

Animal Cloning: A Risk Assessment

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Preface

Preface

The following Risk Assessment is the result of a multi-year effort by staff from the US Food and Drug Administration's (FDA's) Center for Veterinary Medicine (CVM or the Center). Since the late 1990s, CVM has been gathering data and meeting with clone producers and other stakeholders interested in cloning to discuss the safety and regulatory implications of somatic cell nuclear transfer (SCNT), the process most commonly used to generate animal clones during this time period. In the fall of 2000, CVM tasked the National Academy of Sciences (NAS) to perform an independent, scientific review of the available data on the safety of cloning, including holding a public meeting to identify science-based concerns and elicit data and information on clones and their food products from the scientific community. In July of 2001, the Center issued a CVM Update requesting that clone producers not introduce meat or milk from clones or their progeny into food or feed until the NAS report had been completed, and the agency had had a chance to complete its own review of the safety of those food products.¹

In October of 2002, NAS issued its report "*Animal Biotechnology: Science-Based Concerns*." Following an overview of the available data on animal clones, the report indicated that the most likely mechanism for generating hazards to clones would stem from reprogramming of the donor cell genome, and that any harms that might result from that reprogramming would be observed early in a clone's development. They further noted that there were no published data comparing the composition of meat or milk from clones with that from conventionally-bred animals. Nonetheless, the report concluded that there is "no evidence that food products derived from adult somatic cell clones or their progeny pose a hazard (i.e., there is no evidence that they present a food safety concern)" (page 65).

This Risk Assessment is CVM's subsequent independent analysis of all of the available data relevant to assessing the health of clones and their progeny (and other animals involved in the cloning process) or food consumption risks resulting from edible products from these animals. In order to make the Risk Assessment as transparent as possible, all of this information is available to the public, either by virtue of its publication in peer-reviewed journals, or by "publication" in this risk assessment. We have actively sought independent peer-review of these data by providing all of the data in raw form (not summaries) either in the text of the risk assessment or in appendices. In addition, we have also described the means by which the methodology was developed to facilitate peer-review by risk assessors.

¹ http://www.fda.gov/cvm/CVM_Updates/clones.htm

CVM has attempted to be as comprehensive as possible about identifying and using all of the relevant data in its analysis. We have performed extensive literature reviews, engaged in conversations with scientists involved in cloning animals, and requested data on animal health and food composition from scientists, breeders, and food producers. Unpublished data were provided to us in raw, unanalyzed form, which we subsequently analyzed. CVM determined whether a particular publication or dataset was relevant to the analysis. These judgments were framed by the two overarching objectives of the Risk Assessment: determining whether cloning poses any health risks to the animals involved in the cloning process, and whether any hazards arise during the development of clones or their progeny that may pose food consumption risks.

Literature searches for the draft version of the Draft Risk Assessment ceased in early 2006. For the final version of the Risk Assessment, we have conducted updated literature searches (through mid-year 2007), thoroughly reviewed hundreds of additional relevant papers from the peer-reviewed literature, and incorporated this information into the Risk Assessment. The final version also includes additional, unpublished data that were submitted to CVM after the release of the Draft Risk Assessment. We reviewed all of the public comments that we received on the Draft Risk Assessment and associated documents. Careful consideration was given to relevant, science-based information in the comments, and parts of the Risk Assessment have been revised in response to these comments.

In addition to understanding the Risk Assessment's goals, it is equally important to understand what it does not consider. It does not attempt to address the question of whether clones are "normal;" rather it concentrates on identifying the risks that cloning poses to animal health or to humans and animals consuming food derived from clones and their progeny. It also does not attempt to explore issues such as the influence of different donor cell types or cell cycle stages in the "success rate" for producing clones, or the degree to which clones are more or less identical at the phenotypic level. Studies addressing these questions have been used, however, when they provided data useful to the identification of hazards or risks. Similarly, the Risk Assessment does not attempt to parse out the relative effectiveness of different cloning techniques or different laboratories in generating live animals. Results of cloning in species not commonly used for food have been employed only as they have utility as model systems (*e.g.*, mice as models for livestock). Uncertainties associated with those models have been identified.

It is important to note that this Risk Assessment is a scientific document that provides a framework by which science-based questions regarding animal health and food consumption risks are evaluated. CVM recognizes that cloning raises many ethical and

economic concerns. These issues may be important to members of the public, however, they are not within FDA's mission and therefore not within the purview of this Risk Assessment.

Finally, the measures we will take to manage the risks associated with cloning and our recommendations regarding the use of clones or their progeny as food or feed are not included in the Risk Assessment, but are addressed in the accompanying Risk Management Plan and Guidance for Industry.

Chapter I:
Executive Summary

Chapter I: Executive Summary

Cloning is the colloquial term used to describe the process of somatic cell nuclear transfer (SCNT) that falls on a continuum of assisted reproductive technologies (ARTs) currently used in agriculture. In this Risk Assessment, the Center for Veterinary Medicine (CVM or the Center) at the US Food and Drug Administration (FDA) presents a science-based review of the available information on cloning in species traditionally used for food (*i.e.*, cattle, swine, sheep, and goats).

A. Overview

This Risk Assessment addresses SCNT technology, its impact on the health of animals involved in that process, and food consumption hazards that may arise in animal clones and their progeny² in the context of the use of ARTs in conventional animal agriculture. Chapter II is a summary of ARTs currently used in food animal breeding and a detailed explanation of SCNT. Chapter III describes the process of risk assessment, its application to animal cloning, and the nature of the hazards that may arise as the result of cloning. A synopsis of the processes involved in epigenetic reprogramming and their relevance to adverse outcomes noted in animals derived via SCNT and other ARTs is found in Chapter IV. Chapter V addresses potential health risks to animals involved in the process of cloning, including surrogate dams, clones, and their progeny. Chapter VI addresses potential food consumption risks that may result from edible products derived from animal clones or their progeny. Each chapter contains conclusions relevant to that subject; the Risk Assessment is summarized in Chapter VII, and our overall conclusions are presented there. In order to make this process as transparent as possible, all of our methodologies are presented in the text of the risk assessment; the information and data that CVM evaluated are publicly available, either in peer-reviewed publications, or in Appendices to this document. The process by which CVM drew its conclusions is presented in the Risk Assessment, along with explicit statements of potential bias and uncertainty. The document concludes with a complete bibliography, a glossary of terms, and appendices containing data and background information.

The Risk Assessment is the result of a qualitative analysis that identifies and characterizes the nature of hazards that may be introduced into animals as a result of cloning, and puts them in the context of other ARTs currently practiced in the United States. The strongest conclusions that

² For the purposes of this analysis, a clone is defined as an animal produced asexually from a single animal by somatic cell nuclear transfer. Clones are thus genetically identical to their nuclear donor animal. Progeny of clones have at least one animal clone as a parent (but could also result from mating two animal clones) and are produced by sexual reproduction. Clones of clones would be considered as clones (*i.e.*, directly arising from an SCNT process).

can be drawn regarding positive outcomes in risk assessments of this type are “no additional risk” because outcomes are weighed against known comparators. If a finding of “no additional risk” were to be applied to the health of animal clones, it would mean that the cloning process would not pose any greater risk to the health of the animals involved than other ARTs. Applied to the safety of edible products derived from clones, a finding of “no additional risk” would mean that food products derived from animal clones or their progeny would not pose any additional risk relative to corresponding products from conventional animals, or that they are as safe as foods that we eat every day. As with all risk assessments, some uncertainty is inherent either in the approach we have used or in the data themselves. Where uncertainties exist, CVM has attempted to identify the degree of uncertainty and the reasons for its existence.

B. Technology Overview (Chapter II)

Assisted reproductive technologies (ARTs) have been employed extensively in animal agriculture for over a century, and at least one (artificial insemination) has been practiced for several hundred years. These technologies form a continuum that ranges from the fairly minimal assistance provided to animals engaged in natural service through the more recent development of SCNT. ARTs have aided in the genetic improvement of domestic livestock species by the selection and propagation of desirable phenotypes, and accelerating the rate at which those characteristics have been incorporated into national herds. Artificial insemination, for example, permitted the propagation of valuable genomes without the sire being physically present, thereby allowing superior genetics to be spread beyond relatively small geographical areas.

Most commonly used ARTs rely on fertilization as a first step. This joining of egg and sperm is accompanied by the recombination of the genetic material from the sire and dam, and is often referred to as “shuffling the genetic deck.” From a breeder’s perspective, phenotypes resulting from sexual reproduction cannot be predicted—that is, the characteristics of the offspring from a mating may be estimated, but not predicted with certainty. Nuclear transfer, the most advanced of these technologies, does not require fertilization and allows for the propagation of known genotypes and phenotypes without the risk of genetic reshuffling. Thus, SCNT’s greatest immediate impact on animal agriculture may be that it allows the propagation of genomes whose phenotypes are proven. It also allows the propagation of animals whose reproductive function may be impaired, or of very valuable animals that have died. SCNT, like the other newer forms of ARTs (*e.g.*, *in vitro* fertilization, embryo splitting) results in some known adverse outcomes to the animals and possibly the dams bearing those pregnancies.

C. Risk Assessment Methodology (Chapter III)

Risk assessment is a science-based process used to identify hazards that may be present in predefined exposure scenarios, and to estimate the severity and chances of the outcome(s) occurring once that exposure occurs. Because many, if not all, of the individual steps that comprise a risk assessment contain various degrees of uncertainty, risk assessors should explicitly describe the sources of uncertainty and the effect(s) that the uncertainties may have on any judgment of risk. Risk assessment serves as the scientific underpinning from which risk managers may choose different options based on their understanding of, and responsibilities to, the broader contexts within which they operate.

Qualitatively, risk may be thought of as some function of the combination of exposure and the intrinsic properties of the substance or process under consideration by linking an exposure to the likelihood of an outcome. When performing a risk analysis, it is critically important to distinguish between a *hazard* and the potential *risk(s)* that may result from exposure. A *hazard* can be defined as an act or phenomenon that has the potential to produce an adverse outcome, injury, or some sort of loss or detriment. These are sometimes referred to as *harms*, and are often identified under laboratory conditions designed to maximize the opportunity to detect adverse outcomes. Thus, such observational summaries are often referred to as “*hazard identification*” or “*hazard characterization*.” Risk, then, is the conditional probability that estimates the probability of harm given that exposure has occurred. In a qualitative assessment such as this, however, risks can be discussed only within a qualitative context, and no quantitative interpretations should be made.

In order to address the hazards and risks to animals involved in cloning and the food products derived from them four issues must be addressed: identifying hazards and risks; determining the degree to which existing data address the question of risk; characterizing residual uncertainties; and selecting the most appropriate definition of risk for the risk assessment.

This Risk Assessment explicitly excludes transgenic clones from the identification of hazards or risks experienced by “just clones” because of the inability to determine whether the transgenic event or cloning was causally associated with an adverse outcome. In addition, the Risk Assessment has assumed that, at minimum, animal clones, their progeny, and food products derived from them would be subject to the same laws and regulations as conventional animals and their food products.

Source of Hazards/Risks: Because no exogenous genes have been introduced into animals derived via SCNT, the underlying assumption regarding potential hazards that could arise is that anomalies observed in animal clones are due to incomplete or inappropriate reprogramming of

the donor cell nucleus. These anomalies may be macroscopic (*e.g.*, anatomical abnormalities, difference in size or growth rate, reduced fertility, morbidity, mortality) or they may be more subtle in nature. Potential *subtle hazards* would allow an animal clone to develop with apparently normal appearance and functions, but with sub-clinical physiological changes.³ These include alterations in clinical chemistry, hematology, or changes in physiological set-points (*e.g.*, changes in hormone levels). For food consumption risks, relevant subtle hazards that might result from inappropriate or incomplete reprogramming include alterations in the expression of key proteins affecting the nutritional content of food, possibly leading to dietary imbalances. Similar hazards arise in animals generated via other ARTs or natural breeding. The goal of this risk assessment is to determine whether any unique hazards arise that are not noted in comparators, or have not been identified in cattle, swine, sheep, or goats produced via other ARTs or natural breeding.

To address animal health and food consumption risks associated with cloning, two complementary approaches were employed. First, information on the health of animal clones was evaluated within a framework developed by CVM called the *Critical Biological Systems Approach* (CBSA). For food consumption risks, the CBSA was applied in combination with a second approach referred to as *Compositional Analysis*. Following review of all of the available data using the CBSA and Compositional Analysis, a *weight of evidence approach* was then used to draw conclusions regarding risks to animals associated with cloning, and risks to humans from consuming foods produced by animal clones.

The CBSA: This approach divides the life cycle of an animal clone into five functional developmental nodes. Developmental Node 1 incorporates the initial technical steps involved in SCNT (cell fusion) and continues through fetal development. Developmental Node 2 encompasses the perinatal period, including labor induction in the dam, delivery, and the critical few days after birth. The third developmental node, Juvenile Development and Function, covers the period of rapid growth between birth and the onset of puberty. The Reproductive Development and Function Node (Developmental Node 4) includes puberty and reproductive function throughout the reproductive life of clones. The Post-Pubertal Maturation Node (Developmental Node 5) consists of all non-reproductive functions of sexually maturing or mature clones, including growth, weight gain, disease frequency, aging, and, where available, lifespan.

The nature of each component of the risk assessment (*i.e.*, animal health or food consumption risks) shaped the manner in which the available data were evaluated using the CBSA. For example, identification of adverse outcomes for animal health included both the animal clone

³ Such subtle hazards are not typically included in standard food safety assessments.

and the surrogate dam carrying the pregnancy. Emphasis was placed on the clones' development and probability of normal development, compared with other ARTs such as artificial insemination (AI), *in vitro* fertilization (IVF), and blastomere nuclear transfer (BNT). In our assessment of animal health, we considered a wide range of hazards, ranging from macroscopic to biochemical changes (*e.g.*, changes in gene expression, differences in enzyme activity) that might affect the well-being of animal clones. For food consumption risks, animal clones bearing gross anomalies were excluded from the analysis, and emphasis was placed on identifying unique subtle hazards that could have arisen as the result of the SCNT process. The rationale for this approach is found in Chapter IV, and provides the molecular evidence for the role of epigenetic reprogramming as the source of these subtle hazards. Because of the assumption that hazards would be subtle, datasets were evaluated on as fine a level of resolution as possible, including individual animals or even individual analytes per animal in order to have as sensitive a screen as possible for adverse outcomes (and thus potential food consumption risks). In this risk assessment, the most detailed level of resolution used for evaluating animal health has been physiological and biochemical measures of individual animals. It is likely, as technologies mature, that molecular techniques such as genomics, proteomics, and their integrated metabolomic measures will assist in such determinations (NAS 2004).

Compositional Analysis: To reach conclusions about the risks of consuming food produced by animal clones, findings regarding animal health (derived from the CBSA) were considered in conjunction with results of the Compositional Analysis approach. In an attempt to find potential subtle hazards, the data considered in this part of the risk assessment included measurements of gross composition (*e.g.*, carcass composition, percent fat and protein) as well as detailed analyses of vitamins and minerals, fatty acid profiles, and protein characterization of meat and milk produced by clones. The composition of foods produced by clones was compared to the composition of foods produced by comparator animals, and also to published reference ranges for meat and milk. These comparisons formed the basis of our determination of whether meat or milk from clones differs materially from meat or milk from conventional animals, and thus contributed to the overall conclusions regarding food consumption risks.

Weight of evidence: Weight of evidence evaluations do not rely on a single study or even a subset of studies. Instead, they are based on expert judgments on all of the information gathered in the course of a risk assessment. This allows for variability in the amount of information on any particular aspect of the evaluation, as well as inconsistency in endpoints evaluated. Chapters IV, V, and VI contain detailed descriptions of studies that were considered relevant to the hazard identification and characterization, and subsequent risk evaluation. For each adverse outcome identified, the empirical evidence for the causal association of cloning with that outcome was weighed against the empirical evidence indicating that there were associations with other causal agents or processes.

D. The Implications of Epigenetic Reprogramming for Clones and their Progeny (Chapter IV)

Epigenetics has been defined as the study of stable alterations in gene expression potentials that arise during development and cell proliferation. In sexual reproduction, a new diploid genome is created by the fusion of two haploid genomes. The subsequent expression of that genome into a functional organism is governed by a “program.” There are several examples of epigenetic control of gene expression, of which DNA methylation is likely the best characterized.

Mammalian embryos experience major epigenetic reprogramming primarily at two times in their development, both of which have significant implications for cloning. One of these takes place soon after fertilization, and is referred to as preimplantation reprogramming; the other occurs during gametogenesis (the development of cells that ultimately become the sperm and egg). Because preimplantation reprogramming occurs after fertilization, and in the case of nuclear transfer, after fusion of the donor nucleus with the oöplast, it is the most immediately affected by the cloning process, and may be most directly implicated in the development of clones with defects. Gametogenic reprogramming may also be involved in the abnormalities noted in clones, but it likely has more far-reaching implications for progeny, because it generates the gametes used for the sexual reproduction of clones.

The efficiency of producing clones (*i.e.*, the number of live offspring born compared to the number of embryos transferred) by SCNT is very low. The reasons for this low efficiency may be related to inappropriate epigenetic reprogramming. When cloning, the donor nucleus must be coaxed to direct embryonic development as if it were a fertilization-derived zygote. Most of the time, this is not successful. Anomalous epigenetic reprogramming is observed at the global genomic and individual gene level in clone embryos and fetuses, and in similar developmental stages of animals produced using ARTs with significant *in vitro* culturing components. Many of these are lethal, as demonstrated by the low success rate of IVF and the even lower success rate of SCNT. In the small number of successful cases that ultimately result in clones that appear normal and healthy, reprogramming in SCNT-derived embryos appears to be as successful as reprogramming in fertilization-derived embryos. Live and apparently healthy clones may exhibit some level of epigenetic differences relative to fertilization-derived animals, but these differences do not appear to have adverse effects on their well-being or ability to grow and develop normally.

The Center assumes that if clones were to pose food consumption risks, the only mechanism by which those risks could arise would be from inappropriate epigenetic reprogramming, similar to those observed for other ARTs. It is important to note that the genes that are being dysregulated are the “normal,” naturally present genes that comprise the animal’s genome, and have not been

introduced via recombinant DNA techniques from other sources (*i.e.*, clones are not transgenic or genetically engineered animals).

Inappropriate epigenetic reprogramming is not expected in the sexually reproduced progeny of animal clones at levels that exceed those observed in other ARTs or natural reproduction. Unlike their clone parent(s), the progeny of clones are produced by the union of male and female gametes. Production of these gametes *de novo* by the clone parents appears to reset any residual epigenetic reprogramming errors associated with nuclear transfer. Therefore, anomalies present in clones do not appear to be transmitted to the next generation, and the offspring that are produced are normal and healthy. Progeny of clones are thus not anticipated to pose any additional food safety concerns compared with other animals produced via sexual reproduction.

E. Risks to Animals Involved in Cloning (Chapter V)

To identify the potential hazards and assess any resulting risks to animals associated with cloning, Chapter V focuses on the health of clones at all five developmental nodes (pregnancy and parturition, perinatal, juvenile, reproductive, post-pubertal). Health risks to surrogate dams carrying clone fetuses are also considered, and the health outcomes of SCNT are compared with the outcomes of other ARTs. The overall conclusion of Chapter V is that animals involved in the cloning process (*i.e.*, cattle and sheep surrogate dams, and clones) are at increased risk of adverse health outcomes. The increased risks in cattle and sheep clones appear to be limited to the early stages of the life cycle. Although none of the adverse outcomes is unique to cloning, the incidence of these abnormalities observed in animals produced by SCNT is increased compared to animals produced by other ARTs.

Cows and ewes used as surrogate dams for SCNT-derived pregnancies are at increased risk of health problems during pregnancy and parturition. These problems include abnormal placental development and function and complications during late gestation such as hydrops (hydroallantois)⁴ and dystocia (difficult birth) due to excessive fetal size. Overgrowth of the fetus and complications during late pregnancy are collectively referred to as large offspring syndrome (LOS). These conditions also occur with other ARTs that have a significant *in vitro* culturing component, but at a lower frequency. In contrast to cattle and sheep, surrogate swine and goat dams bearing clones do not appear to be at increased risk of complications during pregnancy.

Once clones are born, there are distinct differences between the species with respect to health

⁴The bovine fetus develops in a fluid-filled membrane called the amniotic sac. Surrounding the amniotic sac is a second fluid-filled membrane, the allantoic sac. Wastes from the fetus accumulate in the fluid contained in the allantoic sac. Hydroallantois, also referred to as hydrops, is excessive accumulation of fluid within the allantoic sac during pregnancy.

risks. In swine and goat clones, morbidity and mortality do not appear to be increased during the perinatal period. In calf and lamb clones, however, the incidence of both morbidity and mortality are increased during the perinatal period compared to calves and lambs produced using other ARTs. Clinical signs in perinatal clones associated with LOS include respiratory problems, prolonged recumbency,⁵ enlarged umbilical cord, hyper/hypothermia, contracted flexor tendons, and symptoms associated with abnormal development of the major organs. Survival of these clones appears to be a function of both the severity of the clinical signs and appropriate post-natal management.

Similar to the perinatal period, the risk of morbidity and mortality in clones during the juvenile period varies among species. Compared with animals produced by natural service or ARTs, bovine clones continue to be at an increased risk of morbidity or mortality up to approximately six months of age. These risks appear to be sequellae of the abnormalities first noted in earlier stages of development that persist beyond the perinatal period. In contrast, swine and goat clones do not appear to be at increased risk of morbidity or mortality during the juvenile period. Swine and goat clones, as well as clone calves that are not adversely affected by congenital abnormalities, appear healthy throughout the juvenile period and exhibit normal patterns of growth and development.

As clones approach puberty, no increased risk of adverse health effects have been reported in any of the species evaluated. Clones of both sexes appear to have normal reproductive function, are fertile, and can produce normal offspring via sexual reproduction. Finally, the available information indicates that mature clones are normal and healthy, and there are no increased health risks at this developmental node relative to conventional animals.

Currently, it is not possible to draw any conclusions regarding the longevity of livestock clones or possible long-term health consequences associated with cloning due to the relatively short time that the technology has existed.

Sexually derived progeny of animal clones appear to be normal and healthy. As described in Chapter IV, any residual epigenetic reprogramming errors in clones are expected to be reset during gametogenesis, resulting in production of normal offspring by sexual reproduction. Consistent with these predictions, the data on the health status of clone progeny indicate that there is no increased risk of health problems in these animals compared with conventional animals.

⁵ Respiratory problems and prolonged recumbency appear to be the most common problems associated with perinatal death in clone calves.

F. Food Consumption Risks (Chapter VI)

1. Two-Pronged Approach to Identifying and Characterizing Food Consumption Risks

In order to determine whether epigenetically-caused subtle hazards pose food consumption risks, CVM has developed a two-pronged approach. The first component, the *Critical Biological Systems Approach* (CBSA), incorporates a systematic review of the health of the animal clone or its progeny. Its role in the evaluation of food consumption risks is premised on the hypothesis that a healthy animal is likely to produce safe food products. It accepts that at this time, SCNT is a biologically imprecise and inefficient process, but recognizes that animals are capable of biological repair or adaptation. The cumulative nature of the CBSA allows for the incorporation of both favorable and unfavorable outcomes. The former, provided that all other measures appear to be normal, will result in the finding that the clone is likely to produce edible products that pose no food consumption risks; the latter implies that clones with anomalies are likely to be considered unsuitable for food. The second component, the *Compositional Analysis Method*, assumes that food products from healthy animal clones and their progeny that are not materially different from corresponding products from conventional animals pose no additional risks. It relies on the comparison of individual components of edible products, and the identification of appropriate comparators.

Assessing the safety of food products from animal clones and their progeny⁶ is best accomplished by using both approaches: prospectively drawing on our knowledge of biological systems in development and maturation, and in retrograde, from an analysis of food products. Subtle hazards and potential risks that may be posed by animal clones must, however, be considered in the context of other mutations and epigenetic changes that occur in all food animal populations. No adverse outcomes have been noted in clones that have not also been observed in animals derived via other ARTs or natural mating that enter the food supply unimpeded.

2. Conclusions Regarding Potential Food Consumption Risks

Based on this review of the body of data on the health of animal clones, the composition of meat and milk from those animals and corresponding information on clone progeny, CVM has drawn the following conclusions:

a. Cattle Clones

⁶ Although milk from clones might be marketed for human consumption, CVM anticipates that relatively few animal clones will enter the food supply as meat (*e.g.*, if culled from the herd due to injury or senescence). Relative to clones, it is more likely the progeny of clones will be used to produce meat and milk for human consumption.

Edible products from healthy juvenile bovine clones pose no additional risk(s) relative to corresponding products from contemporary conventional comparators.

The underlying biological assumption for this developmental node is that if anomalies were found in the youngest clones, the juvenile developmental node would be a period of equilibration and normalization as those animals proceeded toward adulthood. Animals experiencing severe developmental abnormalities are not expected to survive. The data are consistent with such a hypothesis.

Juvenile bovine clones that survive the perinatal period are largely healthy and normal. Although some younger clones in this developmental node may be more physiologically unstable than their conventional counterparts, most are able to equilibrate their physiological status and go on to exhibit normal patterns of growth and development. This normalization has been observed consistently in juvenile bovine clones except for those experiencing the sequelae of the developmental abnormalities present at birth. In some cases, these adverse outcomes can persist beyond the perinatal period, resulting in an increased risk to the health of these clones during the first six months of life. Animals bearing these problems are not expected to pass inspection and would not be allowed into the food supply, and therefore are not expected to contribute to food consumption risks. However, no additional subtle hazards that could pose food consumption risks were identified during the juvenile period, as demonstrated by the analysis of clinical chemistry and hematology data, demonstrating that healthy juvenile clones exhibit appropriate physiological responses to developmental signals.

Edible products derived from adult bovine clones pose no additional risk(s) relative to corresponding products from contemporary conventional comparators.

This conclusion is based on application of both prongs (CBSA and Compositional Analysis) of the risk assessment approach. The body of data comprising the CBSA approach is consistent with the biological prediction that there are no underlying biological reasons to suspect that healthy animal clones pose more of a food safety concern than conventional animals of similar age and species.

The data show that healthy adult clones are virtually indistinguishable from their comparators even at the level of clinical chemistry and hematology. These data also confirm the observation that physiological instabilities noted earlier in the lives of the clones are resolved in the juvenile developmental node (see previous conclusions regarding other developmental nodes), and do not reappear as the clones age. There are some reports of early deaths of clones; as these animals would be prohibited from entering the food supply, they do not pose a food consumption risk. Data on reproductive function in cows or bulls of this age cohort indicates that healthy bovine

clones surviving to reproductive maturity function normally and produce healthy offspring. These observations are consistent across studies. Given that reproduction is the most difficult “biological hurdle” placed on an organism, the observation of normal reproductive function provides an additional degree of confidence in the conclusion regarding the appropriate development of these animals.

All of the reports on the compositional analysis of meat or milk from bovine clones show that there are no biologically significant differences in the composition of milk derived from clone and non-clone cattle. Additionally, data from one report show no difference in allergenic potential for meat or milk derived from clone cattle compared to meat or milk from non-clone comparators, and neither meat nor milk from clone or non-clone cattle induced mutations *in vitro*. Finally, none of the reports identified an endpoint that would pose a hazard for human consumption.

b. Swine Clones

Edible products from adult swine clones pose no additional risk(s) relative to corresponding products from contemporary conventional comparators.

This conclusion is based on the same underlying biological assumption as cited for adult bovine clones. Because the data are more heavily weighted towards adult, market sized animals, judgments regarding the safety of food products from swine clones are provided in one aggregate set of comments.

Once piglet clones are born, they appear to be healthy. The most compelling argument for the normal health status of swine clones results from the evaluation of the behavior and physiological status of a small cohort of relatively young (15 weeks), and approximately market age (27 weeks) swine clones relative to closely related conventional pigs. No significant differences were observed in either behavior, epigenetic, or physiological measurements, indicating that these animals were not materially different from the comparators. Another small dataset on swine clones reared in very unusual settings (i.e., deprivation of colostrums, initial husbandry in pathogen-free conditions, switching to commercial settings) is confounded with respect to outcome. Nonetheless, these clones were able to respond appropriately to this stress, and their carcass characteristics, reproductive performance, including semen quality, farrowing rates and litter sizes were within normal reference ranges for conventional swine. No biologically relevant differences were observed in the composition of meat from these clones or their comparators.

c. Sheep Clones

Except by relying on underlying biological assumptions, and by inference from other species, there is insufficient information on the health status of sheep clones to draw conclusions with respect to potential risks that could be posed from the consumption of food products.

With the exception of reports on Dolly, CVM was unable to find any publicly available reports on the health status of live sheep clones. There are several studies addressing methodological issues for optimizing the generation of clones, but these do not address post-natal health. There are reports of anomalies noted in fetal sheep clones that have died or been terminated, and reports on the pathology associated with animals that do not survive. Although these are instructive for understanding the molecular and developmental pathways that may be perturbed during the process of SCNT, these studies have limited relevance to addressing food safety because the deceased animals would not have been allowed to enter the food supply. CVM was not able to find any reports on the composition of milk or meat from sheep clones.

d. Goat Clones

Edible products from goat clones pose no additional food consumption risk(s) relative to corresponding products from contemporary conventional comparators.

This conclusion is based on the same underlying biological assumption cited for the other livestock species, and a relatively small but compelling dataset. Once clone embryos are transferred to surrogate dams and pregnancies are confirmed, the “success rate” for live births is quite high. The animals appear to develop normally through reproductive age, and the available data indicate their physiological responses are appropriate for age and breed. The reproductive development and function of male Nigerian Dwarf goat clones demonstrate that those animals functioned appropriately relative to age- and breed-matched comparators. One male progeny goat was derived from the buck clones; this animal also appeared to function in an age- and breed-appropriate manner. No meat or milk composition data were identified for goat clones.

e. Clone Progeny

Edible products derived from the progeny of clones pose no additional food consumption risk(s) relative to corresponding products from other animals.

Relative to the amounts of meat and milk derived directly from clones in the U.S., it is likely that more edible products (both meat and dairy) will be produced by the progeny of clones. These progeny, unlike their clone parents, are produced by normal sexual reproduction. The underlying biological assumption for health of progeny animals (explained in Chapter IV) is that passage through the process of creating the cells that ultimately become ova and sperm naturally resets epigenetic signals for gene expression, and effectively “clears” the genome of incomplete or

inappropriate signals. This assumption has been supported by empirical⁷ evidence in the mouse model system, which clearly indicates that phenotypic alterations noted in the parent clones are not passed to their sexually-derived progeny. Detailed observations of the progeny of bovine and swine clones demonstrate that these progeny are born healthy, develop normally, and do not exhibit any of the anomalies observed in clones. One extensive dataset on the progeny of swine clones providing direct data on the composition of their meat indicates that these animals are essentially indistinguishable from the comparable progeny of non-clone animals. These empirical data, together with our underlying biological assumption, support the conclusion that edible products from clone progeny pose no additional food consumption risk(s) relative to edible products from any other sexually reproduced animals.

We therefore concur with the high degree of confidence that the outside scientific community (NAS 2002 a,b) places in the underlying biological assumption, and conclude that consumption of edible products from clone progeny would not pose any additional food consumption risk(s) relative to consumption of similar products from sexually-derived animals.

G. Concluding Statements (Chapter VII)

For Animal Health: SCNT results in an increased frequency of health risks to animals involved in the cloning process, but these do not differ qualitatively from those observed in other ARTs or natural breeding. At this time, the overall efficiency of SCNT is low. Cattle and sheep exhibit a set of clinical signs collectively referred to as LOS that do not appear to be present in swine or goats. Surrogate dams are at risk of complications from birth if the fetus suffers from LOS, or from accumulation of fluid in the cavities of the placenta (hydrops). Risks to clones associated with LOS include increased incidence of fetal and neonatal death, and abnormalities that may require additional supportive care during the perinatal period. Clones affected by LOS can recover and mature into normal, healthy animals, but many succumb to complications of LOS during the juvenile period. The risk of morbidity and mortality appears to decrease with age, and after approximately six months of age most bovine clones are normal and healthy as determined by physiological measurements, behavior, and veterinary examinations. Progeny of animal clones also have been reported as normal and healthy.

For Food Consumption Risks: Extensive evaluation of the available data has not identified any subtle hazards that might indicate food consumption risks in healthy clones of cattle, swine, or goats. Thus, edible products from healthy clones that meet existing requirements for meat and milk in commerce pose no increased food consumption risk(s) relative to comparable products

⁷ Empirical refers to that which can be seen or observed alone, often without reliance on theory. In the context of this risk assessment, conclusions drawn on empirical evidence are those that are drawn strictly based on the data. These conclusions may later be put in the context of underlying biological assumptions.

from sexually-derived animals. The uncertainties associated with this judgment are a function of the empirical observations and underlying biological processes contributing to the production of clones. There is less uncertainty about the health of clones as they age and have more time to exhibit the full range of functionality expected of breeding stock.

Edible products derived from the progeny of clones pose no additional food consumption risk(s) relative to corresponding products from other animals based on underlying biological assumptions, evidence from model systems, and consistent empirical observations.

The results of this comprehensive risk assessment agree with the preliminary findings of the NAS (2002a) conclusions that “The products of offspring of clone[s] ... were regarded as posing no food safety concern because they are the result of natural matings,” and “In summary there is no current evidence that food products derived from adult somatic cell clones or their progeny present a food safety concern.”

**Chapter II:
Technology Overview: Somatic Cell Nuclear
Transfer and Other Assisted Reproductive
Technologies**

Chapter II: Technology Overview: Somatic Cell Nuclear Transfer and Other Assisted Reproductive Technologies

Since the beginnings of livestock agriculture, selection criteria have been applied to foster the propagation of animals with traits more desirable to humans. The expansion of herds with desirable traits has been limited, however, by the reproductive capacity of the species or breed and the prevalence of particular versions of genes (or sets of genes) responsible for those traits in the available gene pool. (The gene pool can be considered all of the animals available for breeding.) The female contribution to reproductive success, for example, is limited by species-specific characteristics such as average litter size, frequency of estrus, and gestation length. In natural breeding, male contributions are restricted by the degree of proximity to fertile females and the ability to inseminate females with a sufficient number of normal sperm. Finally, individuals of both sexes are limited to the extent that they may carry the desired versions of genes or combination of genes.

To help overcome some of these complications, various forms of assisted reproductive technologies (ARTs) have been adopted in animal agriculture for over a century, and at least one (artificial insemination) has been used for several hundred years. These technologies form a continuum that ranges from the fairly minimal assistance provided to animals engaged in natural service through those containing components of significant *in vitro* manipulation such as *in vitro* fertilization and embryo splitting, to the more recent development of somatic cell nuclear transfer (SCNT), or what is colloquially referred to as “cloning”⁸ (Faber et al. 2004; Sakai 2005; Oback and Wells 2007b). Beginning with the development and application of modern artificial insemination (AI) methodologies in the first half of the 20th century, ARTs have aided in the genetic improvement of domestic species, including selection of phenotypes such as behavioral and production traits in domesticated animals (Youngquist 1997, Faber and Ferre 2004). By accelerating the rate at which selective breeding goals can be met, improved genotypes have expanded rapidly into national herds in the United States and other countries (Faber et al. 2004; Wells 2005). In turn, this has resulted in lower costs for livestock producers and retail consumers, while simultaneously maintaining or improving the quality and consistency of foods of animal origin.

⁸ The term "clone" originated before the late 1990s. The British biologist J.B.S. Haldane, in a speech entitled "Biological Possibilities for the Human Species of the Next Ten-Thousand Years," used the term in 1963. The Merriam-Webster dictionary, however, dates its use in a biological context to 1903.

Reproductive technology advances have also proven to be powerful tools in curbing the spread of vertically transmitted diseases (i.e., those that are passed from the dam to her offspring during the period immediately before and after birth, either across the placenta or in the dam's milk) (Youngquist 1997). For example, embryo transfer (ET) (see subsequent discussion for a description of this ART) has been used to prevent vertical transmission of *Neospora caninum* in cattle (Baillargeon et al. 2001; Landmann et al. 2002), scrapie in sheep (Wang et al. 2001), Bovine Virus Diarrhea (BVD) in cattle (Smith and Grimmer 2000), and *Brucella abortus* in an American bison (Robison et al. 1998). Embryo transfer is commonly used in laboratory animal research to re-derive valuable strains of gnotobiotic (i.e., animals in which all of the bacterial species are known) or specific pathogen-free research animals when colonies become infected with undesirable disease agents that cannot be controlled through more conservative means.

The following chapter begins with a brief overview of what cloning is, followed by an overview of the continuum of other ARTs commonly in use in current US agricultural practice, placing nuclear transfer technology into context of these breeding practices. Appendix B provides additional details on overall reproductive efficiency observed in current agricultural practice in the US, and Appendix C provides a comprehensive summary of the outcomes observed in ARTs, with particular emphasis on those technologies that contain a significant *in vitro* culturing component. Although all of these technologies are currently in practice, all are continually undergoing development and refinements with the goal of improving efficiencies. A reasonable expectation then, is that success rates (defined as the rate of production of healthy animals) will improve as expertise increases.

A. What is Cloning?

Cloning, or somatic cell nuclear transfer, is a process by which animals are reproduced asexually (embryo splitting and blastomere nuclear transfer are other ways of reproducing animals asexually and are discussed later in this chapter). In cloning, a differentiated somatic cell (a non-germ line cell from an existing animal) is introduced to an oöcyte (a cell that is the immediate precursor of a mature egg) that has had its nucleus (and thus its genome) removed, and then, following some manipulations, is induced to start replicating. If all goes well, the dividing cell is implanted into a female animal (dam), continues to develop normally, and is delivered just as any newborn.

Since the first report of a clone produced by SCNT (Wilmut et al. 1997), several other species have been cloned (Table II-1), although in some cases (e.g., companion animals) only a limited

number of animals have been generated. The reasons for this are multi-fold, but are largely driven by the relative difficulty in producing clones, and the various drivers, economic and technical, that affect the expansion of the technology. For example, the use of clones in expanding elite breeding stock in domestic livestock is perceived to have benefit for breeders and consumers. This risk assessment does not attempt to address those issues, however, and instead concentrates on those domestic livestock clones commonly consumed as food (e.g., cattle, swine, sheep, and goats).

Table II-1: Species of Animals that Have Been Cloned Using SCNT (Resulting in Live Birth)	
Species	First Citation
Sheep	Wilmut et al. (1997)
Mouse	Wakayama et al. (1998)
Gaur	Lanza et al (2000)
Pig	Polejaeva et al. (2000)
Mouflon Sheep	Loi et al. (2001)
Cat	Shin et al. (2002)
Cow	Forsberg et al. (2002)
Goat	Keefer et al. (2002)
Rabbit	Chesne et al. (2002)
Deer	Texas A&M announcement (2003)
Horse	Galli et al. (2003a)
Mule	Woods et al. (2003)
Rat	Zhou et al. (2003)
Wildcat	Gomez et al. (2003)
Dog	Lee et al. (2005)
Banteng	Sansinena et al. (2005)
Ferret	Li Z et al. (2006b)
Swamp Buffalo	Suteevun et al. (2006)
Gray Wolf	Kim et al. (2007)

B. Continuum of Reproductive Technologies

1. Natural Service

Although many people who are not involved in intensive animal agriculture assume that most breeding occurs “naturally”⁹ (e.g., a male animal mates with receptive female), in fact, human intervention is the industry standard for many livestock operations (Youngquist 1997). In the US

⁹ The process of non-assisted mating is referred to as natural “mating,” “coverage” or “service.”

dairy industry, for example, most reproduction involves some technological component, and swine producers rarely use natural mating for their production of offspring. Conversely, in the beef industry most reproduction occurs by natural service, and most of the world's sheep and goat production occurs under free range conditions and depends on natural mating.

Humans have assisted animals in natural mating by monitoring the reproductive status of females, introducing receptive females to the same location (*e.g.*, field, corral, or pen) as the male, and allowing nature to take its course. When this process does not result in sufficient offspring of the desired phenotype, or is otherwise compromised, assisted reproductive technologies can be called into play.

2. Artificial Insemination and Synchronized Estrus

The first ART developed was artificial insemination (AI), which in its simplest form involves the collection of semen from males and its subsequent human-assisted introduction into a physiologically receptive female. It is an important technique for the genetic improvement of animals, as a few select males can produce sufficient sperm to inseminate thousands of females per year, while natural service would provide for the insemination of only a fraction of those animals.

Reports of AI in horses as part of breeding programs have been traced to the Arabian Peninsula in the 14th century (Bearden and Fuquay 2000). AI of a beagle dog was first described by Spallanzani in 1780 (Hafez and Hafez 2000). In 1899, the Russian Czar Nicholas II commissioned I.I. Ivanov to develop an AI program for horses, and by 1933 Ivanov had developed methods for collecting semen and inseminating horses, cows, sheep, and pigs (Foote 2001). In 1931, 19,800 cows were bred by AI in Russia. By 1936, Denmark had established an AI cooperative association, and by 1939, the use of AI had spread to the United States. In 1970, it was estimated that 7,344,420 dairy cows were bred using AI (Webb 2003).

Although there are several methods for collecting semen, most involve training males to ejaculate into an artificial vagina. Semen is then diluted to maximize the number of services that one male can provide. A normal ejaculate from a dairy bull usually contains between 5 and 10 billion sperm; good conception rates generally require about 12-20 million sperm to be introduced. The diluting solution contains factors that help to stabilize and preserve the sperm, as well as antibiotics to inhibit bacterial growth and reduce the danger of spreading any potential disease or contamination. Most collected semen is stored in glass ampoules or plastic straws, and is generally stored either in dry ice and alcohol (-100°F) or liquid nitrogen (-320°F). To date, there appears to be no limit on the amount of time that bovine sperm can remain frozen and regain viability upon appropriate thawing. Since 1997, use of AI in swine breeding has increased

dramatically. A survey of swine producers conducted by the National Pork Board in 2003 indicated that even among small producers (1,000 to 3,000 swine marketed annually) as many as 60 percent of litters were sired by AI in 2003, while for large producers (> 50,000 swine marketed annually) 98 to 100 percent of litters were sired by AI.¹⁰ Rams (male sheep) and bucks (male goats) can also be donors for artificial insemination.

In the US, AI of the female is usually performed either by trained technicians employed by breeding companies or large farms or by the producers themselves. The most common technique employed today for dairy cows involves the use of sterile, disposable catheters that are inserted vaginally and extended through the cervix into the body of the uterus of the recipient cow (whose estrous cycle has been documented). Thawed semen is warmed to the appropriate temperature, and sperm are deposited in the uterine/cervical regions.

The primary advantages of AI to farmers include the ability to use semen from bulls anywhere in the world rather than those that are more geographically proximate, and thus to have desirable genetics available for propagation. It also allows the farmer to use multiple sires in a herd without the attendant costs of maintaining animals that are often difficult to handle and in multiple breeding pastures. AI tends to be less expensive than natural service (a straw of semen generally costs less than transporting a female to the sire and the stud fee) and avoids the potential physical risks to either sire or dam as part of the mating process. The disadvantages of AI include the need to train personnel engaged in the breeding operations on how to detect estrus in females (see subsequent discussion of estrous synchronization), and training or retaining individuals to perform the insemination. Further, care needs to be taken not to rely excessively on a few apparently superior sires so as not to reduce the genetic diversity of the resulting herds.

Sperm collection and AI were further improved by the advent of sperm sexing, or selection of sperm carrying an X (female) or Y (male) chromosome.¹¹ Development of an effective and simple method for producing animals of the desired sex is economically desirable for livestock producers; sperm sexing is currently being used when available and economically feasible (Foote 2001; Faber and Ferre 2004). For example, in the dairy industry, females are desired because males do not produce milk; and excess males often become veal. In the beef industry, however, males are desired because they grow faster. Females can be the desired sex in the swine industry where leaner animals generally receive higher prices; young female pigs (gilts) tend to be leaner than castrated male pigs (barrows) when they arrive at market.

¹⁰ <http://www.pork.org/Producers/EconomicsMarketInfo/Production%20and%20Marketing2003.doc>

¹¹ In normal mammalian sexual reproduction, the female always donates an "X" chromosome, and the male can donate either an "X" or a "Y" chromosome. XX yields a female animal; XY produces a male.

One method that shows the most promise for predetermining the sex of offspring is sexing semen using flow cytometry. This technique is based on the observation that in livestock species, sperm with X chromosomes have about 3 percent more DNA than those with Y chromosomes. Collected semen is diluted, and single sperm are passed through a laser beam that allows for the determination of the amount of DNA in each individual sperm. Based on their relative DNA content, sperm are sorted into “heavier” (female producing) and “lighter” (male producing) fractions. Another method sexes early embryos by removing one or two of the cells from the early embryo, arresting the further growth of the embryo by freezing, and identifying genes found only on the Y chromosome using the polymerase chain reaction (PCR) in the selected cells (Youngquist 1997). Semen sexing is more rapid, less invasive, and more economical, while embryo sexing is impractical at this time, as it is invasive, time intensive, and quite expensive. Further, the potential to damage the embryo by piercing the protective layer around it (*zona pellucida*), removing cells, and freezing the remaining cells in the embryo is quite high.

Estrous synchronization, or the timed induction of estrus (commonly referred to as heat), further permits the efficient use of artificial insemination (Hafez and Hafez 2000). Estrous synchronization was first practiced in the US in the 1960s and allows for the insemination of large groups of animals in a specified window of time when the female is capable of conceiving a pregnancy. The alternative to estrous synchronization is the time- and labor-intensive method of observing females’ behavior to detect estrus, and then arranging insemination at the appropriate time on an animal-by-animal basis. Estrous synchronization is achieved by treating females with hormones (Hafez and Hafez 2000). These synchronization regimens mimic the natural hormonal patterns leading to estrus and ovulation, and allow the producer to control the timing of these patterns. Several FDA-approved hormones are available for estrous synchronization, such as progesterone and prostaglandin F_{2α}.¹² These hormones are commonly used for conventional breeding in cattle throughout the U.S. According to a recent survey of large U.S. commercial dairies, 87 percent of herds use hormonal estrous synchronization methods (Caraviello et al. 2006). The advantage of linking AI to estrous synchronization lays in the ability of contained agricultural practices to operate on a more predictable schedule. For example, cattle breeders can avoid the reduced conception rates that occur during summer’s heat by breeding animals during the cooler spring season. Predictability can benefit farmers by allowing them to allocate resources (*e.g.*, farm labor, veterinary visits) more efficiently, thus lowering production costs.

¹² See http://www.fda.gov/cvm/Green_Book/greenbook.html. During the natural estrous cycle, the ovaries produce progesterone. If pregnancy does not occur, the uterus secretes prostaglandin F_{2α} which causes progesterone levels to decrease and initiates a series of events that culminate in estrus and ovulation. Estrous synchronization methods use exogenous progesterone and/or prostaglandin F_{2α} to mimic the natural secretory pattern of these hormones, and thus control the timing of estrus and ovulation.

3. Embryo Transfer

It is impossible for a fertile female mammal to bear all of her potential offspring. Litter size, gestation time, and post-partum decreases in fertility all limit the potential number of progeny that she can produce. When the female animal reaches the end of her reproductive period, any remaining unfertilized eggs represent potential offspring that have been lost. One solution to this dilemma is to transfer embryos of genetically superior female animals to multiple surrogate dams. This technique, called “embryo transfer,” is particularly useful in species in which a low number of progeny are produced per gestation.¹³ In concept, then, embryo transfer (ET) is analogous to AI in that the total yield of offspring from a genetically superior, in this case, female animal can be increased (Youngquist 1997).

In 1890, rabbit embryos were first transferred from a donor female to surrogate rabbits. The experiment demonstrated that the surrogate’s genetics would not influence the transferred embryo’s genetics or development. In 1951, a successful live bovine ET was accomplished, but non-surgical methods of embryo collection did not succeed until the late 1960s (Hafez and Hafez 2000).

Currently, it is possible to flush large numbers of viable embryos from a superovulated cow with minimal stress to the animal (Hafez and Hafez 2000). Superovulation of the donor animal is generally accomplished by injecting the animal with follicle stimulating hormone or other exogenous gonadotropins before she enters estrus. The hormones induce production of a large quantity of ovarian follicles containing mature, preovulatory oocytes. Insemination is performed at appropriate times relative to ovulation depending on the species and breed. Using standard synchronization methods, the estrous cycle in recipient surrogate mothers is synchronized in parallel with the cycle of the donor to be ready to accept embryos for implantation and gestation. When embryos are about a week old, they may be flushed out of the donor dam’s uterus non-surgically¹⁴ by guiding a catheter through the cervix and into the uterus. Saline solution is injected into the uterus via the catheter and then flushed, along with the embryos, back out of catheter into a collection vessel. Embryos are isolated from the flushing solution, and examined microscopically to determine whether they are of sufficiently quality to implant. If they meet the criteria for further use, embryos can be transferred immediately to a waiting synchronized recipient animal using the same non-surgical technique that was used to collect the embryos. Alternatively, embryos can be frozen for later use or split into halves (see embryo splitting

¹³ Cattle usually produce one calf, with a twinning rate of approximately 1-4 percent depending on breed; sheep and goats generally produce one or two offspring, with an occasional triplet delivery. Swine, on the other hand, usually bear multiple piglets in a litter, and require multiple fetuses to maintain the pregnancy.

¹⁴ Non-surgical techniques for collection of embryos are preferable to minimize risk to the health of the donor. In cattle, the transcervical (non-surgical) method is used routinely to collect and transfer embryos. In sheep, goats, and pigs, embryos are typically collected and transferred either transcervically or by laparoscopy, which involves insertion of two thin, lighted tubes into the abdominal cavity through small incisions (Hafez and Hafez, 2000).

discussion below). Fresh or thawed embryos are inserted into surrogate mothers, where they attach to the lining of the uterus. Following transfer of embryos, no additional hormones or special treatments are required and the pregnancy is allowed to progress normally.

This technique, referred to as MOET (multiple ovulation and embryo transfer), is often used in relatively intensive cattle breeding programs, but is less developed in other livestock species. Similar to fertilizing many females with sperm from one superior male, MOET provides the breeder the ability to expand genetic traits exhibited in superior females. Further, the ability to freeze embryos allows for the preservation of “genetic stock” to be used at a later time. Its prevalence in livestock breeding, however, is much lower than AI, as it is considerably more expensive (Wilmot et al. 2002).

The International Embryo Transfer Society (IETS), a professional society whose membership includes breeders and researchers, estimates that a total of approximately 612,178 *in vivo* derived bovine embryos were transferred worldwide in 2005 (Thibier 2006). This is an increase of approximately 62,000 embryos from the previous year. Most of those transfers occurred in North America (45.2 percent), with the rest taking place in South America (20.5 percent), Asia (18.9 percent), and Europe (~14 percent).

4. *In vitro* Fertilization

The first *in vitro* fertilized (IVF) offspring was a rabbit born in 1959 (Chang 1959a). Since that time, IVF offspring have been born to mice, rats, hamsters, cats, guinea pigs, squirrels, pigs, cows, monkeys, and humans (Bearden and Fuquay 2000). IVF allows for the production of offspring from animals where other ART methods fail due to difficulties with either the female (blocked oviducts, non-responsive ovaries) or male (marginal semen quality and/or quantity), or where disease is present. In cattle, it is also used for the production of embryos from sexed semen because of the low sperm counts resulting from current sexing protocols, and for the further extension of the semen of superior sires due to the relatively low level of sperm required for *in vitro* fertilization. (IVF procedures are also used to assist human couples with limited fertility.)

The overall technique for IVF is similar among species, and involves significant manipulations *in vitro*, or outside the body of animals. In livestock species, oocytes are collected from the ovaries of either living or deceased animals whose genetic potential is desirable (Goodhand et al. 1999). Ovaries can be obtained by transvaginal aspiration from live animals, or from a deceased animal at time of slaughter. Slaughterhouse ovaries are cross-sectioned and the contents of all of the follicles are collected; mature oocytes are collected, evaluated for quality, and used for fertilization. Immature oocytes must be allowed to continue to develop in a maturation medium.

Either fresh or frozen-thawed semen can be used for fertilization. Sperm need to be capacitated *in vitro* in order to penetrate the zona pellucida and fuse with the ovum or to undergo the same maturation process that they would normally undergo in the female reproductive tract. Capacitation involves a series of cellular changes to the sperm including increased motility, calcium uptake and protein binding (binding to proteins produced by the female reproductive tract). *In vitro* capacitation is accomplished by creating a medium designed to simulate the female reproductive tract and allowing the sperm to incubate in it for a period of time. Sperm are then added to ova, incubated in culture medium for approximately 8-22 hours, and the resulting fertilized ova, called zygotes, are washed, examined for appropriate development, and allowed to continue to divide for up to seven days, again in culture. At that time, if embryos appear normal, they may either be frozen for future use or inserted into the uterus of a reproductively competent female.

The IETS reported that the number of *in vitro* embryos transferred in 2005 increased by 11 percent over 2004 numbers to more than 265,000. Almost all of these transfers were performed in South America (particularly in Brazil) and Asia (especially Korea and China).

The numbers of *in vivo* and *in vitro* embryo transfers for other species (sheep and goats) were considerably lower, with 34,458 sheep embryos transferred, mostly in Australia, New Zealand and South Africa, and approximately 5,000 goat embryos transferred, mostly in South Africa and Asia. According to IETS statistics, approximately 66,000 swine embryos were transferred in 2005 (a huge increase over the previous year's number of 16,016), with almost all occurring in Vietnam and Korea (Thibier 2006).

5. Embryo splitting

Genetically identical individuals derived from a sole embryonic source can arise naturally, as in the case of spontaneous monozygotic twinning, or *in vitro* via the manual separation (splitting) of early stage embryos. Embryo splitting may be considered the first true "cloning" procedure involving human intervention, and was first described by Willadsen and Polge in 1981, when monozygotic twin calves were produced.

Embryo splitting, or the mechanical separation of cells,¹⁵ can be used in very early embryos. Briefly, two-cell embryos derived from either *in vitro* fertilization, or embryo rescue following *in*

¹⁵ Common nomenclature for the early stages of development following fertilization include the **zygote**, which includes the fertilized egg contained in the **zona pellucida**, through about the 8 cell stage of development (3 days in the cow, and 3-4 days in the sow). The **morula** refers to the time period (between about 4-7 days in cows, and 4-5 days in sows) following fertilization in which cells continue to divide within the **zona pellucida**, but there is

in vivo fertilization (as described for embryo transfer) are held in place with micropipettes under a microscope. The *zona pellucida* (the clear layer of protein surrounding the oocyte and fertilized ovum) of these embryos is opened, and the two-celled embryo is then split into individual cells with a finely drawn needle or pipette. One of the cells is left in the original *zona pellucida* and the other is either placed into an empty *zona pellucida* or allowed to develop without a *zona pellucida*. These so-called demi-embryos can be cultured *in vitro* for a few days, inspected for appropriate growth and then transferred directly to synchronized recipient dams or frozen for future use. Similar procedures can be used to multiply embryos that have developed beyond the 2-cell stage (Willadsen 1980). Each of the individual cells or blastomeres from a single early embryo is totipotent. That is, each cell retains the ability to generate a fully functional individual identical to the other individual(s) derived from the other cells of the original embryo.

Although commercial applications of embryo splitting have been tracked by breeders' associations, the technology has never gained significant market penetration for several reasons. It is a very expensive and time consuming procedure that has not provided the yield initially anticipated for the technology. For example, actual calf yield from blastomere splits is approximately 105 calves per 100 embryos, while direct transfer of intact embryos yields approximately 60 calves per 100 embryos (Wilmut et al. 2000). Unless embryos are sexed at the time of splitting, however, breeders may end up with half of their animals being of the undesired gender, thus incurring twice the cost for the desired offspring. In addition, even if the resulting calves are of the desired gender, their production potential is not known, making the procedure an expensive gamble.

6. Blastomere Nuclear Transfer

The next evolution of ART evolved from additional manipulations of the blastomere cell, and involved its fusion with an enucleated oocyte. This method expands on the relatively simple early stage embryo cell separation procedure described previously by allowing the use of cells from later stage embryos. In this case, embryos of the eight to sixteen-cell stage, compact morulae, and the inner cell mass from blastocysts can be used as donor nuclei (First and Prather 1991a). Fusion of these later stage blastomere cells, which have lost their totipotency, with enucleated oocytes, reprograms the blastomere nuclei to allow them to develop as zygotes. Blastomeres from bovine embryos up to the 64-cell stage can be fused with enucleated freshly

no discernable migration of cells into any particular region. At about 7-12 days in cattle and 5 days in swine, a group of cells migrates to a portion of the spherical mass, forming an *inner cell mass*, with the remainder forming a ring of cells around a central hollow core (blastocoele). This is referred to as the *blastocyst*. The inner cell mass continues to develop into most of the body mass that will constitute the fetus and the ring of cells around the perimeter, which is referred to as the *trophoblast*, will eventually make up the placenta.

fertilized oocytes and cultured to develop into genetically identical individuals (Keefer et al. 1994). Cell nuclei derived from the inner cell mass of expanded blastocysts transferred into enucleated host cells are also capable of development resulting in offspring (Sims and First 1994).

This technology, which may be considered the true antecedent of somatic cell nuclear transfer, had limited commercial applicability for the same reasons as embryo splitting: high cost, high loss rate, and the inability to predict phenotypic performance or the gender of the resulting offspring.

7. Somatic Cell Nuclear Transfer (SCNT)

In 1962, biologist John Gurdon of Oxford University pioneered the method of the two step “nuclear transfer” process in frogs: the enucleation of a recipient oocyte and the subsequent transfer of a differentiated somatic cell nucleus to that oocyte. Gurdon’s experiments showed that despite the differentiated status of the donor nucleus, reconstituted cells appeared to reprogram, or dedifferentiate, the nucleus and enable it to function much as a naturally produced zygote. These zygotes successfully developed into viable embryos that hatched and grew into tadpoles. Because the tadpoles had all come from the gut cells of the same adult frog, they all had the same genetic material and thus were all clones. However, Gurdon’s nuclear transfer tadpoles clones failed to metamorphose into frogs. When scientists attempted to apply this technology to other species such as mice, cattle, or other mammals, the developmental program could not be reset (Gurdon and Uehlinger 1966; Byrne et al. 2002).

Scientists continued to tackle the problem and in 1986, Randall Prather and colleagues, then working in Neal First’s laboratory at the University of Wisconsin-Madison, cloned a cow from early embryonic cells using nuclear transfer (Prather et al. 1987). Although this was an example of blastomere nuclear transfer, it effectively set the stage for Dolly’s birth a decade later, on July 5, 1996. Dolly the sheep, the first organism ever to be cloned from adult cells, was created by Ian Wilmut and Keith Campbell using a technique similar to that used to create the first sheep from differentiated embryo cells (*i.e.*, a blastomere clone) in 1995 (Wilmut et al. 1997).¹⁶

In July 1998, Ryuzo Yanagimachi, Toni Perry, and Teruhiko Wakayama of the University of Hawaii announced that they had cloned fifty mice from adult cells using the “Honolulu

¹⁶ Blastomere clones differ from those produced by somatic cell nuclear transfer because they arise from cells taken from early embryos. In theory, these cells are less differentiated than somatic cells, and the expectation is that the dedifferentiation process would be less onerous than for cells that have terminally differentiated (*e.g.*, ear skin cells or kidney cells). Blastomere clones are also less attractive for animal agriculture in that their phenotypes have not been proven; that is, no actual animal exists whose characteristics can be evaluated and chosen for propagation. See Section C of this Chapter for further discussion.

technique” (Wakayama et al. 1998). This was particularly significant because mouse embryos begin to divide almost immediately after the ovum is fertilized, and scientists had believed that this would not allow sufficient time for reprogramming to occur. Sheep, on the other hand, because their ova do not divide for several hours after fertilization, were thought to be an “easier” species to clone, as the natural delay between fertilization and division might be replicated in SCNT, possibly giving the oocyte time to reprogram its new nucleus.

SCNT is a relatively new technology described by many as complex, technically demanding and inefficient, that continues to be developed and improved. As such, there is no set “method” that is universally employed, although the basic steps outlined below are common to most SCNT procedures at the time that this overview was written.

a. Donor cell

For species in which the cloning process has been relatively well developed, the first step is identifying the animal to use as a nuclear donor. Animals to be used for breeding purposes are selected because they have been shown to be genetically superior to herd mates for the trait(s) to be propagated. Somatic cells can be collected from the ear (hole punch) or skin (surgical incision or needle aspiration), although many other cell sources have been used. Multiple factors may influence success or failure of the nuclear transfer process. Coordination of the cell-cycle stage of the donor nucleus and the recipient egg cytoplasm appears to be important for successful development of embryos. In general, the selection of a cell type for commercial cloning from an adult animal has evolved to choosing a collection method that is relatively noninvasive and minimizes stress to the live animal donor.

Several characteristics have been identified as contributors to the degree to which any given donor cell or type of cell will likely result in a successful cloning event (Sung et al. 2006; Oback and Wells 2007a). One example is the “replicative state” of the donor cell. In general, cells in culture accumulate nutrients, grow, and when they reach certain conditions, divide. Cells that adhere to a solid substrate, such as the bottom of a tissue culture dish tend to grow until there are so many of them that they begin to touch each other. Once that happens, they generally stop dividing, and go into a “resting state” with respect to replication (referred to as G0). Cells can also be directed into G0 by depleting the nutrients in their growth medium. Some laboratories have concluded that cells in G0 are the most effective donors (Wilmot and Campbell 1998, De Sousa et al. 2002). Conversely, other laboratories have found that actively dividing cells make good donors (Cibelli et al. 1998a, Lanza et al. 2001). Some laboratories find that cells from embryos or fetuses are the best donors (Batchelder 2005), while others are successful at cloning cells from aged or even deceased animals (Hill et al. 2000a, Tian et al. 2001). Another

characteristic that has been shown to influence the degree to which cells make good donors is how “inbred” the donor animal is (Rideout et al. 2000). These researchers have determined that “hybrid vigor” is important for the success rate of animal cloning and the more inbred the donor animal, the less likely it is that cloning will occur successfully. Further, some species appear to be more amenable to cloning than others (*e.g.*, goats compared with cattle, see Chapter V), and some species have not been cloned at all. At this time, the best conclusion that can be drawn with respect to the degree to which a cell (or animal) will serve as a “good” donor is that the technology is not sufficiently mature to predict with certainty which set of conditions will optimize cloning efficiency.

Once a cell has been isolated from culture, depending on the laboratory, either the entire cell or just its nucleus is transferred under the *zona pellucida* of the enucleated oöcyte using a very thin glass micropipette (Solter 2000) to await fusion.

b. Oöcyte

The cell type used as the recipient for the donor cell to be cloned is the mature oöcyte, the version of the ovum that participates in fertilization during sexual reproduction. The oöcyte contains all of the non-nuclear cellular components required for the early development of an embryo. Oöcytes can be obtained from ovaries collected at slaughterhouses or from live animals using aspiration techniques (see previous discussion of *in vitro* fertilization). Because the oöcyte donates only its cytoplasm (the oöplast), it must be enucleated prior to fusion with the donor. The nucleus is generally removed by microaspiration, using a finely honed needle (PIFB 2003¹⁷).

c. Fusion

In order to begin the development process, the membranes separating the oöplast and the donor nucleus (or cell) must be fused. This can be accomplished in two ways: (1) by the administration of a brief electrical pulse, or (2) chemical fusion. Electrical stimulation appears to be the more commonly used technique and involves the application of one to several microbursts of a mild electrical current in the vicinity of the cells. This induces the formation of pores between the somatic donor cell and oöplast which functionally makes the two cells one. This process also stimulates embryonic development,¹⁸ which if successful, results in the development of blastocysts that are transplanted into surrogate mothers (Cervera et al. 2002).

¹⁷ <http://pewagbiotech.org/events/0924/proceedings2.pdf>

¹⁸ This step is also sometimes referred to as activation. During sexual reproduction, physiological activation of the oöcyte is stimulated by fertilization. Since sperm-mediated fertilization is subverted in SCNT, activation must be stimulated artificially *in vitro* by electrical current; addition of chemicals that increase intracellular concentrations of calcium (calcium ionophores) or inhibit protein synthesis; or a combination of electrical and chemical stimuli.

Technical modifications aimed at increasing the success rate of cloning by improving the efficiency of the enucleation and fusion approaches are steadily evolving. For example, Oback et al. (2003) have developed a method that removes the *zona pellucida* from the oocyte, aligns the donor cells with enucleated oöplasts, and uses electrofusion and chemicals to activate the cells to begin dividing. The results of this technique seem to show similar success rates for generating cattle clones as the cloning techniques more commonly used, with the advantage of being faster to perform (in the authors' hands), and requiring less expensive equipment. Peura (2003) has also described a modified technique for preparing fused donor/oöplasts in which sheep oocytes whose *zona pellucidae* had been removed were enucleated after fusion with donor cells, reversing the order in which those steps are usually performed. This technique appears to provide a higher rate of development of the blastocyst stage, implying that some factors present near the oocyte chromosomes may be of assistance. Other factors that may influence the success rate of SCNT are the timing and method of embryo activation relative to fusion. For example, Sung et al. (2007) found that simultaneous electrical fusion and chemical activation of SCNT bovine embryos resulted in a higher rate of development to the blastocyst stage compared to embryos that were activated four hours postfusion. Compared to bovine SCNT embryos that were chemically activated, Schurmann et al. (2006) reported that postimplantation development of SCNT embryos was improved by using a non-chemical, more physiological method of activation (by transferring donor nuclei into enucleated oocytes that had been fertilized *in vitro*).

Over the next few years other technical refinements may be developed, some based on improved technical practice, and others on increased knowledge about basic molecular mechanisms involved in the developmental process. These should increase the success rate of cloning, and decrease the potential for adverse events to occur.

d. Transfer to recipient

Just as the case for other ARTs with an *in vitro* phase, the developing clone is transferred at the blastocyst stage into a surrogate dam in which the estrous cycle has been synchronized using standard methods. After transfer of the embryo clone(s), the pregnancy is allowed to proceed normally; no additional hormones or special treatments are required to establish or maintain pregnancy.

In cloning's earliest days, the surrogate mother was often chosen to be distinctively different from the donor animal with respect to some clearly visible trait. For example, Dolly's donor animal was a Finn Dorset sheep, a breed with white faces. Dolly's surrogate mother, however, was chosen to be a black-faced sheep, so that if a white-faced sheep were born, it would be clear that it was not a genetic relative of the surrogate mother. In addition to choosing a distinctively

different embryo recipient, Dolly's identity was also confirmed by DNA fingerprint analysis of the donor cell line from which she was derived (Wilmut et al. 1997). DNA fingerprint analysis enables definitive confirmation that an animal clone was indeed derived from a specific cell type, and is now the method of choice for confirming genetic parentage of animal clones (First et al. 1994).

C. Critical Biological Events in SCNT

Although SCNT is an inefficient process with a relatively low success rate, the extraordinary nature of the technology and the demands that it places on the biological system being manipulated should not be overlooked. Unlike the fertilized egg or early embryonic cells that may be considered totipotent (capable of becoming *any* cell in an organism) or pluripotent (capable of become many cells in an organism) "generalists," donor cells tend to be specialists. That is, they have differentiated to such a degree that their genomes have been "reconfigured" in ways that are, as yet, not fully understood in order to carry out the particular function for which they have been destined by their particular developmental fate. Kidney cells, therefore, do not transcribe the milk producing instructions of the mammary gland, yet they continue to carry those genes. The question then, is how to "reprogram" the full set of instructions contained in the genome such that "normal" development can occur. The following is a general overview of the events that are thought to occur during the SCNT process. There are several excellent reviews of the overall process or individual components that interested readers can reference for more details, and Chapter IV deals with some of these issues in more detail (Kikyo and Wolffe 2000; Sinclair et al. 2000; Solter 2000; Young and Fairburn 2000; Fulka et al. 2001; Rideout et al. 2001; Novak and Sirard 2002; NAS 2002a, b; Colman 2002; Dean et al. 2003, Santos et al. 2003; Morgan et al. 2005; Vajta and Gjerris, 2006; Meissner and Jaenisch, 2006; Oback and Wells, 2007a).

In principle, SCNT has demonstrated that cell differentiation can be reversed. Genetic reprogramming, the process of altering the gene expression pattern associated with the differentiated cell to one that is appropriate or early embryonic development, is normally carried out at two stages in the development of fertilization-derived embryos: after fertilization, and during the development of gametes (the sperm and ovum). The actual molecular events involved in reprogramming are not fully understood, although they may be categorized into a few overall steps. These include altering the way in which the chromosomes are packaged by changing the chemical nature of the proteins involved, and changing the chemical structure of the DNA in portions of the molecule that are not responsible for base-pairing (NAS 2002 a,b). A more complete description of these processes is found in Chapter IV.

The nucleus of a cell contains a complete copy of all of the genes required for life. This information is encoded in genes. Physically, genes are the linked nucleotides that comprise

DNA, or the “master molecule” of biology. The total genetic material of an organism is referred to as its “genome,” and consists of long strands of DNA packaged in chromosomes, which come in pairs except for those specifying the sex of the resulting organism. The number of pairs of chromosomes differs among species. Cattle, for example, have 30 pairs, pigs have 19, sheep 27, and goats 30. (Humans have 23 pairs of chromosomes.)

Chromosomes can exist in different “conformations” depending on the stage of the cell cycle. When DNA needs to be moved, as in when a cell divides, or when a sperm needs to deliver the male genome, chromosomes are tightly condensed. During the rest of the cell’s life cycle, chromosomes tend to exist in less tightly coiled conformations so the information encoded in the DNA is more accessible for processing. Specific proteins are responsible for holding chromosomes in different conformations. In all cells but sperm, these proteins are called histones; in sperm chromosomes are packaged by proteins referred to as protamines.

When an ovum is fertilized by a sperm, a complex series of molecular events ensues that is referred to as “chromatin remodeling” (chromatin is another term for the protein:DNA complexes that make up chromosomes.) Although the exact steps are not known, the overall process involves stripping away the protamines packaging the paternal DNA, removing histones from the ovum’s DNA, and allowing the newly associated DNA molecules to reform chromatin in a way that allows the fertilized ovum and early embryonic cells to replicate and be “totipotent”(capable of developing into a complete organism). Many proteins are involved in this, only a few of which have been identified, and it is likely that there are chemical markers on the DNA bases that are altered (such as methylation). Chromatin remodeling is likely very different in SCNT. Disassembly of the tightly condensed sperm chromatids and the subsequent removal of protamines do not occur because there is no sperm present. Instead the oöplast must decondense and repackage the chromosomes of the donor somatic nucleus.

In order to perform the functions of life, cells have to convert the information in the DNA to ribonucleic acid (RNA) (a process referred to as transcription), and then to translate that RNA into proteins, which are the molecules that carry out life’s functions. This coordinated set of activities is referred to as gene expression. Alterations in the expression of a given set of genes are often referred to as “epigenetic effects” (or “around gene effects”) because they do not require changes in the base-pairing properties of the DNA that comprise genes. Instead, they reflect changes in the structure of the chromosome around the gene (such as control regions), or on the nucleotides, but outside the portion of the molecule involved in coding. (See Chapter IV for a more complete discussion). A classic example of the manifestations of epigenetic effects is the different fingerprint or freckle patterns observed in human twins. These individuals have exactly the same coding regions, but small changes in the non-coding regions of the DNA result

in different phenotypes. Other examples of epigenetic control of gene expression include the coat color or color patterns of many mammals.

D. Outcomes Observed in ARTs

As this risk assessment is being prepared, biologists are just beginning to understand the highly complex interactions that must occur to choreograph the millions of molecular interactions that signal the expression or silencing of genes in a particular cell or at any point in its life cycle. Although the exact mechanisms by which these effects occur are not fully understood, in all forms of reproduction, ranging from natural mating to SCNT, these processes may go awry in early development. Although most of the animals born following ARTs with significant *in vitro* components appear to be completely “normal,” some of the outcomes are not so successful. In particular, some of the adverse outcomes noted in these “high *in vitro* component ARTs” appear to have common defects in gene expression, particularly in the overgrowth outcomes (Humpherys et al. 2002).

Published studies involving cattle, sheep, and mice demonstrate that embryos produced using *in vitro* systems may differ in morphology and developmental potential compared to embryos produced *in vivo* (Kruip and den Daas 1997; Young et al. 1998; Farin et al. 2004; Farin et al. 2006; see Appendix C for a more detailed discussion and additional references). For example, common abnormalities have been noted in fetuses (Farin and Farin 1995) and calves (Behboodi et al. 1995; Sinclair 1999) associated with the transfer of bovine embryos produced using *in vitro* maturation (IVM), *in vitro* fertilization (IVF), *in vitro* culture (IVC) systems, and SCNT (Hill et al. 2000b). One set of reported adverse outcomes following transfer of embryos from cloning or *in vitro* production systems is often referred to as Large Offspring Syndrome (LOS). These include lowered pregnancy rates, increased rates of abortion, production of oversized calves, musculoskeletal deformities and disproportionalities, as well as hydroallantois (abnormal accumulation of fluid in the placenta) and other abnormalities of placental development.

The phenomenon of “large calves” was first described by Willadsen et al. (1991). The syndrome has also been identified in fetal and newborn lambs and in mice where the embryos were cultured *in vitro* (Eggan et al. 2001). Offspring with LOS tend to exhibit difficulties with placentation (Farin et al. 2004; Bertolini et al. 2004; Lee et al. 2004; Batchelder 2005). In cattle and sheep, the placentae of developing fetuses with LOS are unusually large for their species, and tend to have abnormal development of placentomes (the sites of attachment between fetal and maternally derived tissues of the placenta). LOS fetuses tend to have longer than usual gestation lengths, and often labor in the dams must be induced followed by Caesarian section

deliveries. The newborns tend to be large for their breeds, and often have abnormal or poorly developed lungs, hearts, or other affected internal organs (liver and kidney), which makes it difficult for them to breathe or maintain normal circulation and metabolism. LOS newborns may appear to be edematous (fluid filled), and if they are to survive, often require significant veterinary intervention. Problems have also been noted in muscle and skeletal development of animals with LOS. These animals also often have difficulty regulating body temperature. (For a more detailed discussion of LOS, see Chapter V).

Although the cause of LOS is not known with certainty, it is likely be related to changes in gene expression (*i.e.*, epigenetic changes) that result from the *in vitro* manipulation and culturing of embryos. A review by Young et al. (1998) suggests that *in vitro* culture alone is adequate to perturb the embryo. This hypothesis is supported by data from Sinclair et al. (2000) where *in vivo* matured and fertilized eggs recovered from superovulated sheep donors, cultured *in vitro* for 6 days, showed an 18-36 percent increase in mean birth weight at day 125 of gestation, depending on the culture system used (Sinclair et al. 2000, Young et al. 1998). Table C-1 (Appendix C) provides a more comprehensive summary of adverse outcomes noted in different ARTs.

This is an area of extensive research in the cloning and developmental biology communities. It is likely that advances in the understanding of these mechanisms will lead to significant improvements in the rates of successful outcomes of all ARTs that include a significant *in vitro* component, including cloning.

E. Future of Reproductive Technologies in Modern Agricultural Practice

Modern agricultural practices will likely continue to employ all of the reproductive modalities described in this overview. The factors that may influence which practices are used will likely be a function of the breeder/farmer/ranchers' needs and opportunities. Seidel (2006), in a foreword to a symposium on ARTs, emphasized that current differences in the reproductive management of cattle in different parts of the world are driven by multiple considerations. Some of these have to do with the nature of the differences in the husbandry of beef (mostly pasture based) and dairy (mostly intensively housed in the US, more pasture based in Australia and New Zealand) cattle. Some are economic (ARTs are much more expensive than natural matings), some are practical (the fertility of dairy cattle has declined significantly in the last 20 years, making ARTs more attractive; using ARTs in beef cattle is not practical for ranchers who look for replacement by natural coverage), and some are technological (the ability to choose genetics more precisely versus the developmental problems associated with ARTs with significant *in vitro* components).

Technological issues will be addressed by continued research and development in this field. To that end, several professional and scientific societies (e.g., the International Embryo Transfer Society, various animal science organizations and breeding associations) have been actively involved as clearing-houses for information and interaction.

SCNT has the potential to impact animal breeding in as fundamental a manner as artificial insemination. Given its current high costs (approximately \$20,000 for a live calf) and relatively low success rates (< 10 percent), SCNT will likely be used to improve production characteristics of food producing animals by providing breeding animals, just as any breeding program would select the most elite animals for breeding, and not as production animals. In this way, cloning does not differ from any of the other ARTs that have been described in this chapter. Cloning has the relative advantage of allowing for the propagation of animals with known phenotypes to serve as additional breeding animals. This is critically important in breeding programs, especially when it may take years to “prove” the merit of a sire or dam. Second, it allows the propagation of animals whose reproductive function may be impaired. It has already been used to increase the available genotype of a particular dairy cow with low fertility; her clones appear to be exhibiting normal fertility (PIFB 2003). Third, it allows the propagation of valuable deceased animals from which tissue samples have been appropriately collected or preserved, which may have profound implications for species or breeds nearing extinction. Finally, for the first time, cloning allows for the careful study of the “nature-nurture” interactions that influence breeding programs by allowing a large enough sample of genetically identical animals to be raised in different environments, or with different diets. Such studies have been impossible to perform prior to the advent of SCNT and are likely to yield important information for developing livestock species to live in areas that have, until this time, been marginal for food animal production. This is of particular importance to the developing world, where even slightly increased wealth generally favors the incorporation of animal-based agriculture.

Regardless of the degree to which cloning may be adopted in animal breeding programs, FDA’s role in performing this risk assessment is clear: the agency’s responsibility is to determine whether cloning poses any risk to animals involved in the cloning process, and whether the consumption of food products from clones or their progeny poses any additional risk compared

with food from conventionally produced animals. This Risk Assessment presents the method by which we evaluated data on clones and their progeny, the data themselves, and the agency's conclusions, including discussions of uncertainty.

**Chapter III:
Developing the
Risk Assessment Methodology**

Chapter III: Developing the Risk Assessment Methodology

A. Charge

In July of 2001, the Food and Drug Administration's (FDA or the Agency) Center for Veterinary Medicine (CVM or the Center) issued an open letter (CVM Update 2001) to producers of animal clones to ask them to refrain from putting edible products from those animals into the food supply until the Center evaluated the safety of those foods.¹⁹ This request had already been made to companies engaged in cloning food-producing animals during the previous year. The overall strategy chosen by the Center was to perform a risk assessment in order to determine what hazards might be introduced into animals as the result of the cloning process, to characterize the resulting potential risks, and to develop risk management proposals commensurate with the identified risks.

B. General Discussion of Risk/Safety Analyses

1. Risk and Safety

Risk and safety can be thought of as two sides of the same coin. In general, the answer to the question of "Is it safe?" is addressed scientifically by determining the conditions under which the substance or action in question is not safe, and then limiting exposures to conditions outside those limits. Because knowledge is always incomplete, and not every circumstance can be controlled, there is no such thing as "absolute safety" or "zero risk." Risk assessors attempt to identify conditions under which risks are estimated to be as low as possible, and risk managers use that information in developing policies to protect human or animal health. The methodology used to characterize potential risk is referred to as risk assessment.²⁰ One of the real values in performing a risk assessment is that in addition to arriving at an outcome, the process of arriving at an answer provides a framework by which data can be organized, analyzed, and interpreted. By dividing the risk assessment process into discrete steps, and then reintegrating them into an

¹⁹ http://www.fda.gov/cvm/CVM_Updates/clones.htm

²⁰ Appendix A provides an overview of risk and safety assessments, especially as they have evolved to address issues relevant to cloning, and may be useful background reading for individuals not familiar with the processes.

overall characterization of potential risk, risk assessment allows both the details and the “big picture” to be addressed for complex problems.

Discussion of uncertainty must accompany every risk assessment. Uncertainties may stem from a lack of fundamental understanding of biological processes and/or from data gaps that may be filled with the appropriate empirical studies; they may be exacerbated by intrinsic variability in datasets. Given that the process of risk assessment identifies data gaps and helps direct the acquisition of data that decrease uncertainties, it should not be thought of as a process that is performed just once, but rather a recursive process in the responsible development of research programs, new products, and science-based regulatory strategies.

2. Risk Assessment vs. Risk Management

Risk management can be defined as the set of activities of identifying and evaluating alternative strategies (often regulatory) to deal with the risks characterized in the risk assessment, and then selecting among them based on social, economic, ethical, and political conditions or criteria (NAS 1996a). Risk managers choose among different options based on the risk assessment, which is generally regarded to be relatively value-free compared to the risk management phase,²¹ and their understanding of and responsibilities to the broader social or economic constructs within which they operate. Risk-benefit or risk-risk decisions are risk management, as they involve an active choice between two or more possible courses of action. A risk management plan based on this Risk Assessment is presented in the accompanying document.

C. Risk/Safety Assessment of Cloning

In order to address the hazards and risks to animals involved in cloning and the food products derived from them (and their progeny²²) four issues must be addressed: identifying hazards and risks; determining the degree to which existing data address questions of safety; characterizing residual uncertainties; and selecting the most appropriate risk metric for the Risk Assessment:

²¹ Although risk assessment is based on science and relatively value-free, it generally contains a few policy-based judgments such as the selection of health protective (conservative) defaults when data are incomplete or when choosing among datasets of equal quality. The selection of policy-driven alternatives should be explicitly discussed in the risk characterization, and the implications of such choices should be described in a risk assessment.

²² An animal clone is one arising directly from a somatic cell nuclear transfer event. A progeny animal is one derived from sexual reproduction that has at least one animal clone as a parent (but could result from two animal clones mating).

- (1) **Identifying hazards and risks.** As there are no existing risk paradigms for animal clones and the food products that may be derived from them, this assessment attempts to identify hazards and risks based on the available data and consideration of the biological processes affected by cloning.
- (2) **Determining the degree to which existing data address questions of animal health or food consumption risk.** Many of the peer-reviewed publications on cloning report on the ability to generate live animal clones from various donor cell sources and culture conditions; the frequency of successful outcomes (where success is defined as a surviving dam and a live offspring with no apparent abnormalities); and the nature and frequency of developmental errors. The nature of the published reports, with some exceptions, reflects the institutions producing them: academic laboratories tend to report the development of new technologies and the observation of abnormalities, while corporate entities tend to report successful implementation of the technology, including summaries of the health status of animal clones. These studies are useful in identifying potential hazards to the health of animals involved in the cloning process and characterizing any risks that may stem from those hazards. The number of reports directly addressing food safety is much smaller than the number of reports reviewed for animal health.

Reports from the peer-reviewed literature likely suffer from “publication bias,” an artifact of the criteria used to determine the “attractiveness” of publication in leading peer-reviewed journals. In general, investigators tend to submit to journals, and journals tend to publish, novel findings or hypothesis-testing results rather than surveys of the health of cohorts of animals. With a few notable exceptions, the literature on animal clones tends to consist of reports of studies of the role of various technical manipulations on the success of cloning procedures, descriptions of initial successes of cloning in species that have not yet been cloned, or descriptions of adverse outcomes. Much of the work in which cloning has been refined (and is therefore more likely to be successful) is being performed by the private sector. Given the competitive nature of the breeding and biotechnology industries, as well as the need to maintain business confidential information, much of the important information on more recent cloning outcomes has not been published or made publicly available. In order to keep the current analysis transparent to the public, however, this assessment only cites information that has been published in peer-reviewed journals, or otherwise made available to the agency by companies engaged in cloning, with explicit permission for release to the public.

- (3) **Characterizing residual uncertainties persisting following a review of the existing data.** Due in large part to the novelty of the technology, the concentration of data at the earliest stages of clone development, and limited data directly addressing food safety,

uncertainty will persist in any estimates of risk associated with animal cloning. As with all science-based uncertainties, additional data may increase the confidence with which judgments are made. The decision as to “how much is enough,” however, is a function of the nature of the risk(s) (*i.e.*, its severity), the quality and consistency of the data (*i.e.*, the weight of the evidence), and the tolerance of the risk management policies for uncertainty.

- (4) **Selecting the most appropriate risk metric for this risk assessment.** The most appropriate standard to apply to the potential risk(s) associated with the consumption of foods derived from animal clones and their progeny is whether such food poses any additional risk relative to that derived from sexually-derived animals. For the purposes of this risk assessment, conventional animals are defined as those animals derived by any reproductive means other than SCNT.

D. Transgenic Animal Clones

This risk assessment addresses “just clones,” that is, animals derived via SCNT whose donor genomes have not intentionally been modified by molecular biology techniques. Transgenic clones, on the other hand, are clones whose donor cells contain exogenous heritable DNA inserted by molecular biology techniques. They are considered to occupy a different “risk space” from “just clones” because the transgenic event (the insertion of a heritable DNA sequence) is intrinsically accompanied by a series of potential risks. These include those associated with the DNA construct and those associated with the product of the gene (if there is a gene product). Organisms derived from transgenic cells will have risks specific to the inserted construct, its insertion site, and its subsequent expression. Although it is entirely possible for transgenic clones (or any transgenic organism) to be produced safely and to be a safe source of edible products, the risks associated with each animal must be determined separately on a case-by-case basis, because of the added genetic material.

Nonetheless, much of the literature on animal clones reports on experiences with transgenic clones. In some cases, the transgenic nature of the animals is explicit (*e.g.*, Hill et al. 1999), but in many others, only careful reading or tracing back references cited in the methods section of the papers allows the reader to learn the transgenic status of the clones (*i.e.*, Lanza et al. 2001, Cibelli et al. 2002). The question is whether any information from the transgenic clone reports can inform the identification of hazards and characterization of risks associated with “just clones.”

After a careful review of the key papers addressing transgenic clones, CVM has decided that it is not possible to determine with certainty whether any particular adverse outcome is due to the process of cloning, the transgenic nature of the donor cell, or some combination of the two. Clearly, the insertion of exogenous DNA introduces a set of hazards not present in non-transgenic clones, and by inference, the creation of a different set of risks. If transgenic animals appear to be normal, the logical inference is that neither cloning, nor transgenesis (or the combination of cloning and transgenesis) has perturbed the animals' development. This is the case of transgenesis and cloning posing no significant (or apparent) risk. In either case, this risk assessment puts greatest weight on reports of outcomes from non-transgenic animal clones, and uses studies of transgenic clones for secondary or corroborative purposes. Nonetheless, given the large proportion of the peer-reviewed literature that reports on transgenic clones comprise, these studies have been cited with the preceding caveats. A more complete discussion of this topic is found in Appendix D: Transgenic Clones.

E. Methodology Development

When considering how to develop a risk assessment methodology for animal cloning, it became apparent the need to develop a framework that could be applied to both animal health and food consumption risks. Early in the development of this risk assessment, there were no studies explicitly evaluating the safety of food products from animal clones. Therefore, our initial analyses of both animal health and food safety were based largely on the health status of the animals producing food. As development of the risk assessment progressed, data on the composition of edible products from clones became available, and these data were included in our analysis of risks related to human consumption of foods produced by animal clones.

Interpreting hazard and risk from the same dataset but for different sets of receptors (animal health - the animals involved in producing clones; food safety - the consumers of the food products) requires shaping the manner in which the data are evaluated to suit the ultimate outcome of the assessment. For example, identification of adverse outcomes for animal health requires evaluating data on both surrogate dams carrying pregnancies and resulting clones. For food consumption, however, animal clones that would be condemned at slaughter, as currently practiced with conventional food animals, were excluded from the analysis, and emphasis is placed on the identification of unique hazards to food consumers that could arise as the result of the cloning process. As described in the following section, and Chapter VI, this requires evaluating the dataset at a finer level of resolution than for animal health outcomes.

The net effect of the different ultimate outcomes of the animal health and food consumption risk assessments is that although the datasets considered by both assessments may overlap

considerably, the manner in which they are evaluated differ, and the conclusions generated from the same (or largely overlapping) datasets vary with respect to the amount of risk present.

1. Hazard Characterization

Identifying and characterizing potential hazards is the first step in characterizing the nature of risks due to cloning (see Appendix A). CVM therefore sought to develop a framework in which adverse outcomes associated with cloning could be presented in a systematic manner that would facilitate interspecies comparisons of outcomes.

For food safety purposes, the scientific and regulatory communities have traditionally operated under the principle that domestic animals (*i.e.*, cattle, swine, sheep, and goats) commonly consumed for food have not developed specialized organs producing toxicants to kill prey or avoid predation (*e.g.*, venom producing glands). Further, because the components of animal tissues are necessary for life, and closely resemble the processes in humans, it is highly unlikely that “silent” pathways to produce intrinsic toxicants exist. Thus, “it is convention that animal metabolites are not considered to be natural toxicants” (Watson 1998).

In order to generate a viable clone, the differentiated genome of the donor cell or nucleus must be reprogrammed by the recipient oöplasm. Because no additional genes are being added, and the presumption is that there are no silent pathways to produce intrinsic toxicants, the only method by which hazards may arise in animal clones is from the incomplete or inappropriate reprogramming of the genetic information from the donor somatic nucleus (*i.e.*, epigenetic effects). These phenomena are described in more detail in Chapter IV.

Where, then, would the potential hazards in clones arise? As outlined in Chapter II, during the development of an embryo, a complex series of molecular events are responsible for balancing gene expression from the maternal and paternal genomes, and directing the appropriate expression of genes in the developing embryo and mature mammal. This process is referred to as “reprogramming.” Alterations in gene expression due to those changes are referred to as “epigenetic” variability, and are present normally in conventional animals, including humans.

The most severe errors in reprogramming will result in death, obvious malformations, or metabolic derangements, and are reflected in the low “success rate” of cloning, the perinatal difficulties observed in some newborn clones, and occasional examples of altered metabolic pathways in very young animals (see Chapters V). These are clearly the subject of the animal health risk assessment. Because animals found to have a disease or condition that would render them adulterated (*e.g.*, unfit for consumption, unhealthful, unwholesome) are prohibited from

entering the human food supply, however, the only remaining food consumption hazards arising from gene dysregulation would be those that allow an animal clone to develop with apparently normal functions, but with sub-clinical physiological anomalies.

These *subtle hazards* are outside the conventional range of hazards commonly the subject of food safety analyses, and can be divided into three overall classes:

- (1) Alterations in gene expression that lead to phenotypic variability such as coat color, size, behavior, longevity;
- (2) Disruption of immune function; and
- (3) Alterations in metabolism leading to changes in physiological “set-points” such that the animal has apparently compensated and appears to be normal on gross inspection, but whose physiology may be aberrant.

It is important to note, however, that changes in gene expression in individuals sharing identical genotypes have been observed in conventional animals and in humans. This phenomenon is often referred to as phenotypic variability, and can be seen at the human level in the different fingerprint and freckle patterns that identical (monozygotic) twins possess. Non-clone mice of identical genotypes fed different levels of certain nutrients can have different coat colors, and exhibit significant differences in body weight and lifespan (Cooney et al. 2002).

2. Potential Risks

Risk is defined as the probability of an adverse outcome given that exposure has occurred. This concept is often presented in the format of the “risk equation” that may be expressed as

$$\text{Risk} \propto f_{\text{outcome}}(\text{exposure}, \text{hazard})$$

or, stated more simply, risk is some function of exposure and hazard.

The “risk equation” can be run in the forward or reverse direction. Characterizing risks from a set of hypothetical hazards is a case of running the equation in the forward direction: it allows the estimation of the probability that adverse outcomes might occur once changes that create hazards have occurred. Such approaches are useful when there is some understanding of the underlying biological processes being evaluated. For example, if incomplete genetic reprogramming (a change that may result in a hazard) were to result in animals with altered

calcium transport mechanisms, a possible animal health risk could be bone weakness or malformation, and a possible food consumption risk (a probability of an adverse outcome) could be compromised human nutrition resulting from a diet of milk containing lower than expected calcium levels (the adverse outcome). In the case of the animal health risk, the degree of risk could vary from insignificant, in which no physical symptoms were present, to severe, in which the animal could experience misshapen or fragile bones leading to difficulties in walking. Because animals found to have a disease or condition that would render them adulterated (e.g., unfit for consumption, unhealthful, unwholesome), only the animals without obvious visible anomalies (and therefore less severe calcium transport anomalies) would be sources of edible products. The food consumption risks then could possibly arise from a lower available calcium pool accessible to milk production, and thus a potential nutritional risk to individuals consuming milk from such animals.

Analysis of end products such as milk constituents is an example of running the risk equation in the reverse direction: it captures the potential outcome(s) of the biological changes, and allows for the identification of exposures and hazards responsible for the risk(s). The nutritional hazard identified in the preceding example might be detected more efficiently by a compositional analysis of milk. Compositional analyses, however, are limited by available analytical methods and comparators. As far as CVM is aware, no complex food (e.g., bacon, beef steak, milk, cheese) has been fully characterized with respect to its chemical composition.²³ The organisms that are or make up foods are comprised of hundreds of thousands of chemical substances that can be influenced qualitatively and quantitatively by diet, environmental conditions, and genetics. Attempts to characterize all of the chemical constituents of “milk” or “meat,” then, are neither practicable nor desirable (NAS 2004). Instead, milk and meat analyses have tended to be limited to characterizing proximates (e.g., water content, proteins, fats, carbohydrates, minerals, ash), or, when necessary or desired, to profiles of particular nutrients, anti-nutrients, or individual components of interest (e.g., vitamin content, fatty acid profiles, or protein composition).

3. Proposed Approaches

a. Animal Health Risks

The Center determined that at this point in the development of the technology, risks to animal health are best characterized using a retrospective approach. In other words, CVM approached this issue by recording and cataloguing adverse outcomes in a biological context, rather than by elucidating specific examples of gene dysregulation and searching for their physiological

²³The International Life Science Institute (ILSI) is currently coordinating an effort to generate a database of the known chemical constituents of major food crops (e.g., corn, soy, wheat).

sequellae. The *Critical Biological Systems Approach* (CBSA), described below, provides a framework in which this may be accomplished (Figure III-2). In general, the Center has relied on integrated physiological measurements to survey animal health, although it is likely that genomics, proteomics and metabolomics will see increased use for such purposes in the future. At the time that this risk assessment was prepared, however, these methods had not been sufficiently developed and validated to allow them to be used as survey tools.

b. Food Consumption Risks

Determining the safety of food products from animal clones and their progeny, at least in its earliest stages, is likely best accomplished by using both approaches: prospectively drawing on knowledge of biological systems in development and maturation, and in retrograde, from an analysis of food products. An intrinsic and valuable part of this analysis is cataloging the available information, and identifying data gaps and uncertainties that may in turn suggest research that could serve to decrease the identified uncertainties. The following sections describe the methodology CVM has proposed to accomplish a rigorous, science-based analysis of potential hazards and risks associated with the consumption of food products derived from animal clones and their progeny.

Prior to undertaking such an analysis, however, subtle hazards and potential risks that may be posed by animal clones must be considered in the context of other mutations and epigenetic changes that occur in all food animal populations. Some are considered beneficial, and have been selected for by animal breeders when a desirable phenotype is obtained. For example, not-so-subtle genetic mutations that have occurred at least twice in nature are the development of double-musled beef breeds such as the Belgian Blue and Piedmontese, which arose from different mutations in the myostatin gene (McPherron and Lee 1997). These animals appear to be healthy, although sexual maturity appears to be delayed relative to other breeds, and female fertility appears to be somewhat lower. Nonetheless, these animals are used in selective beef breeding programs in several countries as they have 20-30 percent more muscle mass than cattle with the wild-type myostatin gene, feed efficiency is increased, and the meat is considered to be more tender although lower in fat content. Meat from these animals is presumed to be food, and as such enters the food supply with no additional regulatory scrutiny. Epigenetic changes that occur on a regular basis include variations in pigmentation patterns (*e.g.*, coloration patterns on Holsteins) and are perhaps most easily thought of as those differences observed in identical twins, such as different fingerprints and freckle patterns.

Finally, it is important to remember that any discussion of subtle hazards and potential risks associated with the products of animal clones is not conducted in a regulatory vacuum. All food,

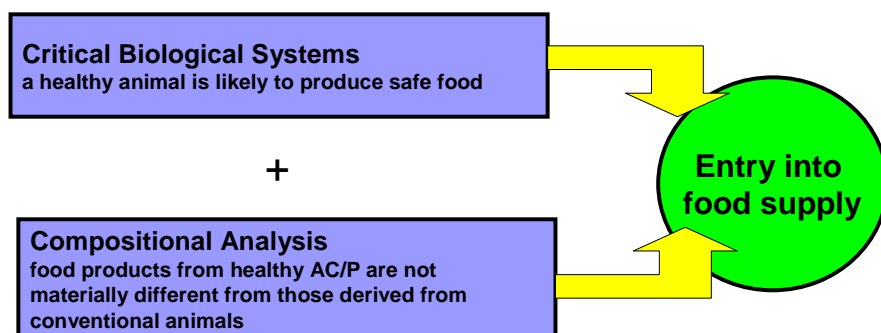
including that from animal clones, must meet existing regulatory requirements in order to be marketed lawfully in the United States.

F. Two-Pronged Approach to Assessing Food Consumption Risks

Given the assumption that food derived from clones will be in compliance with existing regulatory requirements for food products from conventional animals, CVM proposes a two-pronged approach for evaluating the potential risks associated with the food products of animal clones and their progeny (AC/P) (Figure III-1). The first component, the *Critical Biological Systems Approach* (CBSA) is based on the hypothesis that a healthy animal is likely to produce safe food products, and incorporates a systematic review of the health of the animal clone or its progeny. The second component, or the *Compositional Analysis Method*, is based on the operating hypothesis that food products from healthy animal clones and their progeny that are not materially different from corresponding products from conventional animals are as safe to consume as their conventional counterparts. It relies on the comparison of individual components of edible products, and the identification of the appropriate comparators.

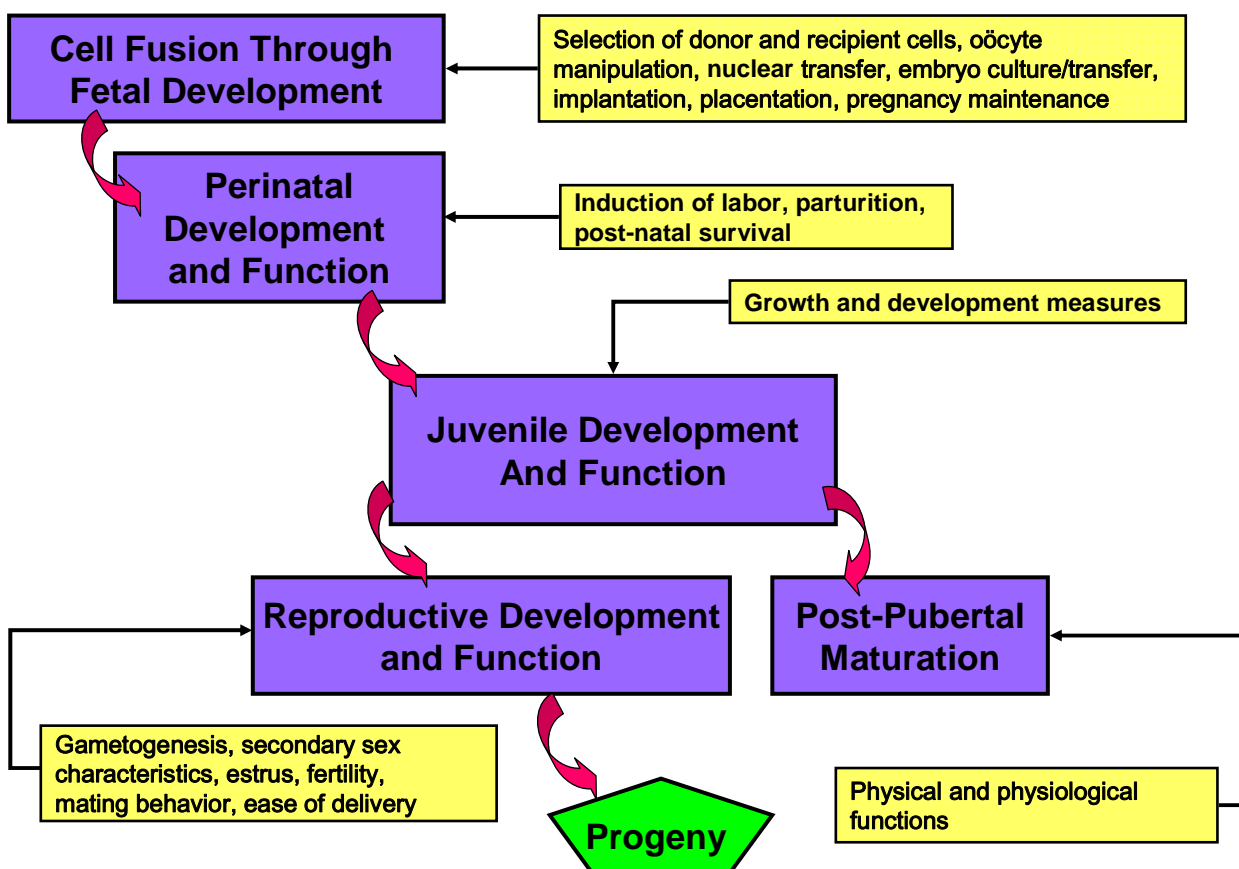
Figure III-1:

Two-Pronged Approach to Evaluating Food Safety



1. Critical Biological Systems Approach

Figure III-2: Critical Biological Systems Approach



a. Overview

The CBSA (Figure III-2) is based on a cumulative evaluation of health status indicators of animal clones. Mechanistically derived, it considers SCNT and the subsequent development of the animal clone from a biological “systems analysis” perspective, and thus may be thought of as being “HACCP²⁴-like.” It accepts that at this time SCNT is a biologically imprecise and

²⁴HACCP is the Hazard Analysis Critical Control Point approach adopted by USDA and FDA for assuring the safety of certain food products undergoing some degree of processing.
http://www.fsis.usda.gov/Science/HACCP_Models/index.asp

inefficient process resulting in few live births relative to the number of implanted embryos, and that some animals are born with obvious defects or subtle anomalies. It also assumes that biological systems are capable of repair or correction, either intrinsically or following human intervention. For example, animals that may have difficulty surviving on their own immediately after birth may develop into healthy, reproducing individuals if provided support in the form of respiratory assistance and warmth during the period immediately after birth. Alternatively, these animals may not recover, and may remain “sickly” or unthrifty until they are culled.

The cumulative nature of the CBSA allows for the incorporation of both favorable and unfavorable outcomes. The former, provided that all other measures appear to be normal, will result in the judgment that the animal will produce food that is safe for consumption; the latter implies that animals with anomalies may be unsuitable for food.

b. Evaluation Nodes

The CBSA selects five key developmental stages of an animal clone’s life, analogous to the “critical control points” of the HACCP analysis. These stages provide biologically-based developmental “collection nodes” (Developmental Nodes) (indicated in Figure III-2 by periwinkle-colored boxes) that also serve as agronomically appropriate points at which to collect data. Examples of the types of data that could be collected are illustrated in Figure III-2 as yellow boxes. It is important to note that these Developmental Nodes address functionality and not necessarily discrete time points, as the latter will vary among species and breeds.

Developmental Node 1 incorporates the initial technical steps involved in SCNT, including cell fusion through implantation, and subsequent embryo and fetal development. Chapter IV covers many of the early common molecular events that occur during this time period common to mammals; Chapter V reviews these issues as they impact on the health of clones and their surrogate dams; and Chapter VI reviews these steps from the perspective of identifying food consumption risks.

Developmental Node 2 encompasses the Perinatal period, including late gestation, labor induction in the dam, delivery, and the critical time period of approximately 0-72 hours after birth. This developmental node allows for the analysis of animal health data relevant to both the surrogate dam and the clone, although few food consumption risks are anticipated to occur at Developmental Node 2 because clones of that age would not be consumed as food.

The third developmental node (Developmental Node 3), Juvenile Development and Function, encompasses the period of rapid growth between birth and the onset of puberty, and may vary in duration among the species considered.

The Reproductive Development and Function Node (Developmental Node 4) encompasses puberty and reproductive function throughout the reproductive period of the animal. Food consumption risks arising from milk production may first be encountered at this point of the animal's life. Because of the complex integration events that must occur for effective reproduction to take place, this developmental node is critically important for evaluating the health and functionality of animal clones. Proper reproductive function indicates that the complex and inter-related physiological systems required for the development and delivery of functional germ cells (and, in the case of females, viable offspring) are functioning appropriately.

The Post-Pubertal Maturation Node (Developmental Node 5) encompasses all non-reproductive functions of sexually maturing or mature animals, including growth, weight gain, disease frequency, aging, and lifespan, where available.

Table III-1: Summary of Developmental Nodes and Implications for Food Consumption Risks		
Developmental Node	Types of Observations/Data	Potential for Entry into Food Supply
1: Cell fusion through implantation, embryo and fetal development	Selection of donor and recipient cells, oocyte maturation and activation, fusion method, days in culture, culture conditions, number of fusions, number of blastocysts formed (if measured). Number of implantations, early and late gestation losses, placentation, pregnancy maintenance, morphological anomalies.	None
2: Perinatal period including immediate pre-partum, delivery, and up to 72 hours post-partum	Number of animals delivered with/without assistance, survival, morphological abnormalities, post-parturition survival, physiological/biochemical characterizations of surviving/dead animals.	Minimal, due to low likelihood of entry into food supply as meat, except for injured animals.
3: Juvenile Development (cattle: pre-weaning; swine, sheep, goats: post-weaning period)	Survival rate, measures of growth, physiological and biochemical markers of health status.	Relatively low, but possibly as meat (e.g., veal, lamb, suckling pig).
4: Reproductive Development and Function	Development of secondary sex characteristics, spermatogenesis, oögenesis, gender appropriate behavior, age of pubertal onset. Fertility measures for males and females. For females, mothering behavior, milk production.	Milk
5: Post-Pubertal Maturation	Growth, weight gain, muscle/fat ratios, milk production.	Meat, Milk

Because the value of clones lies in their use as breeding stock (and is reflected in their relatively high cost), “founder” animal clones are not likely to be slaughtered initially for meat. It is anticipated that most of the food products, especially meat, from clone lineages will enter the food chain as the progeny of animal clones, or their subsequent offspring. Milk from dairy clones could enter the food supply, following breeding and delivery of offspring. Meat from clones could enter the food supply if, for instance, conditions outside the producer’s control forced herd culling (*e.g.*, loss of funding), or when older animals reach the end of their functional utility (*e.g.*, loss of fertility in breeders). Table III-1 summarizes the Developmental Nodes, the types of data likely to be collected at each node, and the potential for the entry of clones into the food supply.

G. The Weight of Evidence Approach

The final step in the Risk Assessment was to consider the information derived from the CBSA and the Compositional Analysis approach as a whole, and then draw conclusions regarding risks, if any, to the health of animals involved in the cloning process and humans consuming food from clones and their progeny. With respect to animal health, individual statements of risk were derived for each species and at each of the five developmental nodes. For food safety, individual statements of risk were derived for meat from each species and for milk from clone cattle.

Weight of evidence evaluations do not require a balanced (or minimum) number of studies from each component of the assessment because they look across all of the studies to determine the degree to which results are consistent, and if not, what potential sources of differences may be. In this Risk Assessment, the weight of evidence approach consisted of four steps:

- (1) *Evaluation of the empirical evidence (i.e., data on molecular mechanisms, physiological measurements, veterinary records, and observations of general health and behavior) for the species being considered;*
- (2) *Consideration of biological assumptions predicated on our growing understanding of the molecular mechanisms involved in mammalian development;*
- (3) *Evaluation of the coherence of the observations with predictions based on biological mechanisms; and*
- (4) *Evaluation of the consistency of observations across all of the species considered, including the mouse model system.*

When drawing conclusions in this risk assessment, empirical evidence was critically evaluated not only within an experimental context (*i.e.*, compared to observations and data from comparator animals and foods), but was also evaluated for *coherence*. The concept of coherence is derived from Hill (1965), and refers to the degree to which the observations are compatible with known biological knowledge and principles.

Consistency, a concept also described by Hill (1965) requires close conformity between the findings in different samples or populations, or in multiple studies conducted by different investigators. In this risk assessment, empirical data derived from clones and foods from clones were evaluated for their consistency across all of the species for which data were available. Like the criterion of coherence, the degree of consistency of the data influenced the strength of the conclusions reached in this Risk Assessment, both for risks to animal health due to cloning and food consumption hazards associated with meat and milk from clones.

The availability of empirical evidence available on each topic was variable and, in many cases, the observations of multiple studies on the same hazard were not consistent. For each hazard, the empirical evidence demonstrating a risk associated with cloning was weighed against the empirical evidence (if any) demonstrating the absence of a risk. This comparison was both quantitative (*i.e.*, based on the number of studies supporting presence vs. absence of risk) and qualitative, putting results into the appropriate physiological context and placing more weight on well-designed studies that provided the most useful information. In some cases, there was insufficient information available to draw meaningful conclusions.

H. Limitations of the Risk Assessment

This is a qualitative, comparative risk assessment that does not attempt to assign a quantitative value to estimates of risk or safety. The strongest conclusions that can be drawn regarding positive outcomes in risk assessments of this type are “no additional risk” because outcomes are weighed against known comparators. If a finding of “no additional risk” were to be applied to the health of animal clones, it would mean that the cloning process would not pose any greater risk to the health of the animals involved than other ARTs. Applied to the safety of edible products derived from clones, a finding of “no additional risk” would mean that food products derived from animal clones or their progeny would not pose any additional risk relative to corresponding products from non-clones, or that they are as safe as foods that we eat every day. As with all risk assessments, some uncertainty is inherent either in the approach we have used or in the data themselves (for a more complete discussion of the uncertainties in this Risk Assessment, see Chapter VII).

Chapter IV:
Epigenetic Reprogramming:
Implications for Clones and their Progeny

Chapter IV: Epigenetic Reprogramming: Implications for Clones and their Progeny

The previous Chapters of this Risk Assessment have introduced the concept that incomplete or inappropriate epigenetic reprogramming appears to be one of the primary underlying causes for the relatively low success rate of cloning, and the source of potential subtle hazards for the consumption of food from animal clones. Although a complete discussion of the rapidly emerging field of epigenetics is beyond the scope of this risk assessment, readers are directed to a series of excellent reviews for more details (Reik and Walter 2001b; Surani 2001; Bird 2002; Li E 2002; Davidson et al. 2003; Kelly and Trasler 2004; Santos and Dean 2004; Tian 2004; Allegrucci et al. 2005; Holliday 2005; Morgan et al. 2005; Eilertsen et al 2007; Reik 2007). An overview of the topic, however, is useful to put the issue of the source of potential subtle hazards in clones into context.

Briefly, epigenetics has been defined as the study of stable alterations in gene expression potentials that arise during development and cell proliferation (Jaenisch and Bird 2003), or alterations in DNA function without alterations in DNA sequence (Jones and Takai 2001). The central idea behind the concept of epigenetics is that although the DNA sequence of almost all the nucleated somatic cells in the body of an adult mammal is identical (except some very specialized cells whose development requires DNA rearrangements), the phenotypes of those cells can be quite different because alternate subsets of genes are expressed at different times in development and during cellular differentiation. In other words, each cell type in an organism has its own epigenetic profile or signature (Morgan et al. 2005).

Epigenetic changes have been implicated as the source of the anomalies noted in clones and other ARTs. The primary biological assumption is that as no exogenous genes are being introduced into the genome being copied and expressed (as in the case of clones) or being expressed following the union of two gametes (as in the case of the other ARTs), alterations in gene expression are responsible for the adverse outcomes noted in the resulting animals. Although much of the focus of the ongoing research in this rapidly expanding field is directed towards gathering and understanding observations of epigenetic changes in early development, epigenetic changes also occur later in life. They are part of the normal and necessary way that organisms adapt to their environments.

For example, Fraga et al. (2005) have demonstrated that monozygotic or “identical” human twins begin life with very similar epigenetic patterns. Over time, however, they accumulate epigenetic differences so that their epigenetic profiles become quite different. Smoking, diet, and other life

experiences are proposed as exerting influence of the epigenetic differences observed between genetically identical twins, with more differences in life experiences correlated with more different epigenetic profiles later in life. Epigenetic changes have also been associated with various disease states that arise from the dysregulation of normal genes (reviewed in Egger et al. 2004, Jiang et al. 2004).

Epigenetic differences are also noted in conventional animals, and may reflect the status of the uterine environment. Cooney et al. (2002) investigated the effects of maternal methyl food supplements²⁵ prior to and during pregnancy on the epigenetic control of various health outcomes using an experimental system based on the expression of an epigenetically-regulated mouse coat color. The genome of mouse strain employed in this study includes an endogenous retrovirus containing viral genes and promoter enhancer sequences referred to as long terminal repeats (LTRs), which can drive the expression of retroviral genes and murine genes in their vicinity. When the LTR is active (relatively demethylated), it overpowers the endogenous mouse promoters, and allows the constant transcription of the genes giving rise to the “yellow” phenotype. This phenotype exhibits a solid yellow coat color, obesity, predisposition to cancer, diabetes, and a relatively short lifespan. When the LTR is suppressed (relatively methylated), the *agouti*²⁶ gene locus is regulated by its own promoters, and is expressed cyclically and only in hair follicles. The phenotype of these mice is lean, healthy animals of normal lifespan and *agouti*-patterned coats. In this study, pregnant dams were fed diets containing three levels of dietary 1-carbon sources or cofactors. The lowest consisted of typical laboratory mouse chow; the intermediate level was supplemented with choline, betaine, folic acid and vitamin B-12, while the highest supplementation included three times the supplement level of the intermediate diet plus methionine and zinc. In addition to evaluating coat color, Cooney et al. (2002) also determined the relative degree of methylation of the LTRs driving the *agouti* gene locus. They observed that as the level of methyl donors in the diet increased, phenotypes of progeny animals shifted towards the *agouti* phenotype. Corresponding changes were observed in the methylation status of the LTR, with increasing methylation in the animals whose dams received higher levels of methyl donors in their diets.

Another example of altered phenotype dependent on methylation status has been identified in inbred sexually-derived mouse strains. In this case, mice that carry a particular allele (specifically *axin-fused* which is referred to as *Axin^{Fu}*) variably express a “kinked tail” phenotype rather than a normal tail morphology. The variability in heritability of the phenotype at this locus

²⁵ Methylation of DNA is performed by specific enzymes (methylases) that obtain methyl sources from either the diet (as folates or folic acid) or from endogenous one-carbon metabolism. The latter requires essential dietary components such as methionine, zinc, and vitamin B-12 to act as cofactors in the synthesis of intermediates that give rise to 1-carbon donors

²⁶ The *agouti* coat color is a continuous spectrum of variegated coat color patterns on a yellow background. In cats, this coat color pattern is referred to as “tortoiseshell.”

has also been shown to be a result of epigenetic inheritance (Rakyan et al 2003). Similar to the variable coat color phenotype described by Cooney et al. (2002, above) the variable expression of the kinked tail phenotype has been shown to correlate with differences in methylation status of a particular type of transposable element at the locus (intracisternal A particle). Additionally, the variability of expression is influenced by the parent that contributed the *Axin^{Fu}* allele. The authors hypothesize that the variability in the phenotype is related to resistance of this type of retrotransposon to epigenetic reprogramming and further influenced by differing levels of reprogramming of the genome contributed by the male or female parent. The two models are not mutually exclusive in that the retrotransposons at the loci are similar and reprogramming may be influenced by maternal methyl donor status and different for maternally and paternally derived alleles (Cropley et al 2006).

Because the field is relatively new, and the scientific community has not identified all of the mechanisms involved in epigenetic remodeling, with few exceptions (*e.g.*, X-chromosome inactivation), the direct links between any one mechanism (or a series of mechanisms) and the health outcomes in live animals are not clear. Animals produced by non-SCNT ARTs, including natural mating, may have different epigenetic profiles, and even exhibit developmental abnormalities, but are not considered to pose unique food consumption risks.

A. Overview of Epigenetic Reprogramming in Early Embryonic Development

In conventional breeding, a new diploid genome is created by the fusion of two haploid genomes; one each from the sperm and the egg. The subsequent expression of that newly formed diploid genome to generate a functional multicellular organism is governed by a “program.” This term was first used by the genetic pioneers Jacob and Monod, who in 1961 proposed that “...*the genome contains not only a series of blueprints, but a coordinated program...and a means of controlling its execution.*” More than a half-century later, researchers are still trying to understand how that control is exerted.

Multiple mechanisms respond to the cell’s developmental stage or its environment by acting as positive (more transcription²⁷) or negative (less transcription) control elements. Transcriptionally active regions of DNA (or heterochromatin) may be considered to be “open” so that various molecules involved in DNA processing can gain access to certain regions, whereas “euchromatin” is physically tightly condensed, or “closed” with respect to access by other molecules, and transcriptionally silent. The picture emerging through current research (see

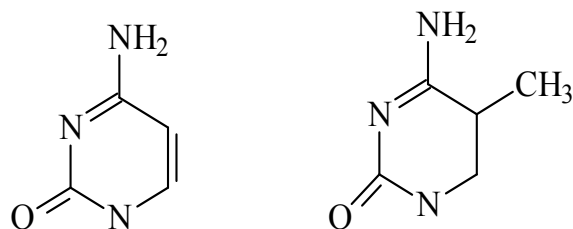
²⁷ Information encoded in DNA is converted into RNA by a process referred to as transcription. Those RNA molecules (messenger RNA) that encode information for protein synthesis are converted to proteins by the process of translation.

citations above) suggests that the overall system is extremely complex, with many degrees of “openness” existing.

One of the examples of this complexity is manifested via the extent and variety of modifications that can occur to DNA itself and its associated histones (positively charged proteins that are responsible for maintaining chromosome structure). These modifications include DNA and histone methylation at a number of positions, acetylation, phosphorylation, and ubiquitination of histones (Kanka 2003; Quivy et al. 2004; Cheung and Lau 2005; Fuks 2005; Verschure et al. 2005). Although histone modification seems to be important for fully appreciating the complete range and stability of regulation possible as well as the subtleties of the system, the methylation state of the DNA is central to the epigenetic regulation of gene expression. DNA methylation has been the subject of considerable research (reviewed by Holliday 2005, Scarano et al. 2005), as scientists begin to understand its role in gene regulation. The bulk of the discussion of epigenetics in this chapter centers on DNA methylation primarily because, to date, most of the studies of epigenetic changes in animal clones examine changes in methylation states.

DNA methylation refers to the addition of methyl groups to the 5 position of cytosine, a non-coding portion of the nucleoside, to regulate the appropriate expression of genes (see Figure IV-1). Methylation tends to occur in areas of the chromosome that are rich in sequences that contain stretches of repeating cytosine-guanosine residues (CpG islands), which tend to be positioned at the 5' ends of genes.²⁸ Most of these regions are unmethylated regardless of developmental stage, tissue type, or gene expression level. DNA methylation in somatic cells is generally faithfully restored at each replication cycle (for dividing cells), although changes in methylation levels are often associated with aging, or occur in abnormal cells (Bird 2002, Jaensich 2004). Methylation may affect gene transcription by physically impeding the access of cellular transcriptional machinery to coding regions, or by attracting proteins that bind specifically to the modified CpG sequences, thus impeding the transcriptional machinery (Cezar 2003).

Figure IV-1: Cytosine and 5-methyl cytosine Addition of a methyl group at the 5 position of the base is shown.



²⁸ The 5' end of a gene is often considered to be at the start of the coding sequence on the DNA molecule. The nomenclature is derived from the position of a hydroxyl group in the deoxyribose sugar ring at the beginning of the strand of the DNA.

Mammalian embryos experience major epigenetic reprogramming primarily at two times in their development, both of which have significant implications for cloning. One of these takes place soon after fertilization, and is referred to as preimplantation reprogramming; the other occurs during gametogenesis (the development of cells that ultimately become the sperm and egg). Because preimplantation reprogramming occurs after fertilization, and in the case of nuclear transfer, after fusion of the donor nucleus with the oöplast, it is the most immediately impacted by the cloning process, and may be most directly implicated in the anomalous development of clones with defects. Gametogenic reprogramming may also be involved in the anomalies noted in clones, but it likely has more far-reaching implications for progeny, because it generates the gametes used for the sexual reproduction of clones, (although, by definition, the absence of gametogenic reprogramming in the somatic cell donors used for SCNT poses a high biological burden for the preimplantation reprogramming (Jaenisch et al 2004)). Most of the literature on epigenetic dysregulation in clones and animals produced using other ARTs addresses preimplantation reprogramming; the literature on gametogenic reprogramming often evaluates endpoints related to the sexual reproduction of clones (Yamazaki et al. 2003).

1. Preimplantation Reprogramming

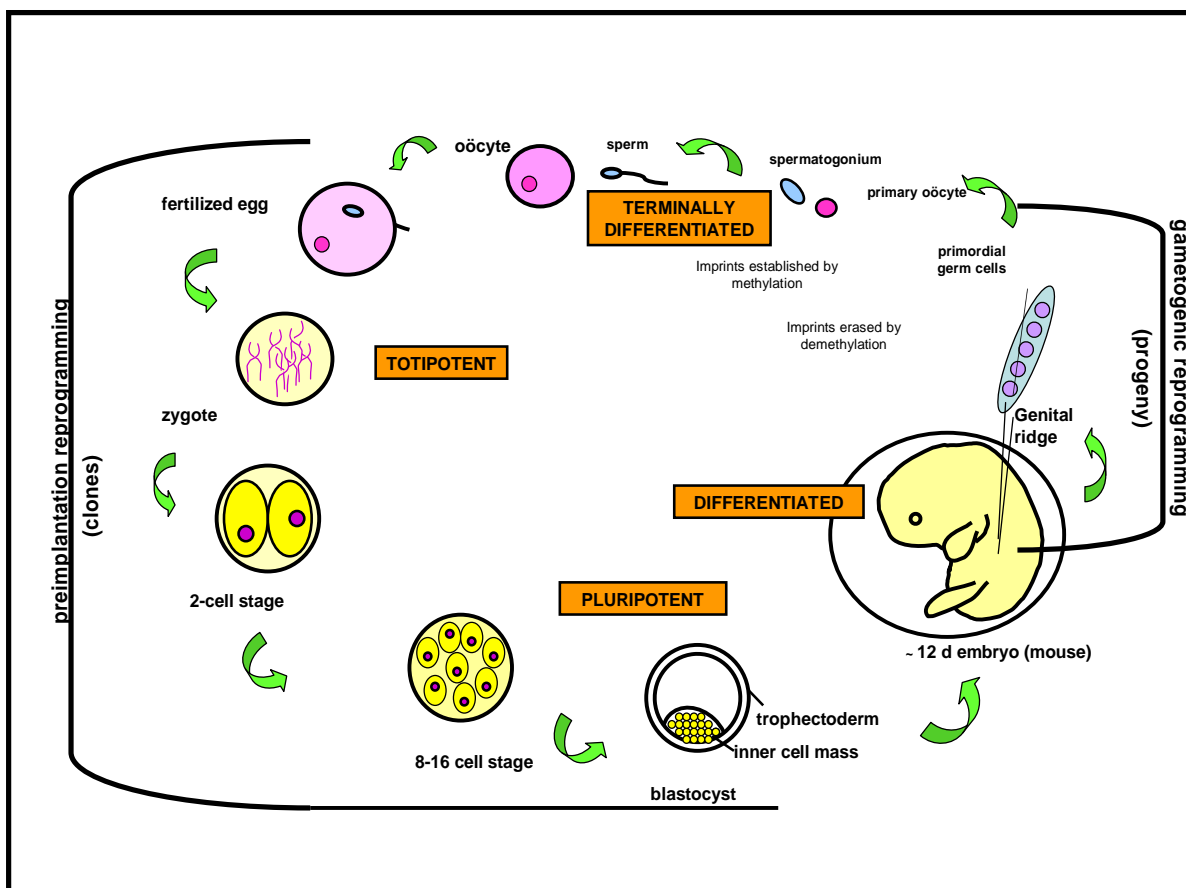
a. Fusion and Cleavage

In sexual reproduction, mammals use cells of highly different morphology and function to deliver haploid genomes. Sperm are small relative to the oöcyte, and package their highly condensed DNA by tightly coiling the DNA around a set of proteins called protamines. The oöcyte's genome is packaged more loosely around a different set of proteins called histones, also found in somatic cells (Cezar 2003). In order for the embryo to form a unique genome, the two chromatin structures must be resolved into one that is capable of coordinated gene expression. A number of factors present in the oöplasm of the ovum, only a few of which have been identified, are thought to aid in this remodeling (Kang et al. 2003a; Chen et al. 2006; Fulka and Fulka, 2007). In the first hour after fertilization, the sperm head swells, the nuclear envelope of the sperm breaks down, and protamines are replaced with histones (Santos and Dean 2004). The chromatin then decondenses, and the male pronucleus²⁹ forms (Mann and Bartolomei 2002). The female genome completes its second meiotic division, expels the resulting polar body, and then forms the maternal pronucleus. Both the male and female pronuclei begin to replicate DNA, and depending on the species, some transcription may ensue. In mice, transcription occurs in the male pronucleus in the first cell cycle, followed by a larger burst in the second cell cycle (Aoki et al. 1997), while in bovine embryos, transcription is delayed (Mann and Bartolomei 2002).

²⁹ The pronucleus is the structure that contains the haploid genome of the sperm or ovum after fertilization occurs, but before they fuse to make the nucleus of the zygote, or the single-celled diploid organism. Once the zygote has undergone the first division (or cleavage), it is referred to as an embryo.

During SCNT, however, different initial events must take place. SCNT begins with the removal of the nucleus of the oocyte that contains the meiotic metaphase II chromosome-spindle complex, followed by microinjecting or fusion of the donor cell or nucleus into the enucleated oöplast. The presence of oocyte factors probably causes the breakdown of the nuclear envelope of the donor cell (similar to swelling and breakdown of the sperm head). Following oocyte activation (usually by electrical stimulation), the chromatin from the donor nucleus decondenses, and a pseudopronucleus is formed. If successful, DNA replication and cellular division follow.

Figure IV-2: Epigenetic Reprogramming and Embryonic Development



Legend to Figure IV-2. The top of the diagram illustrates the terminally differentiated sperm and egg (oocyte). These gametes fuse to form the fertilized egg or zygote and begin preimplantation reprogramming (emphasized by the bracket at the left). Following the steps counter-clockwise around the figure sequential cell divisions are illustrated with corresponding differentiation from totipotent through pluripotent to differentiated. The right half of the figure represents gametogenic reprogramming (emphasized by the bracket at the right), the epigenetic marking of the primordial germ cells that will become the sperm or eggs of this new individual at sexual maturity and setting up another cycle.

Information from either terminally differentiated gametes (fertilization-derived zygotes) or a terminally differentiated somatic cell (in SCNT) must be reprogrammed so that the resulting zygote is totipotent (capable of developing into any cell type). Totipotency appears to be lost early in development and almost certainly after the blastocyst is formed, when the trophectoderm and inner cell mass begin to separate (see Figure IV-2). At this point in development, the cells are pluripotent—no longer capable of being *any* cell type, but retaining the ability to become *many* cell types. The end process is referred to as “terminal differentiation,” in which cells acquire a set of characteristics that allows them to perform a specific function (*e.g.*, muscle cells contract, neurons transmit electrical pulses, and gametes serve as genome donors for subsequent generations). One of the ways that this overall process is accomplished is by resetting the epigenetic marks of the gametes. At this time, the signals that determine “lineage allocation” are not clear. Fujimori et al. (2003) have noted that each of the two cells in the early blastomere is completely totipotent; some lineage bias is observed when the developmental potency of four-cell stage blastomeres is evaluated. Cells inside the eight and 16 cell stage of the morula appear to be more likely to become committed to the inner cell mass lineage (which becomes the embryo), while those outside appear to be directed to the trophectoderm and the development of placental tissues (Morgan et al. 2005).

The next sections provide an overview of events as they are understood in the development of fertilization-derived embryos, followed by examples of observations noted in clones and, when available, other ARTs with significant *in vitro* culturing components. The examples are intended to be illustrative and not comprehensive, as an encyclopedic review is beyond the scope of this discussion. The important points to be made are that

- Mechanisms of epigenetic reprogramming are complex and not fully understood, even in “normal” fertilization-derived embryos;
- SCNT-derived embryos often do not develop normally, and all the available evidence indicates that this is due to incomplete or inappropriate epigenetic reprogramming;
- Genomes are “plastic” and can accommodate some errors in epigenetic reprogramming, regardless of whether those embryos are derived via fertilization or nuclear transfer; and that
- Some SCNT-derived embryos go onto full gestation and delivery; clones that are born can range from those exhibiting some epigenetic dysregulation to no detectable differences.

Some have suggested (Wilmut 2002b, Jaenisch 2004) that no clone is completely “normal” with respect to its epigenetic profile. Although this is an important point for assessing the overall safety of the cloning process for any particular species, and for determining risk to animals involved in the cloning process, the relevance of “epigenetic normality” to food consumption

risks is unclear. This is particularly true when considering the degree to which epigenetic changes are observed in other ARTs with a significant *in vitro* culturing component, and the accumulation of epigenetic changes expected during the aging process. The most compelling conclusions that can be made about food consumption risks are drawn from assessments of the health status of the animals and the composition of food products derived from them, and not from gene expression studies.

b. Demethylation and Remethylation in Early Embryos

Dean et al. (2001) and Morgan et al. (2005) have outlined how the process of demethylation and epigenetic resetting occur in various mammals. Hours after fertilization, but prior to DNA replication and cleavage, the paternal genome of mice, rats, pigs, cattle, and humans, but not sheep, is actively stripped of the epigenetic methylation markers by mechanisms not fully understood, but that likely require the activity of a demethylase enzyme present in the oocyte (Morgan et al. 2005). This genome-wide methylation erasure appears to be conserved among cattle, swine, and rats, but is not observed in sheep (Wilmut et al. 2002; Beaujean et al. 2004; Young and Beaujean 2004). In mice and cattle, the maternal genome retains its methylation markers during this period, and does not undergo demethylation until the zygote undergoes the first cleavage to yield the two-celled embryo. Demethylation of the maternal genome is thought to be passive, that is, diluted by the lack of remethylation on newly replicated DNA (Cezar 2003). In two to eight cell bovine embryos, Dean et al. (2001) observed a further reduction in methylation, consistent with the passive demethylation occurring during DNA replication seen in the mouse. In contrast, mouse 16-cell embryos continued to remain demethylated, and genome-wide *de novo* methylation did not occur until approximately four cell divisions later, and appeared to occur preferentially in the inner cell mass (ICM). Thus, although the overall process of demethylation and *de novo* methylation appears to be conserved in the species evaluated, the timing of these phenomena may differ among species (Morgan et al. 2005). The more important observation, however, is that the first differentiation event in mammalian embryos (the differentiation of the trophectoderm and ICM and the resulting loss of totipotency of the ICM cells) is accompanied by genome-wide *de novo* methylation.

Fertilization-derived bovine embryos begin to demonstrate global genomic *de novo* methylation in the eight- to 16-cell stage, what is often referred to as the maternal to embryonic transition (MET). During this time, the developmental program that is initially directed by components within the egg (maternal) is replaced by a new program directed by the expression of new genes (Wrenzycki et al. 2005), and is accompanied by different rates of demethylation of maternally and paternally derived genes to give rise to a new methylation pattern for the embryo.

Although the early demethylation described above is global (occurring over the entire genome in general), methylation marks on imprinted single copy genes tend to be protected from demethylation so that parental imprints are preserved in the resulting somatic cells of the developing mammal (Li E 2002). It is unknown whether the extensive global demethylation of the genome during pre-implantation development is essential for normal development.

DNA-methylation patterns unique to the developing mammal are established in the embryo after its implantation in the uterus through lineage-specific *de novo* methylation that begins in the inner cell mass. DNA methylation increases rapidly in the primitive ectoderm, which gives rise to the entire embryo. Conversely, methylation is either inhibited or not maintained in the trophoblast and the primitive endoderm, from which the placenta and yolk-sac membranes develop, respectively (Li E 2002). The net effect is that extra-embryonic tissues appear to have a lower methylation state than embryonic tissues. These global differences in methylation status between the embryonic and extra-embryonic tissues appear to be conserved in mice, cattle, sheep, and rabbits (Morgan et al. 2005).

Reprogramming the donor nucleus in SCNT or the nucleus of the early fertilized embryo has been the subject of considerable investigation over the past few years. Much of this research has been summarized in reviews by Rideout et al. 2001; Jaenisch et al. 2002; Mann and Bartolomei 2002; Cezar 2003; Han et al. 2003; Jouneau and Renard 2003; Smith and Murphy 2004; Young and Beaujean 2004; Wrenzycki et al. 2005; Armstrong et al. 2006; and Eilertsen et al. 2007.

These reviews and the studies contained in them have come to approximately the same conclusions: although some clones may develop into healthy animals, the low success rate of SCNT is likely associated with the inability of clones to reprogram the somatic nucleus of the donor to the state of a fertilized zygote. Similarly, the rates of successful embryo formation resulting in term gestation and live births in ARTs that have a high degree of *in vitro* culturing are likely also due to difficulties in reprogramming (Gardner and Lane 2005, Wrenzycki et al. 2005). The sources of the stresses on the embryos that cause these difficulties may be a reflection of the intrinsic biological differences between fertilization- and nuclear transfer-derived embryos (e.g., the need to fully reprogram a differentiated nuclear donor), or technological (e.g., the *in vitro* environment in which early embryos are cultured prior to introduction into the uterus). The following discussion briefly outlines the current state of knowledge of how this is accomplished in fertilization- or nuclear transfer-derived embryos.

In embryos derived via nuclear transfer, epigenetic modification, such as the waves of demethylation and *de novo* methylation observed following fertilization must also occur, but may be hampered by both the nature of the donor DNA and the partially depleted oöplasm. There are reports of both aberrant and “normal” demethylation and remethylation in clones and

fertilization-derived embryos. Differences may be reflections of different methodologies, source cells, species differences, or may reflect unexplained phenomena. The following discussion summarizes the key observations that contribute to the body of knowledge regarding epigenetic remodeling in SCNT- and other ART-derived embryos.

Some species-specific responses in the degree of methylation reprogramming have been observed, although in general, the overall processes appear to be relatively conserved among the clones of different species. Dean et al. (2001) found that somatic nuclei of mouse, rat, pig, and bovine embryos undergo the genome-wide reprogramming described previously, but that reprogramming occurred aberrantly in many cloned preimplantation embryos. Bourc'his et al. (2001), using a similar method, did not observe active demethylation in bovine SCNT zygotes, although they did observe that the somatic pattern of methylation from donor nuclei was preserved through the four cell stage.

Ohgane et al. (2001) compared the methylation status of CpG islands (CG-rich sequences located at promoter regions) in placenta and skin cells of sexually reproduced mice to similar regions in normal-appearing mouse clones. Most of the methylated regions in fetal clones (99.5 percent in the placenta and 99.8 percent in the skin) were identical to those of the controls, but different methylation patterns were observed in the two different tissues. The sites of discordant methylation were located in regions responsible for expression of tissue-specific genes, despite the absence of grossly observable abnormalities. In bovine preimplantation embryos, however, Kang et al. (2001a) noted that bovine clone embryos failed to demethylate satellite regions of the genome (certain repetitive sequences), and instead maintained methylation levels similar to the donor cell. In a subsequent study, however, Kang et al. (2001b) were able to “rescue” the inefficient demethylation of bovine embryos by providing an additional “dose” of oocyte factors to the early embryo. This work confirms the presence of an active element in the oocyte for erasure of paternal epigenetic methylation, and implies that this component, which may be removed or diluted during the process of preparing an enucleated oöplast, is involved in the appropriate epigenetic modeling observed in zygotes and early embryos derived from fertilization. In a third study, Kang et al. (2001c) investigated demethylation in swine clone embryos relative to those derived by *in vitro* fertilization (IVF). They observed that, unlike mice and cattle, the sequences investigated (centromeric satellite DNA) were negligibly methylated in swine oocytes, and hypermethylated in swine sperm. (Sperm satellite DNA sequences in cattle and mouse tend to be undermethylated.) The satellite sequences of the donor pig fibroblast cells were hypermethylated, and retained that status until the 4-8 cell stage. Demethylation began at that time, and the methylation status of the clone embryos decreased significantly in the blastocyst, just as it did in the blastocysts of *in vitro* or *in vivo* fertilization-derived embryos. Their finding thus indicated that satellite sequences of SCNT-derived pig embryos undergo preimplantation demethylation in a manner similar to fertilization-derived embryos. Analogous

results were observed when another sequence, PRE-1 (from the euchromatin) was evaluated. These results are comparable to the pattern observed in mouse embryos by Dean et al. 2001.

Whether the results obtained from these two DNA sequences studied by Kang et al. can be extrapolated to global DNA methylation or other single-gene sequences in the pig remains unknown. Additionally, the reasons for the interspecies differences between mice and pigs on one hand, and cattle on the other, also remain unknown. Nonetheless, one of the key implications of these observations is that global demethylation soon after fertilization appears to be a prerequisite for successful reprogramming later in embryonic development, and possibly for successful SCNT outcomes.

Kang et al. (2003) have also demonstrated that at least some SCNT-derived bovine embryos are capable of normal remethylation during early embryogenesis. They evaluated the methylation status of a 170 base pair fragment of single copy gene in IVF and SNCT-derived bovine embryos. This sequence is negligibly methylated in both sperm and oocyte DNA, and moderately (approximately 37 percent) methylated in the fibroblasts that served as nuclear donors for SCNT. In single celled zygotes, as well as the 4-to 8-cell stage embryos derived via IVF, the low methylation levels of the sperm and oocyte genomes were observed. No significant changes in methylation status of the IVF-derived embryos were observed at the 8-16 cell stage, but by the time a blastocyst had formed, *de novo* methylation appears to have taken place. In SCNT-derived embryos, the methylation pattern of the donor cell was nearly completely lost by the 4-8 cell stage, and demethylation appeared to be complete by the 4-8 cell stage. At the blastocyst stage, the methylation stage of the SCNT-derived embryo was exactly that of the IVF blastocyst, with the same CpG sites exclusively methylated in both sets of embryos. The authors claim that this study is “the most elaborate example of recapitulation of normal embryonic process[es] occurring in [SC]NT embryos.” Although this study clearly demonstrates the ability of somatic cells to be epigenetically reprogrammed in an accurate manner relative to an IVF comparator, and that these molecular results are consistent with observation of apparently healthy and normal animal clones being generated from somatic cell donors, the predictive value of this particular gene for other single copy genes, or the entire genome has not been demonstrated.

In another study showing differences in methylation states among species, Beaujean et al. (2004) evaluated the global methylation status of fertilized and SCNT-derived sheep embryos. They observed that unlike mice and cattle, sheep oocytes do not appear to demethylate the sperm-derived pronucleus after fertilization. *In vivo*-derived sheep embryos demonstrated that a partial demethylation of the global genome occurred up to the 8-cell stage, with similar qualitative findings in SCNT-derived embryos (fibroblast cell nuclear donor), but to a lesser extent. Interestingly, between the 8-cell and blastocyst stages, both *in vivo*- and SCNT-derived embryos

showed comparable overall levels of methylation, but the distribution of methylation patterns differed among the SCNT-derived embryos and between some of the SCNT-derived embryos and those derived from fertilization. The authors attributed these differences to differences in the overall high-order chromatin structure, rather than simply to changes in methylation. They suggested that many SCNT-derived embryos do not undergo the rapid reorganization of the DNA prior to first cleavage that successful *in vivo*-derived (and a small proportion of SCNT-derived) zygotes do. Further, they suggest that perturbations in methylation (and possibly remodeling) correlate with the lack of appropriate trophoctodermal development and subsequent placental development in later embryos and that these alterations may contribute to the high observed levels of placental defects and embryonic loss during SCNT-pregnancies. Beaujean et al. conclude that although “DNA methylation appears to be marker of reprogramming in all mammalian species examined to date, it is not yet clear to what extent it is a determinant.”

In a recent attempt to more directly address the role of methylation status in reprogramming, Blelloch and colleagues (2007), used cells in a mouse model system containing an allele (variant) of a DNA methyltransferase (Dnmt1) that has reduced levels of activity. As a consequence of the mutation the cells have reduced DNA methylation. They found that fibroblasts containing the mutation could be reprogrammed into embryonic stem cell lines three times more efficiently than comparators whose DNA was relatively more methylated, consistent with the hypothesis that decreased global DNA methylation yields a less differentiated state.

c. Epigenetic Reprogramming in Later Development

This summary covers studies of epigenetic reprogramming from the fetal through adult developmental nodes (as described in more detail in Chapter VI).

In a study of genome-wide epigenetic reprogramming in bovine clone embryos and adults, Cezar et al. (2003) measured the amount of 5-methyl cytosine in DNA from various tissues in fetuses and term pregnancies generated via SCNT and fertilization. Their results showed that the amount of methylation was lower in spontaneously aborted fetal clones, fetal clones sacrificed as part of the study, and tissues collected from pregnancies that had experienced hydroallantois relative to controls. These results are in contrast to others that have found hypermethylation in clones relative to fertilization-derived controls (Bourc’his et al. 2001; Dean et al. 2001; Kang et al. 2001b). Adult clones, however, had similar levels of DNA methylation as adults derived via fertilization. Cezar et al. (2003) concluded that there may be an epigenetic reprogramming threshold that is met by a subset of animal clones. They also proposed that clones surviving into adulthood have the ability to overcome epigenetic challenges determined by their somatic cell origin. These hypotheses are consistent with the observations by Chavatte-Palmer et al. (2002),

Chapters V and VI, and the Cyagra dataset, described in Appendix E, in which early physiological instabilities appear to resolve as the clones mature.

Similar to Cezar's observations, Chen et al (2005) also noted that aberrant methylation likely plays a role in the poor development noted in clones and other forms of assisted reproduction. In their study, the methylation status of aborted bovine clone fetuses, aborted fetuses generated by artificial insemination (AI), and adult cattle generated via AI or cloning was studied. Three genomic regions were evaluated: a repeated sequence and the promoter regions of two single copy genes (interleukin 3 and cytokeratin). All of the aborted fetuses (AI- and SCNT-derived) were females between 60 and 90 days of gestation; adult animals were all classified as "healthy" and between 18 and 24 months of age. The adult animals all had approximately the same level of methylation at all of the loci examined, regardless of method of production. The aborted AI-derived fetuses all had similar, but lower levels of methylation than the healthy adults, as well as different methylation patterns. For the single copy genes, methylation could be classified into two groups: one group had very low methylation patterns in the promoter regions, while the other group had methylation patterns similar to the aborted AI-derived fetuses. One of these fetuses also showed low methylation patterns in the satellite region. Although this study is not conclusive, it does provide evidence that at least for certain regions of the genome, appropriate methylation appear to be correlated with normal development.

Dindot et al. (2004) developed a unique bovine hybrid interspecies model (*Bos gaurus* x *Bos taurus*) to study epigenetic markings and imprinting in gestation day 40 female SCNT-derived fetuses and placentae (derived from cumulus cell donor cells) that were genetically identical to fetuses derived by fertilization. Previously, Hill et al. (2000b) had shown that more than 80 percent of bovine clone pregnancies were lost between days 30-60 of gestation, and attributed the losses to placental anomalies including a reduction in the number of expected cotyledons and a decrease in chorio-allantoic blood vessels. These observations were similar to those of Stice et al. (1996) who reported that no placentomes had developed in NT fetuses that died between gestation days 33-55. Mouse clone pregnancies have also shown increases in placental size (Tanaka et al. 2001). These abnormalities have been hypothesized to arise from anomalies in nuclear reprogramming of the trophectoderm, which gives rise to placental structures including the chorion. By using the hybrid *Bos gaurus/Bos taurus* model, Dindot et al. were able to discriminate between parental alleles by following single nucleotide polymorphisms (SNPs) (changes in the nucleotide sequence of the DNA at only one site that allow for the cleavage or the lack thereof by enzymes that recognize specific DNA sequences). In particular, three genes associated with epigenetic reprogramming were selected including *IGF-2*, Gene trap locus 2 (*GTL2*), and the X chromosome inactivation specific transcript (*Xist*). Clone fetuses and placental tissues were isolated from surrogate dams at gestation day 40; none of the clone placentae developed cotyledons, unlike the fertilization-derived fetuses, which had 4, 16, and 25

cotyledons per pregnancy. Although appropriate allelic expression of *IGF2* and *GTL2* relative to fertilization-derived fetuses was observed in both chorionic and fetal liver tissues of the clones, disruptions of genomic imprinting of the *Xist* locus was found in the chorion, but not the liver tissues of any of the clones. Further analysis of two other regions of the genome in the chorion of the clone fetuses, the satellite I repeat element and epidermal cytokeratin promoter, indicated that the trophectoderm-derived tissues of the clones had higher levels of methylation relative to fertilization-derived controls. No differences in methylation levels were observed in the livers of clones or fertilization-derived embryos. In this study, at least, there were differences in the degree of epigenetic reprogramming between ICM-derived tissues (the fetus proper) and those derived from the trophectoderm, consistent with the observation by Hill et al. (2000b), that clones with aberrant placental structures can survive gestation and be born alive and apparently healthy.

To understand epigenetic reprogramming over the lifetime of mice and to identify differences between the methylation status of animal clones and sexually derived comparators, Senda and colleagues (2007) have extended their previous studies (Ohgane et al. 2001) using a system that allows them to simultaneously examine ~2,000 methylation sites. In this study, they compared the methylation patterns in kidney tissues of newborn, adult (8-11 months old), and aged (23-27 months old) mouse clones with age matched comparators. The study also includes up to third generation sequential clones (animal clones that were generated with SCNT of nuclei from animals that are themselves clones).

This study had three main findings. In the first, the authors determine that the methylation patterns for newborn mouse clones differed from age matched comparators in only three of ~2,000 methylation sites, indicating that these clones did not differ markedly from comparator mice derived from IVF. The second finding indicated that the differences in the methylation pattern between clones and sexually reproduced comparators disappeared by 23-27 months of age. Clones at the intermediate time point measured (11 months) differed from their sexually reproduced counterparts in only one point of the methylation pattern. Finally, sequential cloning (that is, using a cell from a clone as a nuclear donor for a subsequent round of cloning), did not appear to either increase or perpetuate differences in methylation patterns in this study.

Although the extent to which these data from this mouse model system is a compelling indication that the epigenetic dysregulation assumed to be responsible for adverse outcomes in clones may indeed resolve with age. These results may also be useful for observation that although animal clones are at greater risk for health problems at birth, those that reach adulthood are often healthy (See Chapters V and VI).

d. Studies of Gene Expression and Development in Clones and Animals Produced by Other ARTs

The previous sections summarized studies of alterations in methylation associated with cloning; the following section summarizes reports of gene expression and phenotypic observations in similar clone populations. The overview is intended to be more illustrative than comprehensive as the literature on this subject is large and growing rapidly. The studies indicate that for non-viable clone embryos, fetuses, or neonates, key genes are inappropriately expressed. In some cases, viable clones have differences in expression compared to fertilization-derived counterparts, leading investigators to speculate that genomes are plastic and that a certain level of gene dysregulation can be tolerated. In other studies of healthy, live clones, no significant differences can be observed between the expression profiles of animals generated via SCNT or other fertilization-based ARTs. Finally, it should be noted that studies comparing embryos generated via various ARTs (including SCNT) with significant *in vitro* culturing components, appear to be sensitive to the culture environment, with developmental success often being a function of the culture medium used.

Most of the earliest studies of gene expression in clones were performed in mice. Boiani et al. (2002) and Bortvin et al. (2003) evaluated patterns of gene expression in mouse blastocysts derived from SCNT to identify which critical genes were involved in the inability of most of those blastocysts to develop further. In particular, they evaluated the expression of *Oct4* and *Oct4*-related genes in these embryos. (*Oct4* is a transcription factor specifically expressed in stem and primordial germ cells, and appears to be required for maintaining pluripotency and the self-renewal ability of stem cells.) Boiani et al. (2002) compared *Oct4* expression in blastocysts cloned from somatic cell nuclei and germ cell nuclei to that observed in synchronous blastocysts produced by IVF and intracytoplasmic sperm injection (as the control groups independent of cloning but involving micromanipulation). Their results demonstrated that mouse blastocysts derived from clones had abnormal *Oct4* expression, and that the failure of mouse clones embryos to develop beyond the blastocyst stage was related to incorrect lineage determination by the inappropriate expression of *Oct4*. Bortvin et al. (2003) identified 10 candidate genes with expression patterns similar to *Oct4* and compared their expression in preimplantation embryos derived from fertilization to embryos whose SCNT donors were somatic cumulus and pluripotent embryonic stem cells. They demonstrated that successful reactivation of the full set of 10 genes correlated with the development of embryo clones, but also noted that almost 40 percent of the cumulus cell-derived blastocysts failed to reactivate these genes faithfully, even though the blastocysts were morphologically normal. Thus, some other factors were required to maintain the pluripotency of the inner cell mass cells. Marikawa et al. (2005) found that the DNA methylation status of the *Oct4* regulatory element in mouse embryos directly influences the level of gene expression. They further noted that the methylation status of the *Oct4* regulatory element was

highly heterogeneous among alleles in a population of adult somatic cells, and hypothesized that the degree to which *Oct4* can be reactivated in SCNT may be a function of the methylation status of the donor cell(s).

Boiani et al. (2005) further evaluated *Oct4* expression in early post-activation SCNT-derived zygotes, fertilization-derived early embryos and parthenotes in six different culture media. (Lack of expression of *Oct4* precludes further development beyond the blastocyst). Among their first observations was that similar to fertilization-derived embryos, progression to blastocyst did not ensure further development of the embryos, and that some of the primary influences on whether such development occurred could be environmental in origin. They also noted that nuclear transfer embryos appeared to be more sensitive to environmental conditions than the other two types of embryos. They concluded that not only was the ability of mouse clone embryos to progress through development contingent on the nature of the donor nucleus and recipient oöplasm, but that culture conditions could have a significant impact on the expression of key genes required for reprogramming (and subsequent development), and the ability of the blastocyst to continue to develop successfully.

To study the correlation between gene expression, survival, and fetal overgrowth (*e.g.*, LOS-type symptoms), Humpherys et al. (2001) examined imprinted gene expression in mice cloned by nuclear transfer and in the embryonic stem cell donor population from which they were derived. They determined that transcript levels of selected imprinted genes varied widely in placentae from animal clones relative to non-clones, although alterations in the expression of one imprinted gene did not correlate with abnormal expression of other imprinted genes. They also observed that changes in DNA methylation levels at one imprinted locus did not necessarily predict changes at other loci. Certain genes (*e.g.*, *H19* and *Igf2*) were largely silenced in the heart and kidney, and their expression reduced in the livers of animal clones relative to conventional animals. No correlations were observed between changes in gene expression and birth weights, placental weights, or neonatal mortality. Culturing the embryonic stem cells *in vitro* resulted in highly variable levels of gene expression; gene expression in the animals resulting from those cells was even more variable than in the cells in culture, implying that culturing early embryos may contribute to the degree of embryonic gene dysregulation. Furthermore, mice derived from the cells of the same cellular lineage differed in their expression of imprinted genes. Given that viable animals were generated with variable expression of imprinted genes, the authors concluded that “mammalian development may be rather tolerant to epigenetic abnormalities and that lethality may only result from the cumulative effects of a stochastic loss of normal gene regulation at multiple loci...even apparently healthy animal clones can have gene expression abnormalities that are not severe enough to impede development to birth but that may cause subtle physiological abnormalities which could be difficult to detect.” The degree to which such subtle abnormalities could exist within conventional populations of animals is not discussed.

Humpherys et al. (2002) evaluated expression of more than 10,000 genes in the placenta and liver tissues of mouse clones derived from embryonic stem cells and cumulus cells using microarray analyses. The expression of 286 genes was altered in clones derived from cumulus cells compared to the fertilization controls, with a similar level of altered expression detected in the embryonic stem cell-derived clones. The general concordance in the expression differences between the mouse clones from different donor cell types suggested to the investigators that most of the expression abnormalities were common to all placentae of mouse clones rather than specific to animals derived from one particular cell type. Consistent with their previous summary (2001), the authors concluded that differences in gene expression, even those that are highly variable, may be tolerated during differentiation and even in clones that survive.

Several groups have recently extended this type of microarray technology to analysis of bovine embryos derived via nuclear transfer and other ARTs (Smith SL et al. 2005; Herath et al. 2006; Beyan et al, 2007a). Although there are procedural differences among the studies, the results are complementary and consistent. They include the observations that the gene expression patterns in cell lines from which embryos were derived were strikingly different from the patterns observed in the resulting embryos. In addition, gene expression patterns in the embryos were relatively similar overall regardless of the cell type and methods used for derivation of the embryos. Finally, a small fraction of the overall number of messages examined was differentially expressed in embryos derived using different methodologies. Similar to the studies using mouse embryos discussed above, these studies show that significant reprogramming also occurs in bovine embryos. These studies are also consistent in that the reprogramming is currently imperfect and there are a number of messages that are differentially expressed in SCNT derived embryos compared with embryos derived using other ARTs. Additionally, numerous studies have shown that many SCNT embryos will not reach term. The embryos used in these studies were early in gestation, so we do not know if the expression differences reflect significant abnormalities or tolerable variation. However, these studies are useful because they contribute to a better understanding of both development and technical approaches to improving SCNT technology.

Sebastiano et al. (2005) noted that in single cells derived from early preimplantation embryos of mice developed via SCNT and *in vitro* fertilization, a series of genes important to appropriate embryonic development began transcription at approximately the same time in both types of embryos. Different levels of expression, however, were found in the nuclear transfer-derived embryos, particularly as the embryos progressed through development. They concluded that reprogramming was initially quickly shifted towards embryonic development, but that reprogramming was incomplete and inaccurate, particularly in the latest stages of preimplantation.

Several studies have attempted to determine whether the expression of any particular gene(s) could be used as a marker to determine the developmental success of embryos produced via SCNT or other ARTs. Camargo et al. (2005) evaluated differences in gene expression in individual preimplantation bovine embryos produced via SCNT (same donor cell line), *in vitro* fertilization (IVF), or *in vivo* derived embryos obtained following superovulation, artificial insemination, and harvested, and cultured *in vitro* to reach the same degree of development as the nuclear transfer or IVF embryos. Using real time PCR, they studied a panel of 11 genes (including *Oct4*) preferentially activated at the maternal-embryo transition (~ the 8-12 cell stage in bovine embryos), during which demethylation of parental genes (or donor cell genes) largely has been accomplished and *de novo* methylation, in which transcription of embryonic genes becomes predominant. Also evaluated was the expression of a fibroblast gene expressed in the donor cells to determine whether cessation of expression of donor genes was also appropriate. The results indicated that the expression patterns of the 11 genes common to the IVF and SCNT-derived embryos were virtually indistinguishable. Further, the expression of the donor cell gene was appropriately turned off in the SCNT-derived embryos. Compared to expression levels in the *in vivo* derived embryos, however, all transcripts except one, lactate dehydrogenase, in both the IVF and SCNT-derived embryos were found at lower levels. They attributed the differences in expression between the *in vivo*- and *in vitro*-produced embryos to differences in culture conditions. To support this hypothesis, the investigators noted that the IVF and SCNT embryos exhibited similar variability in expression among individual embryos, but different from their *in vivo* counterparts.

Miyazaki et al. (2005) compared the expression of a different set of genes from SCNT- and intracytoplasmic sperm injection (ICSI)-derived 2-4 cell and blastocyst stage porcine embryos. The genes selected have previously been suggested as candidates as markers for identifying embryos that would successfully develop (Daniels et al. 2000) included two genes from the fibroblast growth factor family, *Xist* (important in X-chromosome inactivation), genes encoding interleukin-6 and its receptor, and *c-kit* ligand (another gene important in early embryonic development). Donor cells for the SCNT-derived embryos came from two different cell lines, with different degrees of success at developing blastocysts. Additionally, SCNT-derived embryos were developed using two different activation protocols. Although the percentage of embryos in which expression of these genes was similar between the SCNT- and ICSI-derived embryos, actual levels of transcripts of two of the genes (*FGFr72IIIb*, one of the fibroblast growth factor genes, and interleukin 6 receptor gene) were lower and higher, respectively, in SCNT-derived versus ICSI-derived embryos in one of the SCNT-activation protocols, while *FGFr72IIIb* and *Xist* transcripts were lower than ICSI-derived embryos when evaluating the other method of SCNT activation. No significant differences in gene expression were noted at these early developmental stages between the two different SCNT donor cell sources. No

comparisons were made to *in vivo* derived embryos. It is not clear whether the differences between the results observed by Miyazaki et al. and Camargo et al. are due to experimental design, species, or the genes assayed.

Both appropriate and inappropriate gene expression have been observed later in the development of fetuses, neonates, or more mature clones. Yang L et al. (2005) used real-time PCR³⁰ to compare the expression levels of three imprinted genes associated with growth regulation (*Igf2r* and *Igf2*) or imprinting regulation (*H19*) in eight tissues from deceased newborn calf clones, three tissue sources from apparently healthy, genetically identical adult bovine clones, and cattle obtained from a slaughterhouse. The deceased clones all exhibited signs of LOS, and exhibited abnormal and highly variable expression of the genes, despite being produced from one nuclear donor. The decreased levels of expression of *Igf2* (which inhibit fetal growth) in the deceased clones compared to controls were consistent with the decreased expression of the same gene noted by Young et al. (2001), in LOS sheep clones, but interestingly, these levels were not correlated with increased birth weights of the deceased clones. Expression of the three genes in the healthy clones was largely normal, except for *Igf2* in the muscle tissue of adult clones, which was found to be highly variable, although lower than the reported levels for the newborn controls. These results are consistent with the idea that significant dysregulation of imprinted genes results in embryonic or neonatal death, but that those animal clones surviving to adulthood can be epigenetically similar to control animals.

A similar study, this time using real-time PCR to study the expression of IGF binding proteins and IGF receptors in Japanese Black beef bovine embryos derived using via SCNT, BNT, IVF, and *in vivo*, production was performed by Sawai et al. (2005) to determine whether the rates of developmental failure seen in nuclear transfer embryos could be related to changes in this complement of genes. The results indicated that the amounts of *Igf-1* receptor mRNA did not differ significantly among the types of embryos; in contrast, the amounts of mRNA of the *Igf-2r* differed depending on how the embryos were derived. In general, the proportion of embryos exhibiting *Igf-2r* receptor mRNA was more variable in embryos derived via SCNT. This is in contrast to the observations of Wrenzycki et al (2001), who found that the levels did not differ significantly. There are multiple reasons to account for the differences including the source of donor cells, their cell-cycle, etc. Heyman et al. (2002a) also reported that there were no significant differences in the proportion of embryos expressing IGF-2r transcripts among nuclear transfer, IVF and *in vivo* produced embryos. These apparently contradictory observations serve to imply that it is not some process associated with cloning per se (e.g., electrical pulses for cell fusion or other treatments) that adversely affect development, but rather provide further evidence

³⁰ Real-time PCR is a technique that allows for the rapid and precise identification and quantification of genetic material (in this case, RNA) during the actual time that the reaction is running.

that epigenetic dysregulation (i.e., failure of appropriate gene expression) that appears to be responsible for growth dysregulation.

Li S et al. 2005 also used real time PCR to compare expression levels of eight developmentally important genes in six organs from bovine clones that within 48 hours of birth relative to control animals produced by artificial insemination and also slaughtered within 48 hours of birth. Organs that were evaluated included the heart, liver, kidney, spleen, lung, and brain. Aberrant and highly variable gene expression in the clones occurred in a tissue-specific pattern, with the heart most (five of eight genes), and the kidney, least (two of eight genes) again indicating the role of gene expression in the ability of particular tissues and organs to develop appropriately in clones. They also noted that organ systems could be affected independently of others, implying a stochastic process at work. No mention was made of whether a similar study had been performed on live, healthy clone births in this report.

Finally, Archer et al. (2003a) have performed the most comprehensive study of the correlation between epigenetic reprogramming and live clone outcomes in a cohort of female swine clones. (More detailed discussions of the results of this study are found in Chapters V and VI). In addition to evaluating methylation in two different regions of the genomes of these animals and half-sibling comparators, the investigators studied the growth, clinical chemistry, and behavior (Archer et al. 2003b) of these animals. The overall degree of methylation between clones and their half-siblings was the same, with a small random variability in the PRE-1 SINE regions, and one CpG site in the centromeric satellite region. Further, the clones exhibited two patterns in specific phenotypic traits: one set of traits exhibited variability similar to the comparators, and another set showed less variability than the comparators. CpG methylation was measured in PRE-1 SINE (repeat sequence in a euchromatic region) and centromeric DNA (repeat sequence in a heterochromatic region) obtained from skin punch samples. Finally, the clones appeared to have grown and developed normally: no differences were observed between clones and their comparators with respect to growth rates, physiological measures of health, or behavior.

e. Studies of Technical Contributions to Epigenetic Variability in Clones and Other ARTs

In the previous discussion, we have repeatedly referred to the contribution of the methodology used to generate embryos (particularly the culture environment) as playing a critical role in developmental success for cloning and other ARTs. The continuing reports of relatively low efficiency of SCNT have stimulated a wide interest in how a variety of factors affect the efficiency of SCNT in particularly but including various ARTs with an eye to improving the process (Oback and Wells 2007a, Collas and Taranger 2006) particularly, but not exclusively, by improving nuclear reprogramming. A number of factors affecting the status of the cells used for

nuclear transfer seem to be important considerations for improving the efficiency of the process and recent reports identify the morphology, proliferative characteristics, chromosome stability (Giraldo et al. 2006; Mastro Monaco et al. 2006), cell type, culture conditions (Bosch et al. 2006; Inoue et al. 2006; Beyhan et al. 2007b), and stage of the cell cycle (Bordignon and Smith 2006) as important considerations for improving efficiency of SCNT.

Others reports are more directed at identifying the technical conditions that influence cloning efficiency with most focusing on improving reprogramming. Several investigators have examined the contribution of the oocyte to reprogramming in SCNT (Chen et al. 2006, Fulka and Fulka 2007) in the hope of identifying oocyte factors that increase reprogramming and consequently cloning efficiency. Preparation of the oocyte (Li GP et al. 2006) as well as timing (Sung et al. 2007) and method (Schurmann et al. 2006) of activation after fusion of the oocyte and transferred nucleus have been examined for their contribution for the process.

Reprogramming may be improved by the addition of exogenous “remodeling factors” such as nucleoplasmin (Betthausen et al. 2006) or even caffeine (Lee and Campbell, 2006). Although all of these studies (which use a variety of species and most of which harvest the embryos before term) identify factors in the process that are important, the efficiency of SCNT remains low.

Two studies by Wolf and colleagues (Hiendleder et al. 2004a, 2006) also provide insight into the relationship between reproductive methodology, methylation status, and fetal characteristics. The 2004 study compares a number of anatomic parameters to the “global” DNA methylation status of a variety of tissues from embryos produced using AI, IVF, and SCNT. In general, they correlate increased DNA methylation in fetal tissues and increased fetal size. In this study, 80 day AI fetuses were smallest and contained the least methylation, IFV fetuses of the same age were intermediate for both traits and clone fetuses, also 80 days old, were largest and had the most DNA methylation. The 2006 study extends these observations by focusing on the culture conditions used for IVF. This latter study demonstrates that the authors can manipulate the culture conditions so that under one set of culture conditions the fetuses are physically comparable to *in vivo* fertilized comparators while other culture conditions result in consistently large offspring as reported for IVF and SCNT derived fetuses as reported in their 2004 study. However, even in the “normal” size IVF fetuses, global methylation status is not identical to the comparators, indicating that further work needs to be performed to identify and characterize key parameters related to fetal overgrowth syndromes.

2. Gametogenic Reprogramming

The development mechanisms involved in gametogenic reprogramming were initially studied most extensively in the mouse; conservation of mechanisms involved in sexual reproduction are

similar in all species examined to date, although the timing of events differs depending on the length of gestation.

Germ cells (those developmentally destined to become gametes) are first detected as founder population cells at about embryonic day (E) 6.5 in the mouse. By E 7.2, approximately 45 primordial germ cells can be counted in the mouse embryo (Hajkova et al. 2002). These cells begin migration into the genital ridge (the portion of the embryo destined to become the reproductive organs) about 10 days after embryo formation (Hajkova et al. 2002, Yamazaki et al. 2003) (See Figure IV-2). Their epigenetic methylation status at this point resembles that of the rest of the embryo: they contain genomic imprints from the maternal and paternal genomes, and one of the two X chromosomes in female gametes has been inactivated in the somatic tissues (Surani 2001). Once the primordial germ cells migrate into the genital ridge (the thickening near the kidneys of the embryo that gives rise to the ovaries and testes), however, profound changes in their methylation status occur. A period of rapid demethylation ensues, in effect “erasing” all of the epigenetic modifications that were present on the cells prior to their migration (Yamazaki et al. 2003, 2005). This demethylation appears to be selective by affecting single copy imprinted and non-imprinted genes (*e.g.*, coding sequences), whereas the reprogramming of repetitive elements (whose function in the cell is not fully understood but is thought to be structural and regulatory) is more protected and incomplete.

In describing this phenomenon, Surani (2001) states that this “mechanism also erases any aberrant epigenetic modifications, so preventing the inheritance of epimutations, which consequently occurs very rarely.” The mechanism by which erasure of the epigenetic markings, including demethylation, in primordial germ cells is not yet understood. Other “resetting” mechanisms also occur in primordial germ cells, including the restoration of telomere length, and repair of lesions to the coding regions of the DNA (Surani 2001).

Random X inactivation in XX (female) germ cells also occurs during the migration phase of PGCs, coinciding with the timing of X inactivation in somatic tissues (reviewed by Avner and Heard 2001; Heard 2005). Inactivation of one X chromosome in female mammals is absolutely essential to compensate for the potential doubling of the “gene dosage” that a XX genotype would present. Although not fully understood, the process by which this occurs involves coating one of the X chromosomes by an RNA molecule itself encoded by a gene (*Xist*) on the X chromosome, followed by DNA methylation, and covalent modifications of the histones associated with the inactive chromosome. In mice, X inactivation first occurs in the placental trophoblast cells, where the paternal X tends to be inactivated by a mechanism thought to involve the expression of a maternal gene at the blastocyst stages that exclusively inactivates the paternal X chromosomes in the trophoblast cells. The end result is that the structure of the chromosome is

altered from an active, relatively loosely coiled state to a highly condensed and transcriptionally silent DNA molecule (Avner and Heard 2001).

Restoration of epigenetic modification in primordial germ cells in mice appears to take place several days later when the male germ line appears to acquire methylation at 15-16 days after conception. Remethylation of the female germ line in mice does not appear to occur until after birth during the growth of the oocytes, and probably continues until the first meiotic division (a stage in the maturation of the cells destined to become gametes in which the chromosome number is reduced from $2n$ to n) (Davis et al. 2000; Surani 2001). This overall process appears to be conserved in other mammals, although the exact timing may differ according to species.

Although the preceding discussion has focused on methylation as the primary marker of imprinting, it is important to remember that there are other modifications that may contribute to the retention of “epigenetic memory” in germ cells whose identity and mechanism remain to be characterized (Davis et al. 2000; Fazzari and Grealley 2004).

3. Mitochondrial Heteroplasmy

In addition to incomplete or inappropriate epigenetic reprogramming, the relatively low success rate of cloning has been hypothesized to be related to changes in the pattern of mitochondrial DNA (mtDNA) transmission following SCNT (Hiendleder 2007; St. John et al. 2005; Spikings et al. 2006). Because sperm deposit very few of their own mitochondria³¹ during sexual reproduction, mtDNA in developing embryos tends to come almost exclusively from the oocyte and tends to be maternally inherited. During the SCNT process, if intact donor cells are used as nuclear donors, following fusion with the enucleated oocyte, the resulting embryo may have mtDNA from both the donor and recipient cells *i.e.*, mitochondrial heteroplasmy. If the nuclear and mitochondrial DNA originate from different sources, the normal coordination of expression of nuclear and mtDNA may be altered, resulting in altered or impaired energy production in the cell or developing organism.

The extent to which mtDNA heteroplasmy is observed in animal clones is inconsistent (see reviews by St. John et al. 2005; Bowles et al. 2007; Hiendleder 2007). Bowles et al. (2007) reported that donor mtDNA is deleted by normal cellular regulation of mtDNA transmission during embryonic development in both NT embryos and clones themselves. Hiendleder (2007)

³¹ Mitochondria are the only organelles in animals that contain their own DNA, and are considered to be the “powerhouses” of eukaryotic cells. They are the site of the electron transport chain, which is the cell’s major source of energy. Proteins in the electron transport chain are encoded by chromosomal or mtDNA, necessitating tight coordination and regulation of expression between the nuclear and mtDNA. In sexual reproduction, these interactions, which govern mtDNA copy number, mitochondrial morphology, and the number of mitochondria per cell are tightly controlled to optimize energy production (St. John et al. 2005).

observed that most of the sheep, cattle, and swine SCNT clones investigated appeared to be homoplasmic (have nuclear and mtDNA from the same source) or display only mild heteroplasmy. Hiendleder (2007) further noted that factors involved in the process of nuclear transfer (*e.g.*, embryo culture conditions, the choice of donor cell types, and the quality of the oocyte recipient) may affect the level of heteroplasmy,

Even if mtDNA heteroplasmy were present at a significant level in clones, the extent to which it could affect clone health or food consumption risk is difficult to determine. Clones may show little or no heteroplasmy, or may have considerable levels of mtDNA diversity but be phenotypically normal (Bowles et al. 2007). Heteroplasmy may result in impaired mitochondrial function and energy production, contributing to the poor success rate of cloning, but the empirical demonstration of that possibility has not yet been proven. Smith LC et al. (2005) noted that “To date, there is no clear indication that heteroplasmy caused by nuclear transfer procedures in farm animals is detrimental to development.”

4. Conclusions from Studies of Epigenetic Reprogramming

- Inappropriate or incomplete epigenetic reprogramming is the source of the frank adverse outcomes and subtle anomalies that pose animal health risks in animals developed by SCNT or other ARTs.
- SCNT-derived embryos must demethylate the differentiated and generally relatively highly methylated nuclear donors to restore totipotency. The high rate of failure to progress beyond the early stages of cleavage of SCNT-embryos may be a function of the inability to carry out that demethylation, and likely involves other mechanisms, some of which may involve higher-order chromatin remodeling.
- In studies evaluating the differential reprogramming of trophectoderm- and ICM-derived tissues, more dysregulation is observed in the trophectodermally-derived tissues (placental tissues) than in the somatic tissues derived from the ICM. Whether this disparity is a function of the more stringent requirement of appropriate reprogramming of the ICM-derived tissues for survival (embryos and fetuses with significantly altered epigenetic reprogramming simply do not survive) is not known.
- Live and apparently healthy clones can exhibit some level of epigenetic differences relative to fertilization-derived comparators. Many of these differences appear to resolve as the animals age, consistent with the adaptation observed in clone populations studied for physiological and growth parameters. It is not known whether these animals are

tolerant of these differences, or whether a “threshold” of epigenetic differences exists that has not been exceeded in the live and apparently healthy animals.

B. Phenotypic Evidence for Gametogenic Reprogramming

The initial observations confirming the biological assumption that phenotypic expression of underlying inaccurate epigenetic reprogramming observed in clones disappear in the progeny due to gametogenic reprogramming come from the studies of Shimozawa et al. 2002a and Tamashiro et al. 2003, who demonstrated that a phenotype observed in mouse clones was not transmitted to their progeny. These studies have led to the conclusion that “Progeny of animal clones, on the other hand, are not anticipated to pose food safety concerns, as natural mating resulting from the production of new gametes by the clones is expected to reset epigenetic reprogramming errors that could persist in healthy, reproducing clones” (NAS 2002a). Or stating a similar conclusion “. . . epigenetic rather than genetic aberrations are the cause; epigenetic changes, in contrast to genetic changes, are reversible modifications of DNA or chromatin that are usually erased in the germ line” (Hochedlinger and Jaenisch 2002). This postulate can be further summarized as: “All epigenetic problems in the parents seem to be erased when cell nuclei go through the germ line” Yanagamichi (2002), and, “. . .the progeny of cloned animals will be normal” Fulka et al. (2004b).

In the following section, the studies that have led to these conclusions, as part of a summary of the utility of the mouse model for estimating risks in livestock clones are reviewed. It is organized by Developmental Nodes, as in the Critical Biological Systems Approach to evaluating the health status of livestock clones, although several nodes are combined to better reflect the existing mouse dataset.

1. Phenotypic Anomalies Observed in Mouse Clones

a. Utility of Mouse Model

Although the subject animals of this assessment are domestic livestock clones, the use of the mouse as a model system provides some insights into the underlying biology of the cloning process and its implications for food safety, particularly for understanding the role of sexual reproduction in resetting residual epigenetic reprogramming errors. SCNT in mice was first reported by Wakayama et al. (1998) using the “Honolulu technique” at approximately the same time as publication of the “Dolly” paper (Wilmut et al. 1997). Since that time, mice have been cloned from a range of cells from embryonic and adult sources (reviewed by Yanagamichi

2002). The mouse model is useful because of its well-characterized genotypes, small size, short generation period, and shorter life span than larger animals.

b. Pregnancy (Developmental Node 1)

The key measure of the success of SCNT is the normal development, maturation, and reproduction of the animal clones. As with livestock, the efficiency of this process in mice is very low, and in the same range as livestock: approximately 0.2-3.4 percent when calculated from the total number of reconstructed embryos resulting in live offspring (Yanagimachi 2002). In mice, the rate of embryo survival is most reduced early in development, particularly in the days immediately before and after implantation (Yanagimachi 2002). Yanagimachi (2002) also found that more than 90 percent of mouse embryos cloned with cumulus cells had normal chromosomal constitutions, indicating that the poor survival rates are not due to chromosomal problems, again pointing to epigenetic reprogramming as the determining factor in cloning efficiency.

Placental enlargement has been observed in almost all of the studies of mouse clones reported to date (Wakayama and Yanagimachi 1999; Humphreys et al. 2001; Ono et al. 2001b; Tanaka et al. 2001; Ogura et al. 2002; Yanagimachi 2002). Tanaka et al. (2001) performed histological examination of term placentae from mouse clones and evaluated the expression of a number of genes relevant to fetal development. Placentae from these animals were larger than from conventional controls, and exhibited histological changes in all three layers of the placenta (*i.e.*, the trophoblastic giant cell, spongio-trophoblast, and labyrinth layers). Most of the anomalies appeared to be related to the expansion of the spongio-trophoblast layer, which exhibited an increased number of glycogen cells and enlarged spongio-trophoblast cells. Despite these morphological changes, there were no critical disturbances in regulation of gene expression in the placentae associated with term clone placentae. Unlike cattle and sheep, in which clone fetuses tended to be larger than comparators, the average weight of the mouse clone fetuses appeared to be lower than that of comparators, suggesting that a “latent negative effect from somatic cell cloning may occur on fetal growth, potentially due to incomplete placental function” (Tanaka et al. 2001). Despite the morphological changes observed in their study, Tanaka et al. (2001) noted that the placentae “could support full development of the fetus, suggesting that their functions are adequate for apparently normal fetal development” similar to the observation of Hill et al. (2000b) for cattle clones.

Both Ono et al. (2001) and Ogura et al. (2002) reported morphological changes in the placenta of mouse clones similar to those observed by Tanaka et al. (2001). Ono et al. (2001) observed that increased placental size was caused by proliferation of the trophoblastic cells, endometrial

glycogen cells, and unusually large giant cells. They also found limited distribution of maternal blood vessels in the spongio-trophoblast layer and suppressed development of the labyrinth layer. They suggested that these abnormalities would greatly reduce the functional capacity of the placenta and could contribute to high rates of neonatal death in mouse pup clones derived from somatic cells. Ogura et al. (2002) compared the histological findings for mouse clone placentae with those of embryos derived from other micromanipulation techniques, such as microinsemination, aggregation chimera, and pronuclear exchange. Disruption of labyrinth layer morphology was common to placentae from cloning and other micromanipulation techniques, whereas disruption of the basal layer with marked proliferation of glycogen cells was the only phenotype unique to cloning.

The underlying mechanisms responsible for the observed placentomegaly are unknown, but Tanaka et al. (2001) cite their previous findings (Ohgane et al. 2001) of aberrant methylated genomic regions in placental tissues and suggest that slight disturbances in the expression of a number of genes, rather than a drastic change in the expression of a single gene, may impact on placental growth and function. Humpherys et al. (2002) reported that approximately 4 percent of the expressed genes in placentae from nuclear transfer-derived mouse clones differed dramatically in expression levels from those in controls. Placental size was not correlated with abnormal gene expression, indicating that the changes in cellular composition observed in Tanaka et al. (2001) are unlikely to account for the observed expression changes (*i.e.*, changes in placental gene expression did not reflect changes in relative abundance of certain cell types). Ono et al. (2001) and Wakayama and Yanagimachi (2001) speculated that the observed placental abnormalities may be a function of disrupted patterns of expression of imprinted genes important for placental development. However, Inoue et al. (2002), using donor cells from a number of different sources, found that placentae of mouse clones at term were two to three times larger than those of controls, despite the developmentally appropriate expression of imprinted genes in both the placentae and fetuses of mouse clones. They concluded that placental genes were thus regulated by some upstream function that is independent of imprinting and is either dysregulated by nuclear transfer cloning itself, or by some other aspect of nuclear transfer.

More recently, Ohgane et al. (2004) investigated whether placental overgrowth was related to the existence of aberrant DNA methylation at certain loci (and subsequent abnormal gene expression) in mouse clones. They identified a tissue-dependent differentially methylated region within the *Sall3* locus that is hypermethylated in the placenta of all mouse clones examined. Ohgane et al. concluded, given that the methylation rate of the *Sall3* locus correlated with the occurrence of placentomegaly in mouse clones, this was an example of “a genomic locus highly susceptible to epigenetic error caused by nuclear transfer.”

c. Perinatal Period (Developmental Node 2)

As in the pregnancy and parturition developmental node, mouse clones have demonstrated some of the same abnormalities observed in the perinatal periods of larger mammalian clones, including reports of perinatal deaths from respiratory problems similar to that observed in cattle clones (Wakayama and Yanagimachi 1999; Eggan et al. 2001; Yanagimachi 2002). Interestingly, LOS, a relatively high frequency event in cattle cloning, was only evident in one mouse study (Eggan et al. 2001).

Eggan et al. (2001) investigated whether the phenotypic abnormalities noted in mouse clones, such as loss of neonatal growth control, respiratory failure, and high neonatal mortality, were due to the effects of nuclear transfer, or instead reflected some fundamental characteristic of the cell(s) used as donors. Using mouse embryonic stem cells with either inbred or hybrid (F1) genetic backgrounds, they compared the phenotypes of animals created by either tetraploid embryo complementation or nuclear cloning. After evaluating four endpoints (embryos transferred to surrogate dam, pups alive at term, pups respiring after Caesarian section, and pups surviving to adulthood) the authors concluded that genetic heterozygosity (*i.e.*, hybrid vigor) was crucial for influencing the survival of mouse clones. They further concluded that difficulties with neonatal mouse clone survival and respiratory competence were a function of the genetic makeup of the donor cell nucleus, whereas neonatal overgrowth was more likely to be a consequence of the nuclear transfer procedure.

Ogura et al. (2002) reported that more than 90 percent of mouse clone fetuses that developed to term were mostly normal. Birth weights were not significantly different from controls (produced by IVF or spermatid injection), and fetal overgrowth was not observed. This is in contrast to the high incidence of placental enlargement observed in these studies (as discussed earlier in this Chapter). Of the 159 term pups, 12 had abnormalities: umbilical hernia (two cases), respiratory failure (six), developmental retardation (one), severe anemia (one), and intrauterine death shortly before birth (two).

d. Juvenile Period to Reproductive Maturity (Developmental Nodes 3 and 4)

The amount of information on the health status of mouse clones from postnatal development to reproductive maturity is limited. The finding of note within this period was postpubertal obesity in mouse clones reported by a single research group.

Tamashiro et al. (2000) evaluated the postnatal growth and behavioral development of mice cloned from adult cumulus cells relative to control mice specifically generated to eliminate

confounding factors associated with the effects of embryo micromanipulation, *in vitro* embryo culture, embryo transfer, litter sizes, Caesarean delivery, and pup placement with lactating foster mothers. No physical abnormalities were noted at birth or through the course of the study. Body weight at birth was not statistically significantly different between clones and controls. Beginning at approximately 8-10 weeks, however, the body weights of the clone group were significantly higher than that of controls. The late onset of increased body weight in clones was distinguished by the authors from the LOS observed at birth in many mammalian clones. Although preweaning development of these mouse clones was similar to that of controls, there was a delay in first appearance of eye opening, ear twitch, and negative geotaxis (the ability of mice placed on a downward slope to turn and climb upwards). Subsequent tests of spatial learning, memory, and motor abilities in the same subjects did not show any deficits or long-term behavioral alterations. There was no significant difference in activity levels of clones compared to controls up to 180 days of age. The authors concluded that the cloning procedure did not adversely affect the overall postnatal behavior of mice.

Tamashiro et al. (2002) further investigated the obesity phenotype in mouse clones of two different background strains (B6C3F1 and B6D2F1). Comparisons were made relative to two groups: animals manipulated *in vitro* similar to SCNT-derived animals (*in vitro* embryo manipulated, or IVEM, mice), and stock (conventional) control mice. At birth, animals derived from *in vitro* manipulation (mouse clones and IVEM mice) were both heavier than stock control mice. Clones and IVEM mice gained about the same amount of weight over the next eight weeks, after which time the clones became significantly heavier than either IVEM or stock mice. Clones continued to weigh more than controls throughout their lives, unlike control animals whose body weight peaked at approximately 18 months of age. The increased body weight was independent of the strain of mouse used as the nuclear donor. Although mouse clones ate more than the IVEM mice, they consumed approximately the same amount of food as the stock mice. All animals lost the same percentage of baseline body weight when deprived of food, and all animals compensated by increasing consumption when it was returned. Carcass analysis showed that clones had more body fat than either the IVEM or stock mice. Mouse clones had increased plasma levels of leptin and insulin than either control group, whereas plasma corticosterone levels in mouse clones did not differ significantly from the control groups.

The authors concluded that mouse clones are truly obese and are not simply larger than controls. The process of *in vitro* culture appeared to be a factor in body weight, given that both the IVEM mice and clones were significantly heavier than controls. Further, the clones had more carcass fat than the IVEM mice, suggesting that some aspect of the somatic donor cell or the nuclear transfer technique may be a causative factor in the development of obesity. Faulty epigenetic programming was proposed as a possible mechanism responsible for the obesity phenotype observed in these clones.

Further study by Tamashiro et al. (2002) to determine whether a malfunctioning leptin-melanocortin system was involved in the observed obesity, involved administering melanocortin 4 receptor (MTII) and leptin, known suppressors of intake, to mouse clones and examining food intake. Inui (2003), who analyzed the results of Tamashiro et al. (2002) in context of knowledge gained of the leptin-melanocortin system from studies in rodent models of obesity and human obesity, agreed that the phenotype observed in mouse clones is unique, is not due to defects in the leptin-melanocortin system, and may be attributable in part to cloning procedures. Tamashiro et al. (2003) provides a more thorough discussion of the role body weight regulatory systems may play in this phenotype, but concludes that the mechanisms for the observed obesity remain to be elucidated. Inui (2003) proposed that inappropriate placentation may be at least partially responsible for the obese phenotype. This opinion is based on observations in other species, including humans, indicating that decreased intra-uterine nutrient levels can have significant repercussions on later human health. For example, diabetic human mothers have been observed to have births that result in large placentae, altered birth weights, respiratory distress syndromes, and subsequent obesity and diabetes in offspring (the “thrifty-phenotype” hypothesis) (Hales and Barker 2001).

To determine whether the obese phenotype was likely due to events in the cloning process, or the result of a genetic mutation, Tamashiro et al. (2002) mated male and female mouse clones and found that the offspring did not appear to be obese, nor did they have the enlarged placentae commonly found in mouse clones. Obesity was, therefore, not transmitted through the germline, indicating to the authors “that epigenetic modifications that occur during the cloning procedure are eliminated, or ‘corrected’ during gametogenesis.” The authors further proposed that “reproduction by natural mating may be recommended as soon as offspring with specific desired traits are produced by cloning.”

With respect to other possible health outcomes in this developmental period, Ogura et al. (2002) reported that more than 90 percent of mouse clones reached puberty when nursed by good foster mothers, a rate not significantly different from that of microinsemination-derived mice. In this study, cloning did not appear to have any adverse effects on reproductive performance. Of the 25 animals studied, no cases of complete sterility were observed; two female clones delivered only one litter and then became sterile for unknown reasons. No further details were provided.

e. Maturity and Aging (Developmental Node 5)³²

Given that one of the advantages of using the mouse model to study SCNT is the relatively short life span of mice compared to livestock animals, the impact of SCNT cloning on maturity and

³² For a discussion of telomeres and their possible role in aging, see Chapter V.

aging of animal clones has been examined in several reports. Ogonuki et al. (2002) followed weight gain, serum biochemical values, and lifespan in a group of 12 male mouse clones derived using immature Sertoli cells as donors, and compared them to the same values from male mice with the same genetic background derived from natural mating or spermatid injection. At one year after birth, weight gain of mouse clones did not differ from that of natural mating controls. Of the 16 serum biochemical values measured at 3 and 14 months of age, only lactate dehydrogenase (LDH), and ammonia (NH₃) were significantly higher in clones than in control mice. Clone survival rate, however, was significantly different from the two control groups. The first death in the clone cohort occurred 311 days after birth, and 10 of the 12 animals died before day 800. Histopathological examination of necropsy samples of six of the mouse clones revealed severe pneumonia (6/6), extensive liver necrosis (4/6) and tumors (leukemia and lung cancer, 1/6 each). Elevated serum LDH and ammonium levels were consistent with liver damage.

Immune function also was investigated by Ogonuki et al. (2002) in a different group of animals derived from Sertoli cells. In mouse clones as early as 4-5 months of age, antibody production following injection of live bacteria was significantly reduced relative to age- and genotype-matched controls. Phagocytic activity was also lower than controls, although the difference did not reach the level of statistical significance.

Ogura et al. (2002) provided additional information on the same mouse clones. The longest surviving clone died at 857 days of age, with the 50 percent survival point of the mouse clones at 550 days, relative to the 1,028 days for the naturally mated control animals. The average lifespans of the two control groups (natural mating vs. spermatid injection) were not significantly different. The authors suggested that the major cause of early death was related to dysfunction of the respiratory system. Necropsy results showed that all six examined clones had severe pneumonia that resulted in destruction of alveolar structures throughout the entire lobes. Given that the animals were maintained in a pathogen-free environment, and the observed reduced immunocompetence, the authors suggested that the respiratory effects were caused by chronic infection by opportunistic organisms that are usually asymptomatic in immunocompetent mice. Interestingly, the early pneumonia-associated death of mouse clones was restricted to mice of a specific genetic background (B6D2F1). Clones of other genotypes exhibited neither early death nor severe pneumonia.

In his overview of mouse cloning, Yanagimachi (2002) reported that in his laboratory's experience, mice cloned with adult cumulus cells, tail-tip cells, and embryonic neural cells generally had normal life spans with no serious health problems before death except for the postpubertal obesity as described by Tamashiro et al. (2000, 2002). In reviewing the Ogonuki et al. (2002) data, Tamashiro et al. (2003) stressed the importance of considering the age and type of donor cell used in the animal clones, as they may influence the health status of the animal

clone later in life. This is especially important in attempting to extrapolate data to other mouse clones, or other animal clones. Tamashiro et al. (2003) cite the immature Sertoli cells used by Ogonuki et al. (2002) as possibly harboring defects that would result in adverse effects such as the observed hepatic failure and immune incompetence. Tamashiro et al. (2003) summarized their own experience with mouse clones, observing that histopathology at the time of death of their cumulus cell clones indicated that most died of conditions associated with normal aging, and that the lifespan of their clones was comparable to animals followed by the National Institute of Aging.

2. **Conclusions from Phenotypic Studies of Gametogenic Reprogramming in Mouse Clones and their Progeny for Reprogramming in Domestic Livestock Clones and their Progeny**

- Mouse clones offer insights into physiological mechanisms that may be perturbed in animal clones, and provide evidence that certain epigenetic changes may lead to common anomalies in livestock clones. Placental enlargement, an outcome observed in cattle and sheep clone pregnancies, also has been observed in mouse clone pregnancies and appears to be linked to dysfunctional reprogramming of cells of trophectodermal origin. Fetal size, on the other hand, does not appear to be increased in the animals with placental enlargement, and in fact, appears to be decreased in mouse clones.
- The mouse literature also confirms that the genetic make-up of the donor cells is critical in the development and growth of the animal clone, and that cloning methodology (*e.g.*, *in vitro* culture conditions, effects of micromanipulation, methods of oocyte activation, technical skill) may also have a significant effect on cloning outcomes (see also Chapters V and VI).
- At this time, it is not possible to say whether the life span shortening observed in one strain of mouse clones will be observed in other species of clones. The shortened life-spans of mouse clones appear to be due to chronic alterations in metabolism, while the only observed early deaths of livestock clones appear to be due to more acute phenomena. Nonetheless, it is too early to make a definitive judgment on longevity, as most domestic livestock clones have not yet begun to approach even the midpoint of their natural life-spans (See Chapters V and VI).
- Clones are not the only animals that exhibit differences in epigenetic programming relative to their genetic antecedents. There are examples of fertilization-derived

embryos responding to dietary levels of methyl donors in their dam's diets resulting in offspring whose phenotypes differ significantly from their parents. Although the cited case provides a clear molecular correlation between the exposure and outcome, it is important to remember that epigenetic markers are reversible by "nature's design," and are intended to help provide organisms with multiple, interactive mechanisms with which they may adapt to environmental challenges.

The most important implication of the mouse clone literature for domestic livestock clones is the observation that anomalies noted in clones are not transmitted to their progeny. The obese phenotype, for example, is not transmitted to progeny of those clones, and progeny of mouse clones appear to be normal and healthy. This observation is consistent with the biological assumption that gametogenesis effectively "re-sets" epigenetic markings, and allows for the appropriate development of normal organisms (*i.e.*, sexual reproduction). It is also consistent with the limited but consistent observations of healthy, fully functional progeny born to domestic livestock clones. Thus, the empirical evidence supports the assertion that "*Progeny of animal clones, on the other hand, are not anticipated to pose food safety concerns, as natural mating resulting from the production of new gametes by the clones is expected to reset epigenetic reprogramming errors that could persist in healthy, reproducing clones*" (NAS 2002a).

C. Implications of Epigenetic Reprogramming for Animal Health and Food Consumption Risks

The Center assumes that if clones were to pose food consumption risks, the only mechanism by which those risks could arise would be from inappropriate epigenetic reprogramming, similar to those observed for other ARTs. It is important to note that the genes that are being dysregulated are the "normal," naturally present genes that comprise the animal's genome, and have not been introduced via recombinant DNA techniques from other sources (*i.e.*, these are not transgenic or genetically engineered animals).

- Anomalous epigenetic reprogramming is observed at the global genomic and individual gene level in clone embryos and fetuses, and in similar developmental stages of animals produced using ARTs with significant *in vitro* culturing components. Various factors influence the success rate of SCNT and these other ARTs, including the source of the donor cells and oocytes, culture medium, and factors that have not yet been identified. Many of these anomalies are lethal, as demonstrated by the low success rate of IVF and the even lower success rate of SCNT.

- Because abnormalities arise from the dysregulation of intrinsic genes, adverse outcomes that would likely be expected in clones and animals derived via other ARTs are those that result from the inappropriate development of tissues and organs. For example, it would be reasonable to expect both overgrowth phenomena, and the poor development (aplasia or hypoplasia) of tissues and organs. Examples of outcomes that affect the health status of animal clones are presented in detail in Chapter V (Animal Health) and Appendix C, and those that may have an impact on food consumption risks are described in Chapter VI.
- The studies that have evaluated epigenetic reprogramming of live, healthy clones indicate that although there is some variability between clones and their fertilization-derived counterparts, clones are capable of carrying out sufficient methylation-based reprogramming (and other coordinated functions) to allow for survival. Molecular analyses reveal relatively small methylation differences, and either the animals are tolerant of such differences, or the epigenetic differences are below the threshold that poses observable adverse health outcomes.
- It may be, as many have suggested (Wilmut 2002b; Jaenisch et al. 2004), that no clone is completely “normal” with respect to its epigenetic profile. Although this is an important point for assessing the overall safety of the cloning process for any particular species, the relevance of “epigenetic normality” to food consumption risks is unclear. Further, because similar abnormalities have been noted in animals produced using other ARTs, the issue of defining normality becomes significantly more complex. It may be that normality encompasses a range on a continuum, and that animals that are healthy, meet appropriate developmental and behavioral milestones, reproduce and bear healthy young are “normal,” regardless of their epigenetic status. The most compelling conclusions that can be made about food consumption risks, then, are drawn from assessments of the health status of the animals and the composition of food products derived from them, and not from gene expression studies.
- Progeny of animal clones, on the other hand, are not anticipated to pose food safety concerns, as natural mating resulting from the production of new gametes by the clones is expected to reset even those residual epigenetic reprogramming errors that could persist in healthy, reproducing clones (Tamashiro et al. 2002; Yanagimachi 2002; NAS 2002a, 2004, Fulka et al. 2004). Thus any anomalies present in clones are not expected to be transmitted to their progeny.

Chapter V: Animal Health Risks

Chapter V: Animal Health Risks

A. Potential Hazards and Risks to Animals Involved in Cloning

This analysis identifies hazards and characterizes risks to animals involved in somatic cell nuclear transfer (SCNT) in the context of other assisted reproductive technologies (ARTs) in use in current US agricultural practice. Although hazards have been identified in the literature, a systematic assessment of potential risks is difficult, due to the relative newness of the technology, and the variability in outcomes among laboratories and species cloned. This section reviews the publicly available information and applies existing knowledge of animal biology and agricultural practices to cast that information in a risk context.

In addition to characterizing hazards, this chapter identifies information gaps that when filled may provide a more complete understanding of the risks to animals associated with SCNT technology. It is not intended to be an overview of all the techniques used to produce clones, nor does it review studies that attempt to optimize the early stages of clone embryo production. Rather, this analysis focuses on those studies relevant to the overall objective of this risk assessment, namely, identifying the biological risks that cloning poses to animal health

Because of the diversity of approaches in the peer-reviewed studies, CVM has relied on various ARTs including an earlier type of “cloning” called blastomere nuclear transfer (BNT) for context. Current agricultural statistics also are used to provide readers with a frame of reference for these technologies (see Appendix B). Outcomes for various ARTs are located in Appendix C. Peer-reviewed reports of primary findings were used as references for SCNT, while some recent reviews of artificial insemination (AI), embryo transfer (ET), and *in vitro* produced embryos (IVP), as well as primary data reports, were employed as references for the older ARTs.

Most of the studies on SCNT and other ARTs that are of utility for identifying and assessing risk to animals, and that make up the subject of this Risk Assessment are in ruminants.³³ Cattle studies are the most abundant, followed by sheep, swine (a non-ruminant species) and goats. Peer-reviewed research reports on these four species, with supplemental data from studies in mice, primarily have been used as the basis for this assessment. Additionally, CVM evaluated veterinary records, blood clinical chemistry and hematology, and urinalysis provided by two private firms: (1) Cyagra, Inc. provided data on 134 individual cattle clones ranging from birth to approximately one and a half years of age (Appendix E); and (2) ViaGen, Inc. provided data on

³³ Ruminants are animals with a complex or compartmental stomach, such as cattle, sheep, and goats.

11 swine clones and 402 progeny of swine clones through slaughter age (Appendix F). Additional unpublished data were provided by several sources, in the form of veterinary records, blood chemistry and hematology, and reproductive performance on small groups of cattle and swine clones (Appendix G).

Publications from peer-reviewed journals were searched for information relating to health of surrogate dams, animal clones, and clone progeny. Whenever possible, data on contemporary comparators have been used to provide reference rates for purposes of comparison. Where comparisons were not made within a study, the historical literature and other available databases (*e.g.*, USDA National Agricultural Statistics Service (NASS³⁴) or National Animal Health Monitoring Service (NAHMS³⁵)) were searched for applicable comparative information. For example, Table V-1 (Survival Rates of Live-Born Bovine Clones and Comparators) presents data on survival rates of clones and comparators, drawn from both contemporaneous comparators and historical datasets. Descriptions of how other data were analyzed are described in Appendix E (Cyagra Data), Appendix F (ViaGen Data), and Appendix H (Comprehensive Veterinary Exam and Its Interpretation).

B. The Critical Biological Systems Approach to the Analysis of Clone Animal Health: Cattle, Swine, Sheep, and Goats

To provide an assessment of animal health risks that was as comprehensive as possible, the CBSA was applied at all five developmental nodes at multiple levels of observation and analysis. In addition to macroscopic anomalies observed in surrogate dams and clones themselves, the information in this chapter includes available evidence to indicate whether subtle hazards exist that might affect the health of clones. For the purposes of this risk assessment, subtle hazards may be thought of as physiological anomalies that may be present in apparently normal animals, but are not obvious because their identification requires analysis of physiological parameters in blood and tissues (*e.g.*, clinical chemistry, hematological measurements, hormone levels).

1. Pregnancy and Parturition (Developmental Node 1)

Pregnancy is a remarkable time in mammalian development. A carefully orchestrated and incompletely understood sequence of changes in both the pregnant female and developing embryo/fetus must occur to produce a successful outcome: a healthy newborn and mother. Despite this complexity, most pregnancies in domestic livestock proceed normally and result in healthy offspring.

³⁴ <http://www.nass.usda.gov/>

³⁵ <http://www.aphis.usda.gov/vs/ceah/ncahs/nahms/index.htm>

Criticisms of cloning point to the “inefficiency” of the process, which is often translated to mean that successful outcomes are relatively uncommon (Wilmot 2002). Reports of early pregnancy loss or later-term spontaneous abortion of embryonic and fetal clones are frequently cited in the literature (Le Bouhris et al. 1998; Kishi et al. 2000; Chavatte-Palmer et al. 2002; Lee RS et al. 2004). Loss due to defects in the embryo or failure to implant in the uterus of the surrogate dam does not pose a hazard to the dam at this early stage. Rather, the female simply resorbs any embryonic tissues and returns to cycling (Merck Veterinary Manual Online 2005³⁶). Mid- and late-term spontaneous abortions may be hazardous to surrogates if they are unable to expel the fetus and its associated membranes, possibly resulting in metritis (uterine infection), retained fetal membranes (in which the placenta is not expelled), or a mummified (dead, desiccated) fetus. Other complications can occur during pregnancy and labor that may pose a risk to both the pregnant female and the fetus. Developmental Node 1 examines the causes and frequency of pregnancy complications, and the relative risks to both the female and fetus, using other ARTs for comparison where such data are available.

It is important to note that external factors unrelated to breeding method such as animal management and environment that can influence pregnancy outcomes. In evaluating any ART, including cloning, the potential impact of these external influences should be considered before assigning the cause of pregnancy loss to the technology itself. For example, stress is an important risk factor in the loss of any pregnancy, particularly in the preimplantation phase (before the embryo attaches to the uterine lining). Disease, under-nutrition, and severe environmental conditions (*e.g.*, high ambient temperature) are stressors known to interfere with animal fertility and embryo survival (Lucy 2001, Merck Veterinary Manual Online 2005). In these cases, the risk to the pregnancy is directly related to those stress factors, not the technology used, and must be mitigated in order for normal reproduction to resume.

Another factor to consider is the methodology used in the SCNT process. A review of the literature suggests limiting *in vitro* manipulation of the embryo or changing the culture conditions to more closely resemble oviduct/uterine conditions may improve the chances for successful pregnancy outcomes. Many of the abnormalities reported in cattle and sheep pregnancies have not been noted in goats or swine carrying SCNT clones. Of the reports reviewed for this assessment, goat embryos were only cultured through the first or second cleavage stage (less than one day in culture) before transfer to the recipients (Keefer et al. 2002), compared with sheep and cattle, whose embryos were generally cultured to the blastocyst stage (seven to eight days in culture) prior to transfer. Walker et al. (2002) reported success after only brief *in vitro* culture of swine embryos (1-3 hours after activation) before transferring to recipients. Onishi et al. (2000) also reported the successful birth of SCNT pigs following culture

³⁶ <http://www.merckvetmanual.com/mvm/index.jsp>

to the 2 to 8 cell stage (one or two days in culture), while none of the embryos cultured to the blastocyst stage developed to term. In contrast, ViaGen, Inc. has indicated that they have had greater success recently transferring swine clone blastocysts (5 days *in vitro* culture) into surrogate dams (see CVM Memorandum II at www.fda.gov/cvm/cloning.htm). Lagutina et al. (2006) reported the birth of pigs following culture of swine embryos for six days in synthetic oviductal fluid (SOF) supplemented with amino acids, rather than supplementation with serum.

Abnormalities in cattle and sheep clones may result from incomplete reprogramming of the donor nucleus. As noted in Chapter IV, epigenetic reprogramming occurs at different times in embryos in different species, possibly in relation to gestation length. Despite that observation, it is interesting to note that although goats and sheep have the same gestation length (about five months), abnormal pregnancy outcomes are frequently reported with SCNT sheep, whereas SCNT goats have had relatively few problems reported (Wells et al. 1998a; Young et al. 1998; Baguisi et al. 1999; Reggio et al. 2001; Keefer et al. 2002; Ptak et al. 2002). It is important to note that epigenetic remodeling has been studied primarily in mice, swine, and cattle, and that very little is known about the timing and extent of reprogramming in small ruminants.

The biology of placental attachment also may account for differences among pregnancy outcomes in the species evaluated in this risk assessment. In contrast to ruminants with a “cotyledonary” (cotyledon³⁷) type attachment via placentomes (see discussion below on this type of fetal attachment to the uterine lining), swine have what is classified as a “diffuse” type of placenta where fetal attachment occurs over the entire surface of the placenta and uterine lining (Hafez and Hafez 2000). This gross morphologic difference in fetal attachment may influence outcomes of clone pregnancies in the ruminant vs. swine species.

2. Perinatal Period (Developmental Node 2)

The perinatal period (from initiation of labor through approximately one week post partum) is one of the most critical times in the lives of all young animals. Several studies (reviewed by Moore et al. 2002) noted that 75 percent of mortality from all causes for naturally produced and AI beef calves occurred within the first seven days of life.

The process of labor and birth can be as stressful on the neonate as it is on the dam, particularly if complications arise during the process. The newborn must begin breathing almost immediately after birth, either spontaneously or with stimulation from the mother or human attendant. For

³⁷ Cotyledons are the structures in ruminant placentae that form contact points between the fetal-derived placental tissues with the maternal caruncles (attachment points) of the uterus to form the functional units called placentomes. Placentomes allow for the passage of gases and nutrients from the dam to the developing fetus, as well as the removal of waste products from the fetus to the dam’s blood stream, for final elimination.

ruminant animals, as for other herbivores, it is instinctive for the newborn to attempt to stand within the first 5-15 minutes after birth, and to suckle shortly thereafter. Swine are less mature at birth than most other farm livestock, and although they are able to walk and nurse almost immediately after birth, they are not able to control their body temperature (known as thermoregulation) for the first 10 to 14 days of life, and generally require supplemental heat.

In mammals, neonates have little endogenous immune protection from disease during the first few weeks to months of life. Young mammals are dependent on antibodies transmitted from their dams either through the placenta or by consumption of colostrum (the antibody- and nutrient-rich first fluid secreted by the mammary glands after birth preceding the production of true milk). The process of providing immunity to the offspring in this manner is called passive transfer of immunity. In ruminants and swine, the principal means of this transfer is through colostrum. In species where this form of transfer predominates, the neonate must consume colostrum as soon after birth as possible to insure intestinal absorption of functional immunoglobulins, large proteins which contain antibodies (Merck Veterinary Manual Online 2005³⁸). Within approximately 48 hours after birth (although this may vary among species), the neonatal intestine loses the ability to absorb large, functional proteins, (a process known as “gut closure”) and the opportunity for this method of immune transfer is lost (Donovan 1992).

3. Juvenile Developmental Node (Developmental Node 3)

Another critical period in the lives of young mammals is immediately post-weaning to approximately six months of age. In general, health and survival of any young animal post-weaning is dependent on management conditions. Relatively little information has been published in the peer-reviewed literature on health and survival of animal clones during this developmental node. As previously discussed, one clone producer has supplied data (Cyagra, Inc.), including health records and laboratory measurements that have been evaluated along with the published literature; these may be found in Appendix E.

Age at weaning varies among species, breeds, and individual farm management. Swine are typically weaned at about 21 days of age, but may be weaned as early as 10 to 14 days. Sheep and goats may be weaned between 8 and 12 weeks of age. Dairy calves typically receive milk replacer (after colostrum consumption is complete) until 28 to 60 days, when they are weaned to solid feed. Beef calves may remain with their dams and continue to nurse for four months or longer.

³⁸ <http://www.merckvetmanual.com/mvm/index.jsp>

Weaning is a period of stress for all developing animals. Weight loss is common during weaning as the young animal must compensate for the loss of a primary source of nutrition and adapt to what previously may only have been offered as a supplement. Changing diet can induce scours (diarrhea), particularly if it is done abruptly. Diarrhea is a common ailment in all young mammals, and can be serious, resulting in dehydration and death if not treated in a timely manner (Merck Veterinary Manual Online 2005³⁹). In addition, between two and six months of age in ruminants, or as early as 21 days in swine, maternally derived immunity wanes, and the young animal must depend on its own immune system. In some animals, such as beef cattle, this may occur concurrently with transportation stress when they are sold to feedlots or stocker operations, resulting in relatively high losses.

4. Reproductive Development and Function Node (Developmental Node 4)

Due to the complexity of the reproductive system, careful attention was directed to reports of puberty and reproductive function in clones in order to determine whether cloning had perturbed this delicately balanced system. Data from this stage of development in animal clones are sparse, however.

In conventional cattle, inappropriate intrinsic, nutritional, and environmental factors have been shown to adversely influence reproduction in both male and female conventional animals. Under- and over-nutrition can influence the age at puberty and, particularly in the case of under-nutrition, can disrupt the normal estrous cycle. Environmental stressors such as extreme heat or cold can also suppress normal cycling and estrous behavior in females and reduce fertility and libido in males (Lucy 2001). Derangements in metabolic pathways, such as hypothyroidism, genomic disorders manifesting as freemartins⁴⁰ and hermaphrodites,⁴¹ as well as congenital anomalies such as hypospadias⁴² can also result in reproductive failure (Merck Veterinary Manual Online 2005).

Considerable differences exist among species and even among breeds within a species for age at puberty. In cattle, puberty is related to body weight, and a heifer will achieve her first estrus when she reaches approximately 65 percent of her adult body weight. Depending on management, then, heifers will typically begin cycling between 10 and 13 months of age. Goats and sheep mature at a younger age, with first estrus typically occurring between seven and eight months. Dwarf goat and sheep varieties may mature at a much younger age. Nigerian Dwarf

³⁹ <http://www.merckvetmanual.com/mvm/index.jsp>

⁴⁰ Freemartin -- reproductive tract hypoplasia (infantile uterus, not developing appropriately with growth of the rest of the calf, failure to respond to puberty).

⁴¹ A hermaphrodite is an animal with ambiguous genitalia, typically a penis with ovaries or a vulva with testicles. Sometimes this abnormality is not obvious.

⁴² Hypospadias is a condition where the urethra exits the penis on the ventral aspect of the glans penis and not at the tip of the penis where it is supposed to exit.

goats, such as those used in the Keefer et al. (2001a) study, mature as early as four months (Pugh 2002). Swine also mature sexually at a relatively young age, and gilts typically begin cycling between 6 and 8 months of age. Male animals generally reach sexual maturity at similar ages to females of the same breed and species.

In female animals of agricultural species, the estrous cycle is typically 21 days in length, although some variation exists among species. For example, the estrous cycle in sheep is only 17 days. In cattle, both males and females are fertile year round, although fertility may be decreased during parts of the year in regions with hot, humid climates. Sheep and goats originating in temperate zones are seasonal breeders, becoming fertile in response to decreasing day length. Breeds of sheep and goats that originated in the tropics are less sensitive to day length, and some are fertile year round. Swine, like cattle, are year-round breeders. A cow's gestation is approximately nine months, with some breeds having slightly shorter and others having slightly longer gestations. Sheep and goats have gestations lasting approximately five months, with less variation among breeds. In swine, gestation is approximately four months.

With the exception of parturition, the reproductive period is characterized as low risk for the general population of healthy, properly managed agricultural animals. By this point in the animals' growth the immune system is fully developed, and typically assisted by vaccination and parasite control practices. As previously noted, however, heifers are at greater risk of dystocia compared to older cows, largely because they are less than mature size at the time of their first calving. Although it is common practice to select sires with records of producing low birth weight calves ("calving ease"), dystocia continues to be a hazard for heifers. Dystocia is less of a concern in animals that typically bear multiple young, such as swine, as individual fetuses in multiple-fetus pregnancies are usually small compared to single births.

5. Post-Pubertal Maturation and Aging (Developmental Node 5)

Maturity and aging in food animal clones have not been studied extensively due to the relatively short time that cloning has been practiced. Common practice among conventional animals kept for breeding stock indicates that males may be kept to a later age than females, as they generally continue to be fertile for a longer period. Thus, highly valued males would continue in the herd as long as adequate quality semen was still being collected. When fertility of females declines, they are typically sold for slaughter, regardless of age. This decline in fertility generally occurs well before the animal shows other signs of aging or age-related disease.

a. Telomere Length as an Indicator of Aging

Studies have suggested that telomeres, long strands of repetitive DNA that “cap” the ends of chromosomes, are the “biological clock” that controls aging (Lanza et al. 2000, Betts et al. 2001). In all eukaryotic⁴³ cells, the terminal ends of chromosomes are capped by short, repetitive sequences of noncoding DNA that are repeated up to many kilobases in length, in conjunction with specific binding proteins. Telomeres play a role in chromosome stability, protecting DNA from digestion by exonucleases (enzymes that attack the ends of chromosomes), facilitating attachment of chromosome ends to the nuclear envelope, ensuring proper segregation of chromosomes during replication, and ensuring the full replication of coding DNA during cellular divisions (Kuhholzer-Cabot and Brem 2002).

Although the DNA in chromosomes is generally double stranded along its length, the end of the chromosome, or the telomere, differs in that it consists of a single-stranded overhang (called a lagging strand) of variable length that forms a loop. Conventional DNA polymerases (enzymes that replicate DNA) cannot replicate the extreme 5' ends of chromosomes. Instead, these lagging strands are replicated in a series of fragments, rather than as a continuous strand. Each fragment is “primed” by a short sequence of RNA and the gaps between fragments are filled in by DNA polymerase. However, when the RNA primer at the furthest end of the lagging strand is removed, a small gap of un-copied DNA is left that is not filled in by the DNA polymerase. This leads to the loss of 50 to 200 base pairs each time the cell divides. For this reason, telomeres have been proposed to act as “mitotic clocks” that limit the capacity of cells to replicate through the single stranded region, which is interpreted as a DNA damage signal. The net effect is that at some critical telomere length, cell cycle progression is halted, and the cell becomes “replicatively senescent” or incapable of further division. Senescent cells remain viable and metabolically active for very long periods of time with minimal cell death (Schaetzlein and Rudolph 2005).

Telomeres appear to be longest in the nuclei of early stage embryos, and begin to decrease in length starting in the embryonic period. Early stage embryos and immortalized cells in culture appear to have the capacity to rebuild telomeres through the action of an enzyme known as telomerase (Betts et al. 2001, Xu and Yang 2001). Telomerase, the enzyme responsible for telomere replication and elongation, is active during embryogenesis, suppressed postnatally in most somatic tissues, but remains active in germ cells, tumor cells, and in a subset of stem/progenitor cells (as reviewed by Xu and Yang 2003, Schaetzlein and Rudolph 2005). The activation of telomerase appears to occur about the time when the genome becomes activated in the embryo: at approximately the 2-cell stage in mice, or the 8 to 16 cell stage in cattle (Betts and King 2001). The ability of SCNT embryos to rebuild telomeres may depend on species, the

⁴³ In contrast to bacteria, which are classified as “prokaryotes” and have a simple structure, eukaryotic cells have a clearly defined nucleus containing true chromosomes surrounded by a membrane. Eukaryotes also contain other organelles such as mitochondria.

source of the donor nucleus, and culture conditions for early stage embryos (Betts and King 2001, Miyashita et al. 2002).

Concerns over genetic age and potential longevity of SCNT-derived animal clones were first raised after a report by Shiels and coworkers (1999) who noted that telomeres of “Dolly,” the first SCNT clone, were 10-20 percent shorter at one year of age than age-matched conventionally bred sheep (Shiels et al. 1999). Since that report, studies in animal clones have examined the effects of the nuclear transfer process on telomere length and telomerase activity to determine whether the SCNT process “resets” telomere length. Some early studies in cattle suggested that the SCNT process may influence cellular age and senescence. For example, Betts et al. (2001) noted reprogramming abnormalities affected telomerase activity in some early bovine SCNT embryos. In contrast, Cibelli et al. (1998a) cloned from a late-passage cell line (after 30 passages *in vitro*; the lifespan of cells *in vitro* is approximately 31-33 passages). At 40 days gestation, the fetus was harvested and a fibroblast cell line established. These fibroblasts appeared to have an extended lifespan compared to the original donor cells, and underwent another 31-33 passages *in vitro*.

Other studies suggest that reduction in telomere length may be more related to animal species, type of cells used to derive the donor cell line, or duration of time in culture (Shiels et al. 1999, Kuhholzer-Cabot and Brem 2002, Miyashita et al. 2002, Betts et al. 2005). Although telomere shortening may have led to a premature aging phenotype in telomerase-knockout mice (Blasco et al. 1997, Rudolph et al. 1999), convincing data on clones addressing the issue of premature aging are not currently available.

Telomere length variation has not been observed consistently across cloning studies or species. The group that produced “Dolly” stated that her telomeres were of the same length as the cultured mammary gland cells (from a six year old ewe) from which she was generated (Shiels et al. 1999). Dolly’s reduced telomere length was not associated with any other measurable signs of premature aging.⁴⁴ Betts et al. (2001) noted that SCNT sheep generated from cultured embryonic or fetal cells had telomeres 10 -15 percent shorter than age-matched controls. Studies in cattle clones indicated that telomere lengths differ among tissues within an animal, and that DNA from some tissues were more amenable to telomere rebuilding, while DNA of nuclei from other tissues yielded clones with substantially shorter telomeres. For example, Miyashita et al. (2002) have reported that although clones derived from epithelial cells of a 13-year-old cow and clones derived from the oviductal epithelial cells of a six-year-old cow had telomeres shorter than age-matched controls, clones derived from muscle cells of a 12-year-old bull were similar to age-matched controls. Similarly, Kato et al. (2000) noted that telomere lengths in ear fibroblasts of a

⁴⁴ <http://www.roslin.ac.uk/publicInterest/wasDollyOldAtBirth.php>

calf clone were similar to that of the 10-year-old nuclear donor bull, but telomeres in white blood cells of the same clone were similar to those of an age-matched control.

The telomere length of goat clones derived from fetal fibroblast donor cells were shorter than in those from age-matched control animals (Betts et al. 2005). These authors also noted that progeny from goat clones were found to have shorter telomere length in testicular biopsies compared to conventionally derived animals, and the telomere lengths were intermediate to the values obtained for their clone fathers' and age-matched control testes (Betts et al. 2005). This suggests that there was incomplete telomere elongation in the offspring of clones, although as mentioned above it is uncertain whether telomere length is a predictor of longevity.

By contrast, the telomere length of sheep clones (Clark et al. 2003) and cattle derived from adult or fetal fibroblasts were comparable to conventionally bred cattle (Tian et al. 2000, Betts et al. 2001, Jiang et al. 2004) or even slightly increased when near senescent bovine fibroblasts were used for cloning (Lanza et al. 2000).

Using a slightly different technique for measuring telomere length, Meerdo et al. (2005) found no significant difference between blastocysts derived from adult bovine fibroblast cell lines and *in vitro* fertilization-produced blastocysts, but the clone blastocysts had longer telomeres than the two donor cell lines. They also noted detectable telomerase activity in oocytes and a dramatic increase in telomerase activity at the morula stage. A second study in cattle and one in mice also demonstrated telomere elongation during the transition from morula to blastocyst in clone embryos (Schaetzlein et al. 2004). Cellular aging in tissue culture is also reflected by telomere shortening, and its reversal during SCNT was evident in a study by Clark and coworkers by the partial restoration of telomere length after nuclear transfer from late-passage cells (Clark et al. 2003). This and several other studies suggest that gametes have telomerase activity sufficient to lengthen the telomeres through the maturation process (Xu and Yang 2000, Betts et al. 2001, Meerdo et al. 2005).

Wakayama et al. (2000) evaluated successive generations of mouse clones for signs of premature aging and changes in telomere length in chromosomes from peripheral blood lymphocytes. Female mice were reiteratively recloned to six generations (*i.e.*, Mouse G1 was derived from a somatic cell, Mouse G2 was cloned from a cell from Mouse G1, etc. for 6 generations) and four generations in two independent lines. The mouse clones (n = 35) showed no physical signs of increased aging, and behaved normally relative to age-matched controls as measured by tests of learning ability, strength, and agility. There also was no evidence of shortening of telomeres, as had been reported in some studies of livestock clones. In contrast, telomere length increased with successive cloning, although this finding may be confounded by age-related contributions or by characteristics of the donor cells (the cumulus cells used to produce the clones were found to

express telomerase, suggesting that these cells may have long telomeres at the outset). They concluded that “*telomere shortening is not a necessary outcome of the cloning process,*” and suggested the possibility that the differences among the results observed in various species may be due to the selection of cells of longer or shorter telomere length in the different SCNT protocols. Clark et al. (2003) noted that fibroblast cell lines derived from fetal sheep clones had the same capacity to proliferate and the same rate of telomere shortening as the donor cell line from which the fetuses were cloned. This observation led King et al. (2006) to hypothesize that replicative senescence was under genetic control, and not triggered by a pre-determined telomere length.

Recently, Yonai et al. (2005) reported on the growth and production characteristics of six Holstein and four Jersey clones (described in detail in Chapter VI). These clones were derived from oviduct epithelial cells and had shorter telomeres than those observed in conventionally bred old cows (Miyashita et al. 2002). The overall success rate in terms of calf survival beyond the perinatal period was 4.8 percent for the Holstein group and 10.8 percent for the Jersey group. At the time of publication of their article all of these remaining clones had produced two calves and were artificially inseminated and had conceived for a third time. The authors concluded that “*reduced telomere length did not influence productivity between birth and 3 years of age.*”

Thus, although there have been reports of different telomere length outcomes in clones, at this time it is not possible to determine what the exact mechanism for telomere shortening is in clones, as studies have demonstrated that clones do have sufficient telomerase activity to return the shorter telomere lengths of the donor cells to lengths appropriate for normally developing embryos. Further, although some studies indicate that clones have shorter telomere lengths than would be expected, other clones have age-appropriate telomere lengths, and some appear to have longer telomeres. The most detailed study of clones with shortened telomeres indicates that the animals appear to be healthy and function normally. Finally, at this time, because most clones have not been alive for the full “natural” lifespan of their species, it is not possible to predict whether clones with shortened telomeres will exhibit premature aging.

C. Data on Animal Health by Species

1. Cattle

As mentioned above, the majority of available data on health of animal clones and their surrogate dams are derived from studies in cattle. Survival of live-born bovine clones from various studies is summarized in Table V-1. Because relatively few studies included contemporary comparators, historical data from various references and data bases were also incorporated into the table to provide context. For additional information on health and survival of calves derived by other ARTs, see Appendix C.

Table V-1: Survival Rates of Live-Born Bovine Clones and Comparators				
Reference	Transgenic Status (proportion of animals in study)	Surviving/Total Live-Born Clones (fraction)¹	Surviving /Total Live- Born Comparators (fraction)	Comments
Batchelder 2005	None	2/8 (0.25)	6/6 ET 3/3 AI (1.00)	
Chavatte-Palmer et al. 2002	None	21/21 (1.00)	20/20 IVF 176/176 AI (1.00)	Described in Chapter VI
Chavatte-Palmer et al. 2004	None	36/58 (0.62)	NP	Update on animals generated since 1998, includes some animals from the 2002 publication
Cyagra 2003: Appendix E	None	104/134 (0.78)	NP ²	Data from complete comparator birth cohort (animals surviving vs. animals born) not available
Edwards (unpublished)	None	12/27 (0.44)	NP	See Appendix G for complete data
Gibbons et al. 2002	None	8/9 (0.89)	NP	
Gong et al. 2004b	None	12/27 (0.44)	NP	
Green et al. 2007	None	9/9 (1.00)	NP	Used differentiated muscle cells
Heyman et al. 2002	None	11/15 (0.73)	20/25 (0.80)	IVF derived contemporary comparators
Heyman et al. 2004	None	35/50 (0.70)	65/68 (0.93)	AI derived contemporary comparators
Hill et al. 1999	All	6/8 (0.75)	NP	
Hill et al. 2000a, 2001a	All	1/2 (0.50)	NP	
Ideta et al. 2005	None	0/1 (0.00)	NP	
Kato et al. (1998, 2000)	None	13/24 (0.54)	NP	An additional clone died between the perinatal period and 117 days of age (12/24 or 0.50 overall survival)

Table V-1: Survival Rates of Live-Born Bovine Clones and Comparators				
Reference	Transgenic Status (proportion of animals in study)	Surviving/Total Live-Born Clones (fraction)¹	Surviving /Total Live- Born Comparators (fraction)	Comments
Kishi et al. 2000	None	3/4 (0.75)	NP	
Kubota et al. 2000	None	4/6 (0.67)	NP	
Lacham-Kaplan et al. 2000	None	2/2 (1.00)	NP	
Lanza et al. 2000	All	6/6 (1.00)	5/5 (1.00)	IVF and ET derived comparators
Lanza et al. 2001	All	24/30 (0.80)	NP	
Lawrence et al. 2005	None	1/3 (0.33)	NP	The surviving calf died at 9 months due to clostridial infection (subcohort of Edwards above)
Matsuzaki and Shiga 2002	None	8/13 (0.62)	7/7 (1.00)	IVF and AI derived comparators
Meirelles et al. 2001	None	1/1 (1.00)	NP	
Mello et al. 2003	None	1/1 (1.00)	NP	
Pace et al. 2002	Some	82/106 (0.78)	NP	
Panarace et al. 2007	None	225/317 (0.71)	NP	Results reported for Cyagra operations in U.S., Argentina, and Brazil from 2000 to 2005; includes data from Cyagra 2003: Appendix E
Powell et al. 2004	All	5/8 (0.63)	NP	
Renard et al. 1999	None	0/1 (0.00)	NP	Case study on a clone of clone
Shiga et al. 2005	None	4/8 (0.50)	NP	One death associated with Akabane virus
Schurmann et al. 2006	None	6/9 (0.67) IVF-NT 3/4 (0.75) NT	8/8 (1.00)	AI was used as a negative control for this study
Urakawa et al. 2004	None	8/9 (0.89)	NP	
Wells et al. 2004	None	104/133 (0.78)	37/52 (0.71)	Table reflects survival to 3 months, due to unexplained

Table V-1: Survival Rates of Live-Born Bovine Clones and Comparators				
Reference	Transgenic Status (proportion of animals in study)	Surviving/Total Live-Born Clones (fraction)¹	Surviving /Total Live- Born Comparators (fraction)	Comments
				differences in numbers at the beginning of later periods. Reports on number of calves delivered; unclear how many were stillborn. Comparators are progeny of clones
Wells et al. 2003b	Some	22/31 (0.71) 11/24 (0.46)	NP	Non-transgenic (31 calves born alive) and transgenic (24 calves born alive) listed separately
Zakharichenko et al. 1999b	None	1/2 (0.50)	NP	
Hasler et al. 1995	NA	NA	361/428 (0.84)	Historical data on IVF derived beef calves in a commercial operation
Lombard et al. 2007	NA	NA	0.91	Survey of three large dairies in Colorado; based on calves alive 24 hrs after birth
Nix et al. 1998	NA	NA	0.96	Historical comparison from a university herd of beef cattle using AI
Schmidt et al. 1996	NA	NA	13/18 (0.72)	Calves produced by IVF. Two embryos transferred to each recipient, yielding 11 live-born twins and 7 singles; 4 twins dead by 14 days. One singleton dead by 14 days.
USDA/NAHMS 1997 (12/96 – 2/97) ⁶	NA6	NA	0.97	Historical data from beef cattle produced through AI and natural mating in commercial operations

Table V-1: Survival Rates of Live-Born Bovine Clones and Comparators				
Reference	Transgenic Status (proportion of animals in study)	Surviving/Total Live-Born Clones (fraction)¹	Surviving /Total Live- Born Comparators (fraction)	Comments
USDA/NAHMS 2002 (1/02 – 12/02) ⁷	NA	NA	0.98	Historical data from dairy cattle produced through AI and natural mating in commercial operations
Xu et al. 2006	NA	NA	457/458 (1.00)	Used sex-sorted, vitrified, IVF embryos

¹ Survivors through the Juvenile Period/Live births
² NP = not provided; data not available
³ Beef calves; ⁴ Dairy heifers
⁵ NA = not applicable
⁶ <http://nahms.aphis.usda.gov/>
⁷ Ibid

Transgenic Status: All = All of the clones cited in the publication are derived from transgenic donor cells, Some = Some of the clones cited in the publication are derived from transgenic donor cells, None = None of the clones cited in the publication were derived from transgenic donor cells.
 IVF = *in vitro* fertilization
 AI = artificial insemination
 ET = embryo transfer

a. Developmental Node 1: Pregnancy and Parturition

i. Pregnancy

Most abortions in natural service and AI pregnancies in cattle remain undiagnosed due to the expense of laboratory work and the low profit margin in both the beef and dairy industry. Producers and veterinarians become concerned when the rate of abortion exceeds 3-5 percent in a herd. Many causative factors, both infectious (*e.g.*, bacterial, protozoal, viral, fungal) and non-infectious (*e.g.*, genetics, nutrition, stress, toxicity), have been identified (Merck Veterinary Manual Online 2005⁴⁵). Fetal losses later in pregnancy may be more common in goats and swine compared to cattle (Engeland et al. 1997, van der Lende and van Rens 2003, Vonnahme et al. 2002), and are not necessarily associated with disease (Engeland et al. 1997).

Farin et al. (2001) stated that up to 40 percent of pregnancy losses in cattle occur between days 8 and 18 of gestation. A recent study (Silke et al. 2002) indicated that most pregnancies are lost

⁴⁵ <http://www.merckvetmanual.com/mvm/index.jsp>

during the same period in dairy cattle, while a smaller percentage of pregnancies are lost between days 16 and 42 of pregnancy (late embryonic period). Total pregnancy loss in moderate to high yielding dairy cattle may be as high as 40 percent (Silke et al. 2002). Losses at later stages of pregnancy in cattle bred by AI are estimated to be less than 5 percent (Thompson et al. 1998). A study of beef heifers indicated that losses in the first days following embryo transfer are the most common (Dunne et al. 2000), with similar pregnancy rates at days 14 and 30 days of pregnancy, and at term (68 percent, 76 percent, and 71.8 percent, respectively). These early losses do not pose a hazard to the surrogate dam, and the net result is typically a longer than normal estrous cycle (Merck Veterinary Manual Online 2005⁴⁶).

Early embryo loss in other forms of ART may be related to *in vitro* culture conditions that may cause abnormal development and early embryo/fetal death. In a review of studies of *in vitro* produced (IVP) and clone bovine embryos, Farin et al. (2004) reported lowered pregnancy rates and increased rates of abortion associated with IVP.

Similar to other ART, by far the greatest loss of pregnancies resulting from SCNT embryos occurs prior to 60 days gestation in cattle (Le Bouhris et al. 1998, Hill et al. 1999 with transgenic clones, Kishi et al. 2000, Lanza et al. 2000 with transgenic clones, Chavatte-Palmer et al. 2002, Pace et al. 2002 using mixed transgenic and non-transgenic clones). High pregnancy losses during the time of placental formation suggest that embryonic death may be a consequence of faulty placentation, possibly due to a delay in chorioallantoic development, as proposed by Hill et al. (2000b) and Bertolini et al. (2004). Abnormal placentation may lead to a build up of wastes in the fetus and associated membranes, or inadequate transfer of nutrients and oxygen from the dam to the fetus.

Unlike AI and ET, IVP and SCNT pregnancy losses occur at all stages of gestation in cattle, although later term spontaneous abortion is more common in SCNT than IVP (Farin et al. 2006). Clone pregnancies have been lost during the second and third trimesters and have been accompanied by reports of hydrops (discussed in more detail in section 1.a.ii.), enlarged umbilicus, and abnormal placentae (Batchelder, 2005). Indeed, a major factor contributing to mid- and late-term spontaneous abortion of clones of both embryonic and somatic cell origin is abnormal development of the placenta (Wells et al. 1999, Farin et al. 2001, Chavatte-Palmer et al. 2002). Normal placental development is essential to ensure proper exchange of nutrients and gases between mother and fetus (Farin et al. 2001, Bertolini et al. 2004). Placental insufficiency has been cited as a possible cause of fetal loss in cattle, goats and swine bred by AI or natural mating (Lucy 2001, Engeland et al. 1997, Vonnahme et al. 2002). Studies have reported too few and/or abnormal cotyledons present in the placentae of sheep and cattle clones (Farin et al. 2001,

⁴⁶ Ibid

Chavatte-Palmer et al. 2002, Heyman et al. 2002a, Lee RS et al. 2004, Batchelder 2005). Although fewer in number, these abnormal placentomes are found to be larger, weigh more, and comprise a greater surface area for exchange than “normal” placentomes. Enlarged placental surface area in IVP suggests an increase in substrate uptake and transport capacity (Bertolini et al. 2004).

Lee RS et al. (2004) noted pregnancy rates were similar between NT, AI, and IVP at 50 days gestation (65 vs. 67 and 58 percent, respectively), but from that point onward NT pregnancies were continually lost. By day 150, only 40 percent of NT embryo recipients were still pregnant. There were no losses during this time period for either AI or IVP pregnancies. Mean fetal weights at 100 days gestation were not different between the three groups; however, the authors noted that more NT fetuses were two standard deviations above the mean weight of AI fetuses (283 ± 2 g) compared to IVP fetuses (5/6 vs. 1/4). A similar trend was noted among fetuses examined at day 150. Fetal livers and kidneys were larger among NT fetuses compared to AI or IVP fetuses, and one liver and the kidneys from three NT fetuses were noted to have fatty infiltrations. Fatty liver was also diagnosed on post-mortem of one neonatal calf in the recent study by Chavatte-Palmer et al. (2004).

(a). Placental development

Few detailed descriptions of placentae of cattle clones exist. Lee RS et al. (2004) examined placentae of developing SCNT fetuses at 50, 100 and 150 days of gestation. These time periods roughly correspond to the periods before placentome formation is complete (50 days), shortly after complete placentome formation (100 days), and the period when hydrops may first be detected (150 days). The authors noted that at day 50, fetal cotyledon formation and vascularization initiated normally in NT fetuses, but fewer cotyledons successfully formed placentomes compared to AI and IVP control pregnancies. At day 50, 5/10 NT fetuses were noted to have very good vascularization of the cotyledons, compared to 2/5 AI and none of the IVP fetuses, which were said to have pale cotyledons. However, at day 100, the mean number of caruncles among NT pregnancies was lower than for either AI or IVP groups (58 ± 9 vs. 103 ± 15 and 99 ± 16 , respectively). Although numbers of cotyledons were reduced in the NT group, total weight of caruncles was significantly higher in NT fetuses compared to the other groups at day 100, suggesting an attempt to compensate for lower numbers. The authors described NT placentomes as larger than AI or IVP placentomes, and having thicker, fist-shaped structures compared to AI or IVP placentomes, which were typically flat and discoid in shape.

Batchelder (2005) conducted a systematic histological exam of placentae collected at birth from seven cattle clones. She noted all clone placentae exhibited one or more abnormalities of varying severity: moderate to severe edema, enlarged vessels, adventitious placentation, and large areas

devoid of placentomes. No abnormalities were noted for the comparator placentae collected (n=9). In general, clones had fewer (67.4 vs. 98.3) and larger placentomes (6.05 vs. 3.84 kg) compared to the pooled means for AI and ET comparators, and surface area of placentomes was greater and more variable in placentae of clones vs. comparators. The placenta of one clone contained two masses comprised of fatty and connective tissue with hair, but exhibiting no bone or organ development. These may have derived from embryos that failed to undergo complete differentiation, likely due to failure to completely reprogram the donor nucleus to a totipotent (able to become any tissue type) state (See Chapter IV). These may pose a potential hazard (metritis) to the dam if the fetal membranes are not completely expelled at termination of the pregnancy. In this study, all clones were delivered by planned C-section, and the placentae were manually removed.

Constant et al. (2006) noted that placental overgrowth preceded fetal overgrowth in bovine SCNT pregnancies with developing hydrallantois, and that the fetal to placental weight in SCNT pregnancies was lower than for AI or IVF pregnancies after day 220 of gestation. Constant et al. selected 18 SCNT pregnant surrogate cows demonstrating signs of hydroallantois for their study, and compared them with 10 normal AI and six normal IVF pregnancies. Pregnant cows were slaughtered between 180 and 280 days gestation, and fetuses and placentae removed and weighed. Recovered placentae were examined macroscopically and histologically. Differences between SCNT and control pregnancies were noted mainly in placentae and fetal tissues after gestation day 220 in this study. Although placentome number remained constant in AI and IVF pregnancies, placentome number in SCNT pregnancies tended to increase with gestational age. Within the SCNT group, Constant et al. identified two distinct populations: one in which placentome weight and number were within the range considered normal; and one group in which mean placentome weight was heavier than expected given the number of placentomes. Within placentomes from SCNT placentae, histologic examination revealed flattened uterine epithelia with small nuclei, rather than the expected raised caruncular tissue. Fetal connective tissue (the cotyledons) was enlarged even without evidence of edema, with reduced cell density and vessel dilation in some SCNT placentomes. Where edema was observed, it occurred around, but not within, placentomes. In this study, placental abnormalities and hydroallantois were linked to gestational age; however, there did not appear to be a direct relationship between hydrallantois and other placental abnormalities. It is important to note that only placentae and fetuses from pregnancies demonstrating hydrallantois were used in this study. Tissues from apparently normal SCNT pregnancies were not examined.

Failure of epigenetic reprogramming has been cited in numerous studies as a likely cause of early embryo failure and abnormal placental development for SCNT (see Chapter IV). Recent collaborations across laboratories suggest that improper epigenetic reprogramming may also lead to immune-mediated rejection of the developing SCNT fetus during early pregnancy (Hill et al.

2002, Davies et al. 2004). In conventional bovine pregnancies, placental trophoblast cells (cells that interface with the uterus) express antigenic proteins, referred to as major histocompatibility complex (MHC) class I, during the third trimester of pregnancy, and may be involved in the process that leads to the normal expulsion of the placenta following birth of the calf. Work by Davies et al. (2004) indicated that the trophoblast cells of SCNT embryos expressed MHC class I proteins during the first trimester, between 34 and 63 days gestation. These workers also noted accumulation of lymphocytes within the uterine lining, indicating an inflammatory response, in pregnancies where embryos that were immunologically incompatible with their surrogate dams were aborted. Conversely, embryos that were compatible with their surrogates' immune systems survived to term.

Hashizume et al. (2002) noted differences in the expression of placental lactogen (PL)⁴⁷ and pregnancy-associated glycoprotein (PAG)⁴⁸ between Japanese Black cows bred by AI vs. cows carrying SCNT pregnancies at 60 days of gestation. Pregnancy was diagnosed by ultrasound between days 30 and 60 of gestation, and fetal viability confirmed by presence of a heartbeat. Confirmed pregnant cows were slaughtered around day 60 of gestation, and uteri and fetal tissues removed for examination. The differences in gene expression were most notable among cows with immotile NT fetuses, which had low or non-existent expression of these pregnancy related genes. It is likely that these pregnancies would have failed spontaneously if allowed to proceed. These SCNT placentae also exhibited poor caruncular development as well as cotyledonary development with respect to both shape and number of these structures compared to placentae from AI derived pregnancies. In addition, the cotyledonary area was pale and flattened in SCNT placentae compared to AI placentae, which had the expected thickened cotyledons with well developed villi (finger-like structures which interlock and increase the surface area across which fluids and gases flow). The number of SCNT cotyledons was approximately half that of AI placentae in the Hashizume et al. study.

Similarly, Chavatte-Palmer et al. (2006) noted reduced fetal crown-rump length and placentome width in clones compared to AI and IVP controls at all stages of pregnancy. Concentrations of PAG were significantly lower in clones with early pregnancy loss (between days 35 and 90 of gestation) compared to clones with pregnancies that survived to a later date, although there was no difference in PAG levels between clones that died later in gestation versus those that were born alive.

⁴⁷ Placental lactogen is part of the growth hormone family, and has a number of roles during pregnancy which vary among species. Among its functions are stimulation of placental vascular growth (angiogenesis), maternal and fetal intermediate metabolism, and mammary gland development.

⁴⁸ Pregnancy-associated glycoproteins (PAG) are produced by the bovine placenta. The physiological roles of these peptide hormones are unknown. Functions that have been suggested include modulation of uterine prostaglandin secretion and promotion of the immunological tolerance of the developing embryo/fetus by the maternal immune system.

In contrast to the Hashizume et al. study, Bertolini et al. (2006) found increased levels of PL and pregnancy-specific protein B (homologous with PAG-1) in placentomes from IVP fetuses compared to *in vivo* ET fetuses in pregnancies terminated at 180 days gestation. Interestingly, for the production of IVP embryos, Bertolini et al. utilized a culture system intended to induce LOS pregnancies, which contained 10 percent fetal calf serum and a mixture of follicle-stimulating hormone, luteinizing hormone, and estradiol 17 β .

The underlying cause(s) of the higher rate of pregnancy failure and placental abnormalities in SCNT compared to IVP may be related to a number of factors or combination of factors, including the culture media (Thompson 1997; van Wagendonk-de Leeuw 2000; Gardner and Lane 2005), selection of the oocyte (Greeve and Calleson 2005, Roberts et al. 2006), and selection of the donor cell for nuclear transfer (Wells et al. 2003b, Ideta et al. 2005, Lawrence et al. 2005). For both IVP and SCNT, oocytes are generally subjected to a period of maturation *in vitro*, which by-passes the natural developmental processes *in vivo*⁴⁹, and may result in the selection of oocytes that are not capable of sustaining normal embryo/fetal development (Greve and Callesen 2005). Even for non-nuclear transfer embryos, Farin et al. (2004) noted substantial differences in developmental competence of *in vivo* vs. *in vitro* matured oocytes, with blastocyst formation ranging from 50 to 80 percent for *in vivo* matured versus 15 to 40 percent for *in vitro* matured oocytes.

(b). Cell selection and cell culture

Wells et al. (2003b) noted that survival rates to term differed depending on cell cycle of the nuclear donor cells. Putative G₀ cells (cells that apparently were not dividing) used for nuclear transfer had high early pregnancy losses, but no losses after 120 days of gestation, and no reported hydrops. Cells that had begun to divide (G₁ phase) had higher losses to term (21/43 pregnancies lost after 120 days gestation) and higher incidence of hydrops (18/43 (42 percent) of pregnancies), but higher post natal survival than clones from G₀ cells.

Similar to Wells et al. (2003b) findings, Lawrence et al. (2005), found that when somatic cells were “starved” into quiescence by serum deprivation prior to nuclear transfer, more surrogate heifers were confirmed pregnant compared to heifers that received embryos from the serum-supplemented group. However, in contrast with the findings of Wells et al., embryo loss in the Lawrence et al. study was higher in the serum-starved group compared to the serum-supplemented group between 25 and 50 days post-transfer of embryos. Eight of nine pregnancies

⁴⁹ In cattle, a large number of follicles begin to develop during the estrous cycle, but only one (or occasionally two) will be selected to ovulate. The remaining follicles and the oocytes in the ovaries undergo atresia (degeneration) and senescence. The factors that lead to the selection of the ovulatory follicle are not yet completely understood.

in the serum-starved group failed during this early period of development, with the ninth pregnancy failing later in gestation. Although early pregnancy losses were lower in the serum-fed group, (4/11 embryos failed during the period up to 50 days post transfer), later gestational losses beginning on day 174 of gestation resulted in only one viable Jersey calf from this group. Six of the remaining pregnancies resulted in hydroallantois, with fetal deaths occurring between 174 and 255 days. The surviving clone was reported as healthy until nine months of age; two pregnancies were terminated by emergency C-section at 260 and 271 days, but these calves did not survive beyond one week of age. The surviving calf died at nine months of age (see Section 2.c.i).

Fetal muscle cells were serum-starved for one, four, or five days prior to NT to induce different stages of differentiation in a recent study by Green et al. (2007). Three muscle cell types developed *in vitro* were frozen, thawed, and serum-starved for an additional five days prior to NT. Nine apparently healthy calves were born: four from myogenic precursor cells, two from myotubes, and three from muscle fibroblasts. All calves were euthanized prior to weaning, and no additional data is provided on them. The reason for euthanasia was not provided. The authors only state that the calves were euthanized for “unrelated reasons.”

In contrast to the Wells et al. study, Urakawa et al. (2004) reported success using fetal fibroblast donor cells in the G₁ phase. Two cell lines were used, derived from fetuses with the same dam but two different bulls. All embryos that survived to ≥ 6 cells (day 3) continued to develop to the morula/blastocyst stage by day 6. Ten of these blastocysts were transferred into ten recipients, resulting in nine live calves. According to the authors, calving was “uneventful.” Differences were noted between cell lines, in that three calves resulting from one of the lines tended to be heavier at birth than the six calves of the other cell line used (actual birth weights not provided). One of these three heavy-weight calves died after two days without standing. The authors do not report on the health or survival of the remaining eight calves beyond the first six days of life.

Similarly, Ideta et al. (2005) compared development of embryos constructed with G₁ or M phase (the period in the cell cycle when cell division takes place) fetal fibroblasts, and noted that G₁ SCNT embryos had higher rates of development to blastocyst than M phase cells (31 vs. 16 percent). Although these results are considerably lower than those noted in the Urakawa et al. study, the numbers are calculated based on total number of embryos cultured prior to first cleavage, whereas the Urakawa et al. study calculated development based on embryos surviving the first three days in culture. Only five surrogate cows received embryos in the Ideta et al. study, of which three were diagnosed pregnant on day 30 of gestation, and one live calf was delivered. All of the transferred embryos were developed from G₁-phase somatic cells. The single calf died two days after birth. Health of the surrogate dams, method of delivery, and birth weight of the single calf was not reported in this study.

Another study (Schurmann et al. 2006) investigated whether enucleation after IVF would enhance the survival of SCNT derived embryos. In this study, male and female pronuclei and the *zona pelucida* of the single-cell zygote were removed approximately four hours after IVF, and nuclear transfer performed using adult ear skin fibroblasts. *In vitro* development to blastocyst was similar between the controls, derived from unfertilized oocytes, and the early zygote group. Post-implantation development was higher in the group derived from early zygotes or “sperm activated” embryos. More transferred blastocysts survived to establish pregnancies at 35 days gestation in the sperm-activated group compared to controls (30/49 vs. 17/41), and numerically more pregnancies resulted in live calves at birth in the sperm activated group (9/30 vs. 4/17), although this difference was not significant. Survival of calves to weaning was low for both groups, and did not appear to be different between groups (6/9 vs. 3/4).

ii. Parturition

(a) Hydrops

The set of conditions generally termed hydrops refers to abnormal fluid accumulation (edema) in one or more compartments of the placenta and/or the fetus itself, and are variously referred to as hydroallantois, hydrallantois, hydramnios or hydrops fetalis, depending on where the edema occurs (Heyman et al. 2002a,b, Merck Veterinary Manual Online 2005⁵⁰, Pace et al. 2002 (including transgenic clones)). Hydrops is estimated to occur in 1 in 7,500 pregnancies in the general population of cattle (Hasler et al. 1995). The incidence is higher in cattle and sheep recipients of IVP embryos, with one study estimating a rate of approximately 1 in 200 in IVP pregnancies in cattle, or 0.05 percent (Hasler et al. 1995). However, a more recent but smaller study (Block et al. 2003) reported 1/28 (four percent) IVP pregnancies resulted in hydrops. Reports of hydrops in SCNT pregnancies are highly variable, and range from 13 to 40 percent (Table V-2). One research center (AgResearch, NZ), reported that on average 42 percent of SCNT-bearing surrogate cows pregnant at 120 days gestation will experience pregnancy failure, and that 58 percent of these mid- to late-term failures may be attributed to hydrops. This indicates an average hydrops incidence of 24 percent for this laboratory (Forsyth and Wells 2006).

Table V-2 presents a summary of reports of hydrops in cattle from the peer-reviewed literature for clone, IVP, ET, and AI pregnancies. Survival rates of dams developing hydrops generally were not reported. Most studies that discussed outcomes indicated that dams developing hydrops were euthanized.

⁵⁰ <http://www.merckvetmanual.com/mvm/index.jsp>

Table V-2: Incidence of Hydrops in Cattle Surrogate Dams				
Study	Transgenic Status	Incidence (fraction) in clone pregnancies	Incidence (fraction) in comparator pregnancies	Comments
Batchelder 2005	None	1/8 (0.13)	0/6 (0.00)	Comparators were ET (n=6) and AI (n=3)
Block et al. 2003	None	NA	1/28 (0.04)	Study used frozen/thawed IVP embryos
Edwards (unpublished)	None	27/46 (0.59)	NP	
Hasler et al. 1995	NA	NA	1/200 (0.005)	Study based on commercial IVP operation
Heyman et al. 2002a,b	None	3/20 (0.15) 5/21 (0.24)	0/24 (0.00)	IVP comparators
Hill et al. 1999	All	2/8 (0.25)	NP	
Lawrence et al. 2005	None	6/11 (0.55)	NP	Sub-cohort of Edwards
Lee RS et al. 2004	None	2/8 (0.25)	0/9 (0.00)	4 IVP and 5 AI comparators. A third clone fetus was suspected of developing hydrops. All pregnancies terminated at gd 150.
Matsuzaki and Shiga 2002	None	2/13 (0.15)	0/7 (0.00)	2 IVP and 5 AI comparators
Mello et al. 2003	None	1/3 (0.33)	NP	
Pace et al. 2002	Some	30/178 (0.17)	NP	Pregnancies lasting beyond 60 days
Panarace et al. 2007	None	24/37 (0.65)	0/13 (0.00) 0/10 (0.00)	13 ET and 10 IVP comparator pregnancies
Wells et al. 2003b	Some	18/43 (0.42) 1/6 (0.17)	NP	Pregnancies lasting beyond 120 days. Non-transgenic (n=43) and transgenic (n=6) listed separately
Zakharichenko et al. 1999a	None	2/5 (0.40)	NP	
NA = not applicable NP = not provided; data not available Gd = gestation day or day of pregnancy				

Not all cases of hydrops in clone-bearing pregnancies develop into a significant complication or threat. In an interview with CVM staff (see CVM Memorandum I at http://www.fda.gov/cvm/CloningRA_Memorandum_I.htm), U.S. clone producers indicated that many pregnancies result in some excess fluid accumulation in the fetal membranes and tissues.

In most cases this accumulation is mild or moderate, and does not threaten the surrogate dam or calf. The producers interviewed for this assessment indicated that they monitor surrogate dams closely, beginning as early as 150 days of gestation, for any signs of developing hydrops. They indicated that if the veterinarian determines that hydrops is sufficiently severe to threaten the surrogate, the pregnancy is terminated. In contrast, Forsyth and Wells (2006) indicated that in their experience at AgResearch in New Zealand, only the mildest cases of hydrops result in normal calves, and it is now their practice to terminate any pregnancy as soon as hydrops is diagnosed.

A few studies have directly compared cloning procedures with other ART under the same conditions. These studies are limited, with few clones and often fewer comparators from alternative technologies (Heyman et al. 2002a,b, Matsuzaki and Shiga 2002, Lee RS et al. 2004, Batchelder 2005). In one such study, Matsuzaki and Shiga (2002) compared 13 SCNT clones with five AI and two IVP-derived calves used as controls. Five of the 13 clones required delivery by Caesarian section (C-section), while all seven controls were delivered without assistance. Two cows carrying clones had to be induced at 250 days gestation due to rapidly expanding hydroallantois, and the calves were delivered by C-section.

Batchelder (2005) indicated that the largest clone in that study (weighing 71.0 kg at birth) exhibited edema at birth, particularly in the head and neck, suggesting that it suffered from mild hydrops fetalis. This calf was successfully delivered at term by planned C-section, although it died three days after birth. This calf's surrogate dam apparently was unharmed by the complication, although another surrogate dam was euthanized at 211 days gestation due to severe hydrops.

In one of the largest cattle cloning studies reported, Pace et al. (2002) estimated that approximately 6 percent (30/535) of all pregnancies established with SCNT embryos resulted in hydrops, but among pregnancies with clones that lasted beyond 60 days, the incidence of hydrops was 17 percent (30/178). An important consideration in interpreting these outcomes, however, is that approximately 75 percent of the embryo clones in this study were transgenic. Heyman et al. (2002) observed that 3 of 20 (15 percent) recipients of fetal and adult SCNT embryos (non-transgenic) developed severe hydroallantois during the time from approximately six months of gestation to term. In another trial reported in the same paper, five cases of late abnormal pregnancies were detected among 21 SCNT recipients (24 percent) by repeated ultrasonography, and the recipients were euthanized between day 155 and 233 of gestation. Severe hydroallantois was confirmed at necropsy and the size of the placentomes from these pregnancies was measured (142.3 ± 61.7 g vs. 46.7 ± 22.7 g for controls). No abnormalities were reported among the IVF-derived pregnancies in the Heyman et al. 2002 study.

Similarly, a study by Wells et al. (2003b) reported a high rate of pregnancy loss of non-transgenic bovine fetal fibroblast clones after 120 days gestation, with hydrops cited as the cause of pregnancy loss in 86 percent (18/21 losses) of the cases.

Panarace et al. (2007) monitored 37 clone-bearing, 13 ET, and 10 IVP pregnancies by ultrasound, and reported a high incidence of hydrops fetalis (13/37 pregnancies), placental edema (21/37 pregnancies), and hydroallantois (5/37 pregnancies) among clone-bearing pregnancies. Most of the hydrops fetalis cases (12/13) also developed placental edema. Most of these complications resulted in loss of the pregnancy. Only one clone experiencing hydrops fetalis and three clones with placental edema were born alive in this study; only two of these four calves survived. The authors do not report the outcome for the surrogate dams. None of the ET or IVP comparators developed hydrops in this study.

Lee RS et al. (2004) examined survival and development of AI, IVP and SCNT fetuses at 50, 100 and 150 days of gestation. Although there were no significant differences in fluid volume of fetal membranes at day 50 or 100, total fetal membrane fluid volume was significantly higher in SCNT (n = 8) fetuses compared to IVP (n = 4) fetuses (8033 ± 1800 ml vs. 5088 ± 698 ml) at 150 days gestation. For AI fetuses, mean fetal membrane fluid volume was 6500 ± 444 ml. The study noted the high variability in membrane weights and fluid volume among clone fetuses, and stated that 2/8 SCNT fetuses examined had particularly high allantoic fluid volumes (20 and 12 L), which were largely responsible for the high mean fluid volume among clones. The authors stated that these two cases indicated developing hydrops. The authors suspected a third SCNT fetus was developing hydrops, but did not provide data on this case. Fluid volumes were less variable among membranes of AI and IVF fetuses.

In contrast, hydrops has only been detected in one or two cows out of 250 to 300 transgenic clone-bearing surrogate cows, as reported in discussions with U.S. clone producers, suggesting that these results vary considerably among labs performing animal cloning (see CVM Memorandum I at http://www.fda.gov/cvm/CloningRA_Memorandum_I.htm). The producers also noted that hydrops occurred in IVP-derived pregnancies, but less frequently than with clone-bearing pregnancies, although no actual numbers were available.

The causes of hydrops in conventional animals are unclear. Liu and Wintour (2005) stated that hydramnios can result from abnormalities in fetal renal function, and that the large volume of dilute urine produced by the fetal kidney is essential for the maintenance of normal amniotic and, in some species, allantoic fluid volume. Conversely, Constant et al. (2006) concluded fetal abnormalities noted in NT pregnancies developing hydrops were the consequence of placental dysfunction. Although actual cause and effect is not yet clear, it appears that placental function is integral to the development of LOS and its related clinical signs, particularly hydrops and

congenital kidney defects. Lee RS et al. (2004) also suggested that the association between excessive fetal fluid accumulation and renal and placental growth deregulation may indicate impairment of renal and placental function. *“Although the placenta is the major organ regulating the fetal environment, the fetal kidney also plays an important role in the regulation of fetal arterial pressure, fluid and electrolyte homeostasis, acid base balance, and hormone synthesis. In ruminants, fetal urine contributes to the allantoic and amniotic fluid. Reports have appeared of kidney defects and impaired renal function in cloned offspring as well as impaired liver function in cloned mice...”*

Although it is likely related to placental insufficiency, not all abnormal placentae develop hydrops (Hill et al. 2001b). In SCNT, incomplete or improper epigenetic reprogramming and subsequent inappropriate gene expression may be an important factor in placental development and hydrops (see Chapter IV).

(b) Dystocia

Dystocia, or difficult labor, is an identified hazard for any pregnancy that goes to term. A common cause of dystocia is incompatibility between the size of the fetus and the pelvic opening through which it must pass. Although oversized offspring occur in all species, it is more common in animals that typically produce only one or two offspring per pregnancy. Other causes of parturition difficulty include malpresentation of an individual fetus (*e.g.*, breech birth, head or leg out of position), or simultaneous presentation of multiple fetuses in the birth canal. Severe dystocia may increase the risk of retained fetal membranes and metritis (uterine infection), and cause damage to the reproductive tract, including uterine adhesions, uterine rupture and uterine prolapse, and nerve and musculo-skeletal damage (Merck Veterinary Manual Online 2005⁵¹). Such complications could compromise future reproductive capability and result in culling of the dam. Another risk is that dystocia may lead to an emergency C-section. Complications of emergency C-section surgery may include uterine tearing, peritonitis, infected suture line, incisional hernia, and respiratory and circulatory compromise from anesthesia and recumbancy. Stress of labor is also a complicating factor in the case of emergency C-section.

Previous estimates of dystocia in natural and AI-derived bovine pregnancies range between 4 and 6 percent. Nix et al. (1998), in a large study of 2,191 births of natural and AI bred beef cattle at Clemson University reported that 6 percent of births required assistance. Calf birth weight and parity of dam (number of times she had given birth) were the major factors in the incidence of dystocia. Calves heavier than 40 kg were associated with greater calving difficulty. Heifers were more likely to experience dystocia, despite the common practice of selecting sires known to produce smaller calves. Dystocia contributed to the increased neonatal mortality of the calves

⁵¹ <http://www.merckvetmanual.com/mvm/index.jsp>

and decreased reproductive performance of the dams in this study. In another large study that evaluated dairy cattle, 6.3 percent (1,749/27,713) of pregnant cows experienced dystocia (Lucy 2001). USDA estimates the mean dystocia risk in the general cattle population at 4 percent of pregnancies (USDA/NAHMS 1997⁵²).

A more recent study of three large dairies (herds ranging between 1,000 and 5,000 cows) in Colorado indicated the overall incidence of severe dystocia (requiring assistance) at 10.9 percent for all cows on these dairies (Lombard et al. 2007). Again, calf birth weight and parity of dam were important factors in incidence of dystocia, with incidence among heifers (18.9 percent) higher than for multiparous cows (6.9 percent). This study indicated that incidence of dystocia, stillbirth and calf mortality associated with dystocia appeared to be increasing compared to previous reports. However, the report also suggested that large herd size and other management factors, such as training of farm personnel, were also factors in the increased incidence of stillbirth and later calf mortality.

Rates of dystocia in surrogate dams carrying clone pregnancies are difficult to determine as clone producers have often elected to deliver clones via planned C-section as part of their animal care protocol (Wells et al. 1999, Lanza et al. 2000 using transgenic clones, Gibbons et al. 2002, Batchelder 2005). Planned C-section deliveries are associated with decreased parturition risk, and in most cases the surrogate dam recovers without ill effects. Although this does not eliminate the risk associated with giving birth, particularly in the event of hydrops, very few surrogate dams are lost, and most recover normally. The relationship between cloning, large offspring syndrome (LOS), and dystocia will be discussed in greater detail in the next section and in section 2.b.i.a.

(c) Large Offspring Syndrome

Large Offspring Syndrome (LOS) (Table V-3) has been described as occurring at a relatively high frequency in clone-bearing pregnancies, and at a lower frequency in cattle derived from IVP and ET pregnancies, and in some cases may be related to the development of hydrops (Kruip and den Daas 1997; Chavatte-Palmer et al. 2002). This syndrome will be discussed in greater detail in Section 2. b. i. a., as it also has implications for the health and survival of the newborn animal. For the surrogate dam, LOS increases the likelihood of dystocia, frequently requiring human intervention to remove the calf vaginally, or by C-section, due to the inability of the dam to expel the calf without assistance. Reported incidences of LOS in peer-reviewed publications on cattle clones have ranged from as low as 1/12 (8.3 percent) (Miyashita et al. 2002) to as high as 12/24 (50 percent) (Kato et al. 2000). Average birth weight of clones (some transgenic) of various

⁵² <http://nahms.aphis.usda.gov/>

cattle breeds (Holstein, Brown Swiss, Angus and Holstein x Jersey crossbreeds) in the Pace et al. (2002) study was 51 ± 11 kg, with 54/106 (51 percent) live-born calves weighing more than 50 kg at birth. Given the inability to distinguish between transgenic and “just clone” pregnancies in the Pace et al. study, it is difficult to put these numbers into context with other studies of non-transgenic clones. Average birth weight of calves produced by AI or natural service varies depending on breed, and may range from 30 kg in small breed cattle to 45 kg or more in large breed cattle (NAS 1996b).

(d) Other complications

Although other complications associated with SCNT pregnancies have been noted, potential interactions with transgenic manipulation of the donor cell and predisposing conditions in the surrogate dam make it difficult to ascribe the complications exclusively to the cloning process. For example, the ketonuria⁵³ and fatty liver associated with ketosis and “fat cow syndrome”⁵⁴ described by Hill et al. (1999) are not only confounded by the existing obesity of the surrogate dams at the time of diagnosis, but also by the transgenic nature of the fetal clones. Cows that are obese at calving are most likely to develop fatty liver, and cows that develop fatty liver at calving are most susceptible to ketosis. Fatty liver can occur whenever there is a decrease in feed intake and may be secondary to the onset of another disorder. Obesity in late-gestation cattle is a commonly reported problem resulting in anorexia (due to reduced gut capacity), ketosis, fatty liver deposits, and hepatic insufficiency in pregnant cattle (Merck Veterinary Manual Online 2005⁵⁵).

Wells et al. (1999) noted weak or non-existent uterine contractions, poor mammary development and failure to lactate in cattle carrying fetal clones. Actual incidence of these complications is not known, but all have been reported in sheep (Ptak et al. 2002) and failure to lactate was noted in swine surrogate dams (http://www.fda.gov/cvm/CloningRA_Memorandum_I.htm).

iii. Unpublished Data

J.L. Edwards’ laboratory at the University of Tennessee submitted a three page table to CVM summarizing the outcomes of cloning studies in Jersey cattle conducted between 2000 and 2003 (Appendix G.) The table provides data on all five developmental nodes for clones and their surrogate dams, and summarizes the outcomes of 47 late second- and third-trimester pregnancies.

⁵³A metabolic disorder related to energy metabolism, where breakdown products of body fat spillover into the urine.

⁵⁴Pregnant cows that are obese often reduce energy intake near the time of calving, leading to rapid mobilization of body fat which predisposes them to metabolic disorders such as fatty liver and ketosis, an inability to clear the blood stream of breakdown products of fat, known as ketone bodies.

⁵⁵<http://www.merckvetmanual.com/mvm/index.jsp>

Twenty-five pregnancies (53 percent) were delivered by C-section, and five surrogate dams (11 percent) required assistance during vaginal delivery. Twenty-seven pregnancies (58 percent) resulted in hydrops, with 10 surrogate dams (37 percent of hydrops cases) euthanized as a result of this condition. The reasons for the unusually high incidence of hydrops in these surrogate dams have not been identified. In a teleconference with Dr. Edwards, she indicated that most of the C-sections were planned as part of a timed labor induction protocol introduced after several surrogate dams developed rapid onset of hydrops.

b. Developmental Node 2: Perinatal Period

i. Peer-Reviewed Publications

One factor that makes analysis of survival data on clones difficult is the difference in how stillbirth is interpreted between studies of clones compared to studies in the general population of cattle. Studies on cattle cloning reviewed for this risk assessment defined stillbirth as calves born dead. Conversely, studies on conventionally bred dairy cattle typically define stillbirth as death within 24, and sometimes 48, hours of birth (Lombard et al. 2007). This may be attributed to different methods of management between clone producers and conventional cattle operations, especially beef cattle. Due to the expense of generating clones, surrogates bearing clones are likely more closely monitored than dams in commercial cattle breeding operations. Also, in most of the studies on clones reviewed for this risk assessment, labor was induced labor and, in many cases, clones were delivered by C-section. Thus, personnel were present when clones were born, and status at time of birth could be recorded. Under more conventional breeding practices, onset of labor is spontaneous, and neonates are frequently delivered unaided and unobserved.

In the general population of cattle and sheep, neonatal death rates are typically low. Overall, the estimated death rate of beef calves within 24 hours of birth (including stillbirths) was 3.4 percent (USDA/NAHMS, 1997⁵⁶). Nix et al. (1998) found that dystocia affected calf mortality within the first 24 hours, with mortality rates increasing with increasing severity of dystocia. Overall calf mortality attributed to dystocia was 4.5 percent of all calvings in this study (2,191 births). A more recent study (Lombard et al. 2007) indicated that calf mortality among dairy heifer calves, including stillbirths, was increasing (13 percent) compared to earlier studies. Dystocia was the most influential factor on calf mortality in these studies, due to trauma of difficult labor and emergency C-section. Dystocia was also associated with high calf morbidity (illness) in a study of 2,490 beef cattle herds (Sanderson and Dargatz 2000), and in the Lombard et al. study of three large Colorado dairies.

⁵⁶ <http://nahms.aphis.usda.gov/>

Among dairy replacement heifers, the highest losses occur during the first week of life (1.8 ± 0.3 percent deaths for all heifer calves born alive). In these heifers, the most commonly reported illnesses were due to respiratory problems and scours (diarrhea), with incidence of these illnesses peaking during the first two weeks of life (USDA/NAHMS 1994⁵⁷).

Because the number of animal clones available to study is small, it is difficult to draw conclusions on rates of morbidity and mortality of live-born clones. However, some trends appear to be common across most of the studies reviewed. Early reports, beginning in 1998, of clone mortality rates were 50 to 80 percent (reviewed by Solter 2000). Survival rates have improved in some recent studies, with mortality during the first month of life of approximately 18 percent (21/117; Pace et al. 2002 for a cohort of mixed transgenic and non-transgenic clones) and 20 percent (6/30; Lanza et al. 2001 for a cohort of transgenic cattle), with most of the deaths occurring during the first 48 hours postpartum. Similarly, data supplied by Cyagra, Inc. (2003) indicate 22 percent mortality in the first 48 hours (30/134) among non-transgenic clone calves born between 2001 and 2003. A more recent study from this group (Panarace et al. 2007) which covered Cyagra operations in the US (including data submitted directly to CVM), Argentina, and Brazil between 2000 and 2005, indicated a 12 percent mortality rate among live-born calves in the first 24 hours after birth. (For a summary of survival rates among live-born bovine clones, see Table V-1.)

(a) Large Offspring Syndrome

Large Offspring Syndrome (LOS) has been described in calves and lambs produced by ET, IVP, BNT, and SCNT. References describing this syndrome in the following section include descriptions of abnormalities noted for any of these ARTs. As the name indicates, the most readily recognized sign is oversized fetus or newborn, characterized as having a birth weight greater than 20 percent above the average birth weight for that species, breed, and sex. Dystocia and related morbidity and mortality of the young animals are common in cases of LOS when C-sections are not planned. Mortality rates for LOS calves can be high (Behboodi et al. 1995, Farin et al. 2001, Farin et al. 2004, Lee RS et al. 2004). A description of clinical signs associated with LOS is provided in Table V-3. A summary of incidence and survival rates of calves born with LOS and related clinical signs are in Table V-4. Survival of LOS calves is highly variable, and appears to depend on severity of the clinical signs and neonatal management practices. Studies that included such data indicated that survival ranged from 0 to 88 percent of calves diagnosed with LOS.

⁵⁷Ibid

Table V-3: Clinical Signs Associated with Calves Displaying Large Offspring Syndrome (LOS)

Fetal size > 20% above average for species/breed Slow to stand Inability to thermoregulate Weak or absent suckle reflex Large umbilicus with patent blood vessels Deformities of limbs (tendon contracture) and /or head Disproportionate or immature organ development Increased susceptibility to infection Respiratory signs: insufficient lung surfactant, failure of lungs to inflate Cardiovascular signs: patent ductus arteriosus, enlarged heart /ventricle, septal defects Kidney defects (hydronephrosis) Hepatic congestion/fatty liver Hydrops

Stress associated with dystocia, prolonged labor and emergency C-section birth is a risk factor for large calves, regardless of how they are produced (Kato et al. 1998, Kubota et al. 2000, Lombard et al. 2007, Panarace et al. 2007). Lombard et al. (2007) noted that calves born to conventionally bred dams with mild or severe dystocia were at increased risk of hypothermia, respiratory and digestive problems, failure of passive transfer of immunity, and death. Matsuzaki and Shiga (2002) reported that SCNT clone calves born by emergency C-section had a higher mortality rate (4/5) compared to clone calves that were delivered vaginally (1/8). It is not clear whether the higher mortality is entirely due to the emergency surgery or whether adverse factors in the clones themselves contributed to the mortality.

Congenital abnormalities that may be related to fetal oversize include deformities of limbs and head, and may be a function of crowding in the uterus (Meirelles et al. 2001, Zakhartchenko et al. 1999a, Hill et al. 1999 with transgenic clones, Garry et al. 1996 with BNT clones). Intrauterine infections may also be responsible for some of these abnormalities (Kato et al. 2000, Kubota et al. 2000). LOS includes a large number of abnormalities, only some of which may be directly related to dystocia and congenital effects of unusually large size.

Some clinical signs associated with LOS also may be the result of perinatal hypoxia (oxygen deprivation) and metabolic acidosis (blood pH < 7.4) due to dystocia (Vaala and House 2002). Hypoxia during dystocia is due to premature separation of the umbilicus or other interruption in flow of oxygenated blood from the dam to the fetus. Hypoxia increases the production of lactic acid, which results in metabolic acidosis. Mild acidosis is expected in neonates, and is usually transient, but dystocia and the accompanying hypoxia result in prolonged and pronounced acidosis. The associated clinical signs include delayed time to stand; weak suckle reflex; delayed transition from fetal to neonatal circulation; pulmonary hypertension; inadequate lung surfactant production; hypothermia; impaired hepatic function; altered glucose metabolism; and poor

absorption of immunoglobulins from colostrum. These clinical signs are directly associated with dystocia, and occur in conventionally bred calves as well as calves resulting from ART and clones, when delivery is difficult or prolonged (Vaala and House 2002).

Other abnormalities reported to coincide with LOS include respiratory, cardiac, hepatic, renal, umbilical, and immunologic problems, and may occur even among animals with birth weights within the normal range for their breed. These abnormalities may result from dysregulation of developmentally important genes rather than the uterine environment (see Chapter IV), or may be the result of placental abnormalities, as discussed in Section 2.C.i.a. Systemic abnormalities including organ dysfunction result in morbidity and often result in high mortality. Pulmonary abnormalities include immature lung development, insufficient lung surfactant, and failure of the lungs to inflate. Cardiovascular abnormalities include patent ductus arteriosus and ventricular defects (Table V-3).

i. Potential Sources of LOS

In vitro culture conditions are suspected to contribute to development of LOS in IVP-derived embryos (Farin and Farin 1995, Farin et al. 2001, Bertolini et al. 2002a,b). Various culture systems used in different laboratories often use slightly different media ingredients,⁵⁸ such as fetal calf serum (FCS), bovine serum albumin (BSA), and serum derived from cows in estrous (ECS), and may expose developing embryos to hormones and growth factors that may not be in appropriate concentrations for the stage of development, possibly contributing to gene dysregulation (Sinclair et al. 1999, Gardner and Lane 2005).

Lazzari et al. (2002) noted differences in the expression of several developmentally important genes between IVF-derived blastocysts cultured with BSA and blastocysts recovered from cows bred by AI, including the IGF-I and –II receptors, fibroblast growth factor, and several genes important for the transport of glucose.

Farin and Farin (1995) compared bovine IVP embryos cultured in mixed media containing 10 percent ECS and other hormones for seven to eight days with embryos fertilized *in vivo* and collected and transferred on the same day via embryo transfer (ET). Pregnancy rates 53 days after transfer were higher for heifers (a cow that has not yet produced her first calf) receiving ET (15/19 embryos transferred; 79 percent) compared with IVP embryos (7/19 embryos transferred; 37 percent).

⁵⁸ Cells in culture require media that provide the essential nutrients and other chemical components that allow them to grow. Scientists have attempted to simulate the growth environment of the intact organism in culture media by adding certain blood components, usually serum (the portion of whole blood that remains after clotting has occurred).

Behboodi et al. (1995) reported that birth weights were not significantly different between calves produced by AI and IVP-derived calves when embryos were cultured to the blastocyst stage in sheep oviducts; however, birth weights of calves born from embryos that developed into blastocysts *in vitro* were higher than those for calves from embryos that developed in the sheep oviduct or from AI. In this study, 7/8 calves produced from embryos cultured *in vitro* died within 48 hours of birth, compared to 1/8 calves from embryos cultured in the sheep oviduct after fertilization. Hasler et al. (1995) noted that approximately seven percent of clients purchasing cows carrying IVP-derived calves reported high birth weights. In this study, of 428 IVP calves born, 67 died at birth (15.6 percent). Block et al. (2003) reported 5/28 IVF calves were stillborn (17.9 percent), one of which suffered hydrops, and another hydrocephalus. In contrast, a recent study (Xu et al. 2006) reported that out of 458 calves born from vitrified IVF embryos, two calves were born with unspecified physical defects, one of which died within an hour of birth.

Table V-4: Incidence of LOS and related clinical signs and survival rates of neonatal calves produced with ARTs¹						
Study	Transgenic Status	Clone LOS incidence	Survival of LOS clones	Comparator LOS incidence	Survival of comparators	Comments
Batchelder 2005	None	8/8 (1.00)	2/8 (0.25)	2/9 (0.22)	9/9 (1.00)	Comparators were ET (n=6) and AI (n=3). See Table V-5 for clinical signs.
Behboodi et al. 1995	Some	NP	NP	4/8 (0.50) 0/72 (0.00)	NP	8 IVF calves compared to 72 AI calves
Block et al. 2003	None	NP	NP	3/28 (0.11)	NP	All calves derived by IVP
Cyagra 2003: Appendix E	None	73/123 ² (0.59)	56/73 ³ (0.77)	NP	NA	Clinical signs: contracture; septicemia; nephritis; failure to thrive; umbilical, gastrointestinal, cardiac-circulatory anomalies
Edwards (unpublished)	None	14/27 (0.52)	11/14 (0.77)	NP	NA	See Appendix G. Multiple internal organ defects, gastro-intestinal problems

Garry et al. 1996	None	34/40 (0.85)	26/34 (0.77)	0/26 (0.00)	NA	BNT clones, AI comparators. Clinical signs: respiratory and musculo-skeletal
Gibbons et al. 2002	None	8/9 (0.88)	7/8 (0.88)	NP	NA	Clinical signs: respiratory, umbilical, septicemia, hydrocephalus, GI problems
Gong et al. 2004b	None	7/27 (0.26)	0/27 (0.00)	NP	NA	
Hasler et al. 1995	NA	NP	NA	23/343 (0.07)	NP	Data gathered from owners of IVF-pregnant cows
Hill et al. 1999	All	4/8 (0.50)	2/4 (0.50)	NP	NA	Clinical signs: respiratory, umbilical, cardiac, hepatic anomalies; contracture, acidosis, weak suckling reflex
Heyman et al. 2004	None	7/50 (0.14)	NP	NP	NP	Birth weights of AI comparators used to set range for determining LOS in clones
Kato et al. 2000	None	6/17 (0.35)	3/6 (0.50)	NP	NA	Clinical signs (may be result of Akabane virus): musculo-skeletal, kidney abnormalities
Kubota et al. 2000	None	6/6 (1.00)	4/6 (0.67)	NP	NA	Clinical signs: respiratory, polyuria and polydypsia Akabane virus
Lanza et al. 2001	Some	14/30 (0.46)	8/14 (0.57)	NP	NA	
Miyashita et al. 2002	None	1/12 (0.08)	0/1 (0.00)	NP	NA	

Table V-4: Incidence of LOS and related clinical signs and survival rates of neonatal calves produced with ARTs ¹						
Pace et al. 2002	Some	70/106 (0.66)	59/70 (0.84)	NP	NA	Clinical signs: umbilical, respiratory, cardiac, musculo- skeletal, GI; hydrocephalus, bacterial infection
Xu et al. 2006	None	NP	NP	2/458 (0.0004)	457/458 (1.00)	Studied sex-sorted, vitrified IVF embryos
Zakhartchenko et al. 1999a	None	1/2 (0.50)	0/1 (0.00)	NP	NA	Clinical signs: musculo- skeletal and hepatic abnormalities
¹ Data on live-born calves ² Of 134 calves born, 123 were born alive. ³ Denominator is number of calves identified with LOS and/or related clinical signs NA = not applicable NP = not provided; data not available						

Sire selection may also contribute to the large calves resulting from ET and IVP. Knight et al. (2001) indicated that one of the sires used in a two year study had a tendency to produce large ET calves. High birth weights in this study may have contributed to low survival rates in a previous study in the same herd. In cattle, sires may be selected based on their IVP and ET calf birth weight records (Knight et al. 2001). A similar relationship between high birth weights of calves was noted in two other studies of IVP (Thompson et al. 1998, Block et al. 2003).

In a large study comparing birth weights, dystocia incidence, and neonatal death rates in AI, ET, IVP, and BNT produced calves of various beef and dairy breeds from labs in several countries, Kruip and den Daas (1997) noted that on average 31.7 percent of IVP calves (n=308) weighed more than 50 kg at birth, compared to 10 percent for AI (based on 495,000 calf records from the Netherlands). Interestingly, only 15 percent (n=126) of calves produced by BNT had birth weights greater than 50 kg in this study. For one breed (Holstein-Friesian), perinatal losses were similar between AI (n=1,160) and ET (n=45) calves (6.1 ± 0.6 and 6.6 ± 0.6 percent), but loss was higher for IVP calves (14.4 ± 2.3 percent; n=251). Perinatal death loss was higher (11.6 vs. 2.3 percent) for IVP (n=308) compared with BNT calves (n=126) for the six breeds studied (Holstein-Friesian, Belgian Blue, Simmental/Fleckvieh, Limosin, Piedmontese, and Alentejano).

More recent studies in which IVP and SCNT embryos were produced under the same culture conditions reported considerably higher incidences of LOS in fetal and adult cell SCNT-derived

calves compared to IVP (Heyman et al. 2002, Chavatte-Palmer et al. 2002, Matsuzaki and Shiga 2002), indicating that culture conditions may not be the only factor influencing the development of LOS in cattle clones. Average birth weight of adult-cell SCNT clones was significantly higher than IVP-derived calves (53.1 ± 2.0 kg vs. 44.5 ± 2.1 kg) in the Heyman et al. (2002) study. Chavatte-Palmer et al. (2002) found considerable variability in organ development among calf clones, and reported that one apparently normal clone fetus had small kidneys for its size and stage of development. Also in this study, Chavatte-Palmer et al. noted differences in body temperature, plasma leptin, thyroxine (T4) and insulin-like growth factor-II (IGF-II) in surviving clones compared to IVP and AI controls during the first week to 15 days after birth, although the clones appeared normal and healthy. Differences between clones and controls resolved by 50 days of age (see Chapter VI for a more complete discussion of this study). The differences in outcomes between SCNT and IVP pregnancies observed in these studies suggest that some additional factor(s) may be at least partially responsible for the higher rate of abnormalities in animal clones compared to IVP calves, and not solely due to culture conditions. One possible explanation for this increase in abnormalities is incomplete epigenetic reprogramming (see Chapter IV).

In a later study by this same group (Chavatte-Palmer et al. 2004), an additional cohort of 58 live-born calves were followed through maturity. Clone survival after the first week following birth was 76 percent (44/58). Clinical signs and necropsy findings for nine clones that died during the perinatal period included hyperthermia, umbilical hernia, respiratory problems, ascites (abnormal fluid accumulation) in the chest and abdomen, fatty liver, limb deformities, various digestive tract problems, and abnormal or degenerating kidneys.

Culture media requirements may differ between SCNT and IVP embryos. Mastromonaco et al. (2004) compared development to blastocyst for IVP and SCNT embryos using different media ingredients at different stages of the *in vitro* process (oocyte maturation and embryo culture stages). Although IVP embryos had similar rates of development to blastocyst and hatched blastocyst regardless of culture media used, development to blastocyst was greater among SCNT embryos cultured in synthetic oviductal fluid with 2 percent steer serum. Unfortunately, this study only looked at development through day 9 of embryo culture, and did not examine *in vivo* embryo development or subsequent calving outcomes. It is possible, however, that culture conditions impact epigenetic reprogramming, and this may be related to differences in outcomes observed in the Heyman et al., Chavatte-Palmer et al., and Matsuzaki and Shiga studies.

In a recent study comparing SCNT (n=8) to ET (n=6) and AI (n=3), Batchelder (2005) noted large birth weights among three Hereford clones (n=3), ranging from 50.0 to 71.0 kg. By comparison, ET comparator Hereford calves ranged from 31.5 to 48.0 kg (n=3). Curiously, the mean weight for Holstein clones (n=5) was similar to contemporary ET comparators (n=3) (37.1

vs. 39.4 kg), and within the average range for Holstein heifer calves. Neonatal clones in this study had lower RBC (6.8×10^6 vs. 8.6×10^6 cells/ μ l) and hematocrit at birth than their comparators, and remained low for the first hour after birth, but were similar to comparators thereafter. White blood cell counts (WBC) and differential patterns were similar between clones and comparators. Clones exhibited lower blood glucose and lactate levels during the first 24 hours after birth than comparators, but were similar to comparators by 48 hours.

To explore the possible relationship between epigenetic dysregulation and organ abnormalities associated with increased risk of mortality in SCNT-derived neonatal calves, Li et al. (2005) investigated the expression (defined in this experiment as relative levels of messenger RNA (mRNA))⁵⁹ of eight developmentally important genes (*PCAF*, *Xist*, *FGFR2*, *PDGFRa*, *FGF10*, *BMP4*, *Hsp70.1*, and *VEGF*) in nine clones that died within the first two days after birth. At necropsy, seven calves were found to have multiple cardiac, hepatic, and pulmonary defects; two calves had underdeveloped kidneys; two calves had hemorrhagic spleens; and three calves had brain lesions or underdeveloped brain.⁶⁰ Compared to organs from healthy AI-derived calves ($n = 3$), levels of mRNA encoding several genes were altered in heart, liver, spleen, kidney, lung, and brain tissues from the deceased clones. The magnitude of differences in mRNA levels seemed to be the greatest (two- to five-fold) in heart tissue, in which expression of five of the eight genes studied (*PCAF*, *Xist*, *FGF10*, *BMP4*, and *VEGF*) was up-regulated, and least in kidney, in which expression of two genes (*FGF10* and *PDGFRa*) was down-regulated by approximately 60 percent. Across all of the tissues studied, levels of mRNA encoding *VEGF*, *BMP4*, *PCAF*, and *Hsp70* demonstrated the greatest changes (either increased or decreased). The products of these genes are involved in regulation of blood vessel formation, regulation of lung and heart development, transcriptional coactivation, and protection against apoptosis,⁶¹ respectively. These observations support the idea that in some clones, aberrant epigenetic reprogramming may alter the expression patterns of genes that control embryo development and organogenesis. In this study, the level of dysregulation was apparently sufficient to result in congenital organ defects that were not compatible with life outside the uterus. It is not known whether congenital organ defects in animals derived by other ARTs are also associated with altered patterns of gene expression.

⁵⁹ Within cells, mRNA is transcribed from DNA and carries coding information to the intracellular sites of protein synthesis where it is translated into amino acids, the building blocks of proteins. Thus, mRNA encodes the “blueprint” for cellular protein products is often used as a measure of gene expression.

⁶⁰ Specific abnormalities were described in the heart (patent foramen ovale, cardiac muscle putrescence, congestion, enlarged ventricle, enlarged heart, and valvular incompetence); liver (cysts, congestion, fatty, enlargement, single lobe); spleen (hemorrhage); lungs (thickening of cells lining the alveoli, collapsed, congestion, aplasia, more than two lobes); kidneys (small or abnormally shape, renal aplasia with fatty accumulation); and brain (under-development with hemorrhage and edema).

⁶¹ Apoptosis is a form of programmed cell death, and plays an important role in embryonic development.

Batchelder (2005) also noted several clinical signs often associated with LOS in both Holstein and Hereford clones, including delayed time to suckle and stand, hypoglycemia, forelimb flexor tendon contracture, enlarged umbilicus, patent urachus, and respiratory distress. These clinical signs were not always associated with high birth weight. Interestingly, a small number of comparators exhibited some of these same clinical signs. Table V-5 is partly reproduced from Batchelder 2005.

Table V-5: Clinical signs observed in neonatal clones and comparators for Batchelder 2005.		
Clinical Sign	Clones	Comparators
Time to nurse (> 3hrs)	5/8	0/9
Time to stand (>3 hrs)	5/8	1/9
Hypoglycemia ($\leq 50\text{mg/dl}$)	3/8	2/9
Respiratory distress	3/8	1/9
Flexor tendon contracture	4/8	0/9
Enlarged umbilical vessels	8/8	2/9
Patent urachus	5/8	1/9

All calves in this study survived the first 48 hours; however, one clone died at 72 hours after birth, and another at six days after birth. The first clone, a Hereford heifer, was the largest at birth (71.0 kg), and at necropsy was diagnosed with pulmonary hypertension and multiple severe organ abnormalities including diffuse fibrosis of the liver, dysplasia of the biliary system, right ventricle hypertrophy, and patent ductus arteriosus. The second clone died at six days of age after suffering bloat and various other clinical signs involving the heart and lungs.

(b) Other complications

In discussing health and mortality among clones it is often difficult to distinguish between defects resulting from the uterine environment, placentation, and/or difficulties during delivery, and epigenetic factors intrinsic to the clone that impede normal development of the fetus and adaptation following birth. Dystocia, for example, can result in premature separation of the placenta, causing inhalation of amniotic fluid prior to birth, predisposing the neonate to pneumonia in both conventional calves (Moore et al. 2002) and clones (Kato et al. 1998). Respiratory failure is one of the most commonly reported clinical signs in neonatal clones (Table V-3), and appears to result from numerous causes, including inadequate surfactant and failure of the lungs to inflate, as well as pneumonia arising from various causes (Garry et al. 1996, Hill et al. 1999, Chavatte-Palmer et al. 2002). Pneumonia may result from dystocia in natural pregnancies as well as those derived by ART (Moore et al. 2002). However, many of the respiratory conditions reported to occur in association with LOS (failure to inflate) have not been reported for calves from natural service or AI, and may be peculiar to ARTs that involve more

extensive *in vitro* manipulation of the embryo (*i.e.*, IVF and cloning), or may be related to labor-induction protocols (Batchelder 2005).

Calves exhibiting LOS may also show prolonged time to stand and poor or late-developing suckling behavior (Chavatte-Palmer et al. 2002, Pace et al. 2002 (mixed transgenic and non-transgenic clones), Batchelder 2005). Poor suckling may preclude immune transfer in colostrum-dependent species, resulting in decreased ability to respond to immune challenge. Most of these studies, however, indicate that colostrum was administered by tube-feeding if the animal failed to suckle within one to two hours postpartum (Garry et al. 1996 (BNT clones), Hill et al. 1999 (transgenic clones), Gibbons et al. 2002, Batchelder 2005). Poor immune response in such cases may be due to a number of causes: inability of the neonate to absorb immunoglobulins; colostrum that is inadequate in immunoglobulin content; excessive or overwhelming stress; or high levels of pathogens in the neonatal environment. Clone producers have indicated that some calves are born with large umbilici, often with patent (open) blood vessels. This factor may increase the risk of bacterial infection, and clone producers indicated that surgery was generally performed on the enlarged umbilici of calves to reduce the risk of infection (see CVM Memorandum I at www.fda.gov/cvm/cloning.htm; also Appendix E and Batchelder 2005).

Most studies that reported supplemental colostrum feeding did not indicate the source of the colostrum or whether tests of its adequacy (gravimetric density or IgG concentration) had been performed. Two studies reported testing colostrum of surrogate dams for adequacy or blood tests of neonates to determine immunoglobulin status (Hill et al. 1999, Pace et al. 2002). In the study of transgenic clones by Hill et al., several of the surrogate dams were judged to have adequate colostrum. Transgenic calf clones that failed to suckle were administered colostrum by tube and fostered to other cows as needed. Pace et al. (2002) reported testing plasma IgG of calves 12 hours after birth, followed by plasma infusion if plasma IgG concentrations were less than 1,200 mg/dL. Calf clones in this study were reported to have normal serum IgG levels 24 hours after birth. As noted throughout this report, the data derived from clones that are transgenic are extremely difficult to extrapolate to “just clones” (the only subject of this risk assessment) because of the inability to determine the relative contributions of the transgenic modification and the cloning process to the observations.

In Batchelder 2005, clones were provided 2 liters colostrum (either by bottle or esophageal tube) within three hours of birth as well as supplemental plasma by I.V. over 40 minutes. At 24 hours after birth, clones and comparators had similar levels of serum IgG. However, one clone had sub-normal IgG (435 mg/dl IgG), and was classified as having failure of passive transfer of immunity, and a second clone was classified as marginally protected (1500 mg/dl IgG). Batchelder related failure of passive immune transfer to poor metabolic status and respiratory distress.

ii. Cyagra Data: Perinatal Cohort

A complete discussion of the Cyagra dataset, including how it was analyzed and the context in which results should be interpreted, is presented in detail in Appendix E. Briefly, the Cyagra dataset provided information on the overall health status and laboratory tests (clinical chemistry and blood cell parameters (hemograms)) for a group of SCNT-derived cattle clones and their approximately age- and breed-matched comparators. Among 10 neonates, four liver-related analytes were lower in clones than comparators: AST, GGT, cholesterol, and bile acids. Except for the values from one calf that did not survive, all red blood cell analytes were within the comparator group range. Three calves, all of which were infected with rotavirus, had low lymphocyte counts (lymphopenia).

Of the 134 clone calves in the Cyagra cohort, 11 were stillborn. Birth weights were available for 34 of the 123 live-born clones, and ranged from 19.5 kg (a twin calf) to 76.8 kg. Eighteen of the 34 (53 percent) birth weights were at least 20 percent above the average for their breed. Most oversized calves (13/18 (72 percent)) survived the critical first 48 hours after calving. Six of the oversized calves did not exhibit any other clinical signs associated with LOS. Fifty-five additional calves that were not oversized at birth, or for which birth weights were not available, showed clinical signs often associated with LOS; 43 of these animals survived the first 48 hours after calving. The most common clinical sign was umbilical problems (41 cases), followed by tendon contracture (15 cases), ranging from mild to severe. There were also four animals with respiratory signs, five with cardio-vascular signs, three with thermoregulatory problems, two with renal or nephric signs, and five animals listed as having “abnormal development.” Some of the calves exhibited more than one sign, often umbilical problems with contracture, cardiac or respiratory signs.

iii. Unpublished data

Two small datasets on perinatal bovine clones were also submitted to CVM. These are presented in their entirety in Appendix G, along with a more complete analysis. In brief, the body temperatures, pulse and respiration rates of 19 clones of unidentified breeds for the first 72 hours after birth averaged 39.4°C, 95.2 beats/minute, and 57.4 breaths/minute. The second set of data submitted were birth records and veterinary observations for two Holstein heifer calves, indicating the animals were generally in good health, showed normal increases in plasma glucose in response to feeding, and total protein and glucose values similar to non-clone neonatal calves in the Cyagra dataset.

According to the data submitted by J.L. Edwards' laboratory at the University of Tennessee (Appendix G), 19/27 (70 percent) live-born Jersey clones survived the perinatal period. Causes of death included placental abnormalities, suspected detached umbilicus, and hepatic, cardiac and renal lesions or defects. Average birth weight for these calves was 31.6 kg, and ranged from a very low birth weight calf (6.3 kg) to a high of 45 kg. A study at North Carolina State University indicated average birth weight for female Jerseys was 23.64 kg (Washburn et al. undated circular).

c. Developmental Node 3: Juvenile Development

i. Peer-Reviewed Publications

From weaning to puberty, the rates of morbidity and mortality in AI-produced and naturally bred cattle are generally very low. According to USDA statistics (USDA/NAHMS 1996), mortality of dairy replacement heifers (from weaning to calving) was 2.4 percent. In beef replacement heifers, the most common cause of morbidity was pinkeye (1.9 percent) followed by scours (*i.e.*, diarrhea, 1.0 percent; USDA/NAHMS 1997⁶²).

Compared to the perinatal period, there is less detailed information in the literature on the health of bovine clones during the juvenile period. Most studies merely report that animals surviving the first 30 to 60 days postpartum are "healthy and normal" (Campbell et al. 1996, Lanza et al. 2000 with transgenic clones, Heyman et al. 2002). Kubota et al. (2000) reported that veterinary exams, growth curves and blood clinical chemistry were used to determine the health of six clone calves, and that no differences were noted between clones and age matched controls.

In a study of the effects of fetal bovine serum on the development of clones, Lawrence et al. (2005) reported the death of the single surviving calf (from the serum fed group) at nine months of age due to enterotoxemia, attributed to gastrointestinal infection with Type A *Clostridium perfringens*. Various gastrointestinal and immunological problems have been reported in cattle clones, including bloat, poor motility, and poor nutrient absorption (Cyagra 2003: Appendix E, Wells et al. 2004, Batchelder 2005). Some studies of clones have also reported reduced immune responsiveness, although this has mostly been noted in neonatal clones (Renard et al. 1999, Chavatte-Palmer et al. 2004, Wells et al. 2004). Type A *C. perfringens* is a common soil and feed-borne bacteria. Enterotoxemia caused by proliferation of Type A *C. perfringens* and release of toxins in the gut is normally prevented by vaccination and good feeding management. The report of this study does not state whether this calf was vaccinated, nor does it provide details as to the possible source of the infection. The lack of detail in this study makes it difficult to

⁶² <http://nahms.aphis.usda.gov/>

determine whether the death of this calf was related to cloning, or if it was due to management practices.

Heyman et al. (2007) reported that between one quarter to one third of calves produced at the INRA laboratory died by six months of age, but calves surviving beyond that time had normal physiological status. It has been suggested that clones may have different sensitivity to stress compared to conventional animals. Stress affects a number of physiological systems throughout the body, and is often associated with increased secretion of cortisol and catecholamine metabolites (epinephrine and norepinephrine) by the adrenal gland (Minton 1994). These hormones result in physiological changes that allow the animal to adapt to stressful situations. Stress can have a negative impact on immune function and can make animals more susceptible to infection. Therefore, Heyman et al. (2007) postulated that chronic stress in bovine clones might be the basis for the sudden unexplained death of clones reported in some studies (*e.g.*, Chavatte-Palmer et al. 2004; Wells et al. 2004). To test this postulate, Heyman et al. (2007) measured plasma concentrations of cortisol and urinary concentrations of epinephrine and norepinephrine in clones and comparators every two months from 4 to 36 months of age. The variability of measurements was greater in clones, but no differences were found between clones and comparators in concentrations of blood cortisol or catecholamine metabolites, suggesting that apparently healthy clones were not suffering from chronic stress.

Heyman et al. (2007) noted some differences between clones and AI-derived calves up to two months of age, even among clones that were clinically healthy. As an example, these authors note that although hematological parameters were within the normal range, hemoglobin and hematocrit of clones were lower than those of their age-matched AI comparators. The authors do not describe the reference range used in this study. As discussed in Appendix F on the analysis of Cyagra data, it is important to use a range that is relevant to the animals of interest. Thus, the authors' use of an age-matched comparator group raised under the same conditions is appropriate, and their finding that these young cattle clones may have experienced subclinical anemia is likely a valid observation. However, for 21 clones and 19 co-raised comparators followed from four months through three years of age, no differences were noted for daily gain or feed intake, and cardiovascular, respiratory and locomotor functions of clones were considered normal. The authors did note differences in concentrations of fibrinogen (a clotting factor) and albumin in blood chemistry of clones vs. comparators. No differences were noted in subpopulations of mononuclear blood cells, and no differences in antibody response between 17 healthy clones and 17 comparators aged two months to five years, suggesting that immune function of apparently healthy juvenile clones was normal, although the antigen-specific response of clones to ovalbumin was weaker than comparators at eight to nine months of age. However, when this test was repeated with these animals plus eight additional clones and

comparators at 10 to 13 months of age, there was no difference in antigen-specific response between clones and comparators.

Panarace et al. (2007) reported that 42 percent of calves born, including stillbirths, died before 150 days of age (163/388). (In this study, the stillbirth rate, reported as percent of calves born dead, was 18 percent (71/388)). Of 317 calves born alive, 225 (71 percent) survived beyond 150 days. The most common causes of mortality in the calves that died between 24 h and 60 days after birth were enlarged umbilical cord (37 percent), depression/prolonged recumbency (20 percent), and hyper/hypothermia (17 percent). In calves that died without any other sign of abnormality, the most common clinical sign was respiratory distress (19 percent).

In a long term study of health and survival of clones and their offspring, Wells et al. (2004) stated that the most common cause of mortality (either by natural death or euthanasia) of young clones at their facility was musculoskeletal abnormalities (severe tendon contracture and chronic lameness). They also reported two cases of death due to bloat, and an unspecified number of clones dying due to endophyte toxicity. Gastro-intestinal problems, including bloat, have been reported in other studies (Cyagra 2003: Appendix E, Batchelder 2005), but can also result from poor feeding/grazing management in conventional cattle. Endophyte toxicity results from grazing fungus-infected grass by cattle sensitive to the toxin. Wells et al. acknowledge that this trait is inherited in certain lines of cattle, and likely was related to the genetics of the nuclear donor. (The clones affected by this toxicity were derived from the same donor.) Other causes of death among clones (besides those attributed to accident or management problems) included anemia, chronic heart failure, and degenerative nephrosis, problems which have been noted in other studies (Cyagra 2003: Appendix E, Chavatte-Palmer et al. 2004). Growth rates of heifer clones were within the range for conventional heifers raised under typical management conditions in New Zealand (0.677 ± 0.066 kg/day). Heyman et al. (2004) also reported growth rates of 23 clones were within expected limits for Holsteins ($0.7 - 0.8$ kg/day). Growth rate was not influenced by birth weight in these studies.

Shiga et al. (2005) reported on growth rates of four clones (two steers and two intact bulls) of a 12 year old Japanese Black bull. Although the average birth weight of the clones was greater than that of AI-derived comparators (43.1 ± 4.1 vs. 31.3 ± 4.0 kg), post-natal growth rates were similar between groups, and by two years of age, body weight and shoulder height were similar between clones and comparators.

Chavatte-Palmer et al. (2002) monitored the growth and development of clones (n=21) compared to IVP (n=20) and AI (n=176) controls. For each variable measured, numbers of clones and controls varied (see Table VI-2 in Chapter VI). For the first week after birth, the mean rectal body temperature was higher in clones (n=10) than AI controls (n=10), and some temperature

spikes (up to 41° C for periods lasting 24 – 36 hours⁶³) were observed. Body temperatures of clones were reported as remaining elevated for the first 50 days, although data were only provided for the first week. The investigators were unable to determine the cause of the elevated body temperatures in clones: no bacterial infections were detected, and animals did not respond to anti-inflammatory drugs commonly used to lower body temperature. Levels of thyroxine (T4), a hormone that controls metabolic rate in most tissues, were tested to determine if they could explain the temperature difference between clones and controls. Plasma T4 levels were lower in clones than controls during the first two weeks of life, and were similar to controls thereafter. Chavatte-Palmer et al. (2002) noted that lower plasma T4 levels coupled with elevated body temperatures in young calves was consistent with the findings of Carstens et al. (1997b).

Carstens et al. (1997b) measured metabolic rates and increases in other blood parameters related to stress in different breeds of neonatal calves before and after stimulation with norepinephrine. The Carstens study focused on the regulation of brown adipose tissue by norepinephrine. Brown adipose tissue (BAT) is found in neonates of many mammalian species, and, while it contains fat, its primary function is to generate heat (unlike white adipose, which is primarily a fat depot) to keep the newborn warm during cold stress. Brown adipose cells contain large concentrations of mitochondria (which is what makes it brown in appearance). Mitochondria are often referred to as the “power houses” of cells, because they generate energy from nutrients through a process known as oxidative phosphorylation to produce adenosine triphosphate (ATP), the ultimate (short-term) form of energy storage immediately prior to use by the cell to carry out functions that require energy. This process is relatively inefficient, resulting in some energy loss from the system as heat. This heat loss is the primary source of body temperature, which is relatively constant in warm-blooded animals (Blaxter 1989).

Brown adipose tissue metabolism is stimulated by norepinephrine (Blaxter 1989), which is consistent with the norepinephrine release observed in response to stressful stimuli such as cold (Voet and Voet 1995). Unlike other tissues, BAT cell mitochondria contain an extra protein, controlled through the action of norepinephrine, which allows the oxidative phosphorylation pathway to become “uncoupled” from the production of ATP. Although oxidative phosphorylation continues, ATP is not produced. This interruption in the pathway to ATP results in the release of large amounts of energy as heat (Voet and Voet 1995). In some species, BAT persists into adulthood, but in cattle and some other cloven-hoofed species, BAT usually disappears (is broken down and metabolized) following the neonatal period (Blaxter 1989).

In most tissues of the body, metabolic rate is controlled by T4. In animals that do not possess BAT, body temperature is a function of metabolic rate (Voet and Voet 1995). According to

⁶³ For dairy cattle, normal temperature is approximately 38.5 °C.

Carstens et al. (1997b) T4 appears to have differential effects on BAT compared to other tissues, in that elevated T4 suppresses thermogenesis (heat formation and release) in BAT. It may do this by reducing metabolic rate in this tissue, or by blocking the activity of the protein that uncouples oxidative phosphorylation, allowing energy to be captured as ATP, as it is in other cells, and reducing the amount of energy that is lost as heat. In the Carstens et al. (1997b) study, T4 was not affected by norepinephrine challenge, but metabolic rate and body temperature increased, which the authors attributed to increased heat production in the calves' BAT.

Because the higher body temperatures of clones observed in the Chavatte-Palmer et al. (2002) study were independent of T4 levels, it is possible that the hyperthermia experienced by the clones resulted from increased BAT metabolism. However, norepinephrine was not measured in this study (Chavatte-Palmer et al. 2002), so it is not possible to determine whether that was the cause of the elevated temperature levels in these clones. Another question with potential relevance to BAT metabolism is mitochondrial heteroplasmy (see Chapter IV for more on this issue)⁶⁴. This term refers to the possibility that some of the mitochondria from the nuclear donor cell may survive and persist alongside mitochondria from the oöcyte. The Chavatte-Palmer et al. (2002) study did not examine mtDNA of clones exhibiting hyperthermia.

In a follow-up study, Chavatte-Palmer et al. (2004) reported that 38/44 clones (86 percent) surviving the perinatal period lived to six months of age. The authors reported an additional four clones with thymic aplasia or atrophy (underdeveloped or degenerating thymus gland) since the first report of a clone with this condition in their laboratory (Renard et al. 1999). It is not clear from the current study whether these four clones were also the result of multiple rounds of cloning as in the Renard et al. report. To our knowledge, this is the only laboratory reporting thymic aplasia as a clinical problem in clones. On necropsy, the thymus glands of these calves exhibited abnormal tissue organization, suggesting epigenetic errors (see Chapter IV). Three calves in this group died suddenly with few or no clinical signs: two died following the onset of diarrhea and one calf died without any apparent cause. Another calf was diagnosed with diabetes

⁶⁴ Sperm contains relatively few mitochondria compared to oöcytes. Approximately 10 copies of mitochondrial DNA (mtDNA) have been detected in sperm, whereas oöcytes are estimated to contain $317 - 795 \times 10^5$ copies of mtDNA (Hiendleder 2007). Thus, in sexual reproduction, it is assumed that the massive difference in numbers may overwhelm the few paternal mitochondria, leading to the now widely accepted theory that mitochondrial inheritance comes solely from the oöcyte. There have, however, been a few cases of naturally occurring mitochondrial heteroplasmy, which may occur more frequently in interspecies hybrids (Hiendleder 2007). Some researchers working with SCNT have noted that mitochondria from the nuclear donor cell survive and replicate alongside mitochondria from the oöcyte in embryonic and postpartum clones (Steinborn et al. 2002, Takeda et al. 2003, Jiang et al. 2005, Lloyd et al. 2006), but others have noted normal maternal mitochondrial inheritance or very limited heteroplasmy (Evans et al. 1999, Steinborn et al. 2000, Loi et al. 2001, Hiendleder et al. 2003). As with sexual reproduction, persistent mitochondrial heteroplasmy in SCNT has mostly been noted in clones using donor cells of one species or subspecies fused with oöcytes of a different species or subspecies. None of the authors related mitochondrial inheritance to health problems in clones, nor has heteroplasmy in sexually reproduced animal hybrids been linked to disease (Hiendleder 2007).

insipidus. The only post-mortem finding on the diabetic calf was an enlarged pituitary, suggesting abnormal hormonal regulation. The authors also noted that, although hematological values for clones were within the normal range, hemoglobin levels of 25 clones were lower than those of 19 AI contemporary comparators for the first 65 days after birth. This finding appears to reinforce this group's earlier conclusion that clones cannot be considered physiologically normal until approximately two months of age.

Batchelder (2005) also noted periodic moderate to severe hyperthermia in young Hereford and Holstein clones (n=8) until approximately 60 days of age. As in Chavatte-Palmer et al. (2002), hyperthermia was unresponsive to treatment with either anti-inflammatory drugs or mechanical attempts at cooling (fans, alcohol baths). These studies also reported no changes in behavior or other signs of illness in calves with hyperthermia. Respiratory rates in clones followed a similar pattern to body temperature in this study, increasing during temperature spikes. This is expected, as increased respiration rate (including panting) is a means of dissipating body heat for cattle, or could be related to increased oxygen demand by BAT (Blaxter 1989).

Additional endocrine measures evaluated in the Chavatte-Palmer et al. (2002) study included cortisol, insulin-like growth factor-I (IGF-I), IGF-II, IGF binding protein, leptin, and growth hormone. Blood samples for these assays were collected from all 21 clones and 8 AI calves (described above). Cortisol levels were decreased in both clone (n=11) and non-clone calves (n=2) born by C-section relative to calves born vaginally (10 clones and 6 non-clones). By seven days of age, clones and AI controls exhibited similar cortisol levels following an ACTH (adrenocorticotrophic hormone) challenge (ACTH induces the production of cortisol), providing evidence that these calves were not suffering from stress. No differences in levels of growth hormone, IGF-I, or IGF binding protein were observed between clones and AI controls. Levels of IGF-II were relatively high at birth among clones, but rapidly decreased within 15 days, until clones had slightly lower IGF-II levels compared to AI controls. Leptin levels were higher in clones than controls during the first week of life, but were similar to controls thereafter. The role of leptin in ruminant metabolism is not well understood, and the relevance of this measurement to an assessment of animal health cannot be determined. Insulin and glucose response after eating were not different between clones and AI controls in this study (Chavatte-Palmer et al. 2002).

Govoni et al. (2002) also published one of the few studies of postnatal growth and development of cattle clones. Although this study was performed on only four animals generated from the same donor cell, the report is fairly detailed. Holstein heifer clones were paired with age, sex and breed matched controls produced by AI. All calves were pre-pubertal at the beginning of the study (approximately 5 months of age). Control calves were housed in adjacent pens in the same barn as clones. Differences were noted over time between clones and controls in growth

hormone (GH) and IGF-I levels. Over the course of the six month study, GH levels declined in controls, but in clones GH levels began to increase beginning at about nine months of age. Average plasma concentration of IGF-I was generally lower in clones compared to controls. Although IGF-I increased in both groups over the course of the study, clones continued to have lower IGF-I concentrations compared to age matched controls (203.7 ± 13.8 vs. 306.3 ± 13.1 mg/mL).

Growth hormone has been reported as a major modulator of systemic concentrations of IGF-I (Le Roith et al. 2001), and has been demonstrated to stimulate hepatic production of IGF-I after its release from the hypothalamus. Somatostatin, which is stimulated by high levels of IGF-I, suppresses GH synthesis, which in turn causes a reduction in IGF-I synthesis in the liver. Mice lacking functional hepatic synthesis of IGF-I grow normally, perhaps because IGF-I is also synthesized in muscle, but GH levels in these mice are elevated. If cattle and mice were to exhibit similar control mechanisms, the expectation would be that the increased GH levels in clones after 9 months of age would have resulted in a concomitant increase in circulating IGF-I. This was not observed by Govoni et al. (2002). Clones were more responsive than controls to factors controlling GH release, but showed a similar response as controls to inhibiting factors. For example, the magnitude of response to injected Growth Hormone Releasing Hormone (GHRH) was five times higher in clones than controls, although GH returned to basal levels 40-50 minutes post stimulation. Conversely, injecting animals with Somatotropin Release Inhibiting Factor (SRIF) was successful in equally inhibiting response to GHRH in both clones and controls. IGF Binding Protein 2 (IGFBP2) levels were not different between growing clones and controls in the Govoni et al. study. IGFBP3 (another binding protein for IGF-I) levels were lower in clones compared to controls. The altered levels may be related to the lower IGF-I levels in these animals, possibly resulting in down-regulation (reduced synthesis) of this binding protein. The Savage et al. (2003) study, discussed below, noted no abnormalities in growth or behavior of these clones. Neither of these studies noted health problems in the clones, suggesting that these differences were insufficient to cause any metabolic perturbation.

Savage et al. (2003) performed behavioral studies with four Holstein heifer clones and age- and breed-matched control heifers (the same group of animals as reported in the Govoni et al. study). Animals were studied beginning between 32 and 36 weeks of age, at which point there were no differences in weight or height between the clones (205.5 ± 9.9 kg; 117.0 ± 1.8 cm) and controls (211.4 ± 7.4 kg; 119.5 ± 1.4 cm). All calves were raised together under the same management conditions. Based on a series of studies evaluating approach to other animals and novel objects, clones exhibited age-appropriate behaviors, but were reported to be more aggressive and inquisitive than controls, and spent more time grooming and socializing. Clones tended to spend less time in playful behavior than controls. Review of records on the cow that served as the nuclear donor for the clones indicated that she had displayed similarly aggressive and inquisitive

behavior as a young animal, suggesting that at least some of these behavioral traits may be genetically controlled. Clones spent more time in proximity to adult animals in an adjacent pen (which also housed the nuclear donor), and in proximity to the feed bunk compared to control animals. In general, clones were reported to spend more time with each other rather than socializing with control animals. The authors speculated as to whether clones exhibit genetic kinship recognition.

Batchelder (2005) reported aggressive feeding behavior and “insatiable” appetites among eight juvenile clones, as well as increased water consumption vs. ET and AI comparator calves. The clones’ increased demand for water and milk replacer may be related to the higher body temperatures experienced by clones in this study during the first 60 days, and may represent a compensatory response to maintain hydration. Blood values for these older calves were not reported so it cannot be determined whether some other underlying metabolic disturbance (such as differences in energy metabolism) might have contributed to the increased appetite and thirst exhibited by clones. Increased appetite, growth rate and altered glucose metabolism has also been noted in IVP and BNT calves (Garry et al. 1996, Rerat et al. 2005). Overall, no differences were observed in weight gain between clones and comparators in this study; however, when data were analyzed by breed, Hereford clones gained more rapidly than Hereford comparators, while Holstein clones gained less than Holstein comparators during the first four weeks after birth.

The potential for long term effects of embryo manipulation on the resulting animal is a question that has arisen in the past. McEvoy et al. (2000) noted that IVP calves exhibiting LOS at birth (greater than 60 kg) were not different in body weight compared with *in vivo* produced, normal birth weight controls when slaughtered at 13 months of age. LOS cattle had abnormally large hearts when necropsied at slaughter, although this study did not discuss whether the enlarged hearts showed any other anomalies which might indicate functional abnormalities. Rerat et al. (2005) noted that although birth weights were similar between AI (n=8) and IVP (n=11) calves, IVP calves ate more and grew faster throughout the study, and were heavier than AI controls beginning at eight weeks of age and persisting through 16 weeks, when the study ended. Wilson et al. (1995) compared birth weight and growth of calves derived by AI and natural mating (NM) to half- and full-sibling calves from ET or BNT. Male and female calves resulting from BNT had higher birth weights (49.5 vs. 39.9 and 36.8 kg for male BNT, ET, and AI/NM; 47. vs. 37.1 and 34.6 kg for female BNT, ET, and AI/NM, respectively). BNT calves were also heavier than ET and AI/NM calves at 1 year of age (519.0 vs. 497.4 and 497.0 kg for male BNT, ET and AI/NM calves; 429.1 vs. 356.3 and 352.9 for female BNT, ET, and AI/NM calves, respectively). No other physiological measurements were taken in the Wilson et al. study, and health of calves was not discussed. In contrast to the Wilson et al. study, Pace et al. (2002) reported that 52 SCNT clones (some transgenic) raised at the same facility had similar weight gains for the first 120

days, regardless of birth weight. No comparisons were made with contemporary controls in this study, and no other physiological measurements for this age were reported.

Of the six calves surviving the neonatal period in the Batchelder (2005) study, three more calves died or were euthanized during the juvenile period. Two calves died due to complications involving a non-healing umbilical stalk and patent urachus. Another calf died of apparent pneumonia, and was diagnosed with cardiac abnormalities and pulmonary hypertension upon necropsy. Two of the calves exhibited neurological signs, including head twitching and seizures. Three clones (Holstein breed) and all nine comparators survived the juvenile period.

ii. **Cyagra Data: 1-6 Month Cohort**

According to the data provided by Cyagra (Appendix E), nine calves out of 104 died after the critical 48 hour period after birth. Of these, three died or were euthanized during the juvenile or weaning period. Causes of death for these three animals varied. One clone died at 47 days after it failed to respond to therapy to treat severe contracture of the limbs. Another clone was diagnosed with “failure to thrive,” indicating that it failed to gain weight or grow properly. The third clone died at 149 days as the result of gastrointestinal tract problems such as bloating and poor rumen motility.⁶⁵ One clone that survived to weaning was culled for poor conformation. This type of culling is often seen in conventional breeding programs.

Health issues observed in some of the Cyagra clones (see Appendix E) included an increased incidence of umbilical problems (enlargements, excessive bleeding, navel infection), contracted tendons, and cryptorchidism (a condition in which one or both testicles are retained in the body cavity). All of these conditions are seen in sexually-derived animals, but at lower frequencies than in clones. For example, calf clones had umbilical surgery at a much higher rate than non-clone calves. Contracted tendons appeared to occur at a higher incidence in clones relative to sexually-derived animals. Cryptorchidism is quite uncommon in sexually-reproduced bulls, but does appear to be an inherited trait (Blood and Radostits 1989); cryptorchid bulls tend to sire cryptorchid calves, are not recommended for breeding stock, and are thus directed to food production. The health risk posed to the animal by cryptorchidism is the tendency for the retained tissues to become neoplastic (tumor-forming) in long-lived animals.

There was no evidence, based on hemoglobin and hematocrit, of anemia in clones (Appendix E). One clone calf had a hemoglobin count which was above the range for the comparator group, but that observation does not appear to have any health consequences. Other red blood cell measures, such as low mean cell hemoglobin concentration (MCHC) and red cell distribution width (RDW)

⁶⁵ In ruminants, the rumen or largest compartment of the stomach contracts on average once per minute. This is necessary to aid digestion and normal passage of nutrients through the gastro-intestinal tract.

do not indicate a disease process in the absence of indicators of anemia such as low hemoglobin or hematocrit (these measures were within the normal range). White cell measures were generally within the normal range, and there were no differences that could indicate infection or abnormalities in immune function.

As discussed in Appendix E, physiological indicators of growth such as calcium, phosphorus, and alkaline phosphatase were appropriately elevated relative to expected adult levels in both clones and comparators. A few clones exhibited elevated levels of these growth indicators relative to comparators. Although these differences may be attributed to differences in management, it is important to note that the animals in which these elevations occurred were among the youngest in this age cohort. Further, these clones are likely the product of superior genetics bred for improved growth and production characteristics.

Although six of the 42 valid glucose values (some measurements were considered artifactual (see Appendix E)) were higher for clones than comparators, none of the urinalyses were positive for glucose. The elevated glucose is not considered to be clinically relevant as the absence of urinary hyperglycemia indicates that the elevated blood glucose levels were transient, and most likely a short-lived response to stress.

Gamma glutamyl transferase (GGT) levels in three clones were lower than in comparators, and sorbitol dehydrogenase (SDH) levels were lower in three other clones than comparators. Although high levels of these enzymes are indicative of liver damage, the clinical significance of low levels is not known.

iii. Unpublished data

A private veterinary clinic submitted hematology and clinical chemistry data on blood samples from three bull clones. Three samples from each bull were taken over a six week period when the bulls were between five and seven months old. Results of these samples are presented in Appendix G. One bull clone had low cholesterol on the second sampling date, but on the first and third sampling, values were within published ranges. All of the other clinical chemistry and hematology values were within published ranges for cattle.

From the data submitted by J.L. Edwards at the University of Tennessee (Appendix G), 12/27 live-born calves (44 percent) survived the Juvenile Period. One calf died at nine months of age from enterotoxemia due to infection with Type A *Clostridium perfringens* (Lawrence et al 2005). Another six calves died between the ages of 41 days to eight months. Clinical signs observed in these calves included gastrointestinal problems (scouring, rumen stasis, acidosis, anorexia), severe weight loss, pneumonia, umbilical abscess, and hepatic defects.

d. Developmental Node 4: Reproductive Development and Function**i. Peer-Reviewed Publications**

As mentioned in Section 2.a.i., overall, fertility rates among conventionally bred cattle have declined in recent years, particularly among dairy cattle. Heifers generally have higher conception rates compared to multiparous older cows. Even so, a recent survey of 2,688 U.S. dairy herds indicated that overall conception rates among Holstein heifers were only 57 percent (Kuhn et al. 2006) compared to earlier reference texts (*e.g.*, Knobil and Neill 1998), which estimated pregnancy rates among dairy heifers at 90 percent. Part of this difference may be an artifact of the different definitions of conception rate and pregnancy rate⁶⁶ or the use of a single breed in the more recent study by Kuhn et al.

Heyman et al. (2004) reported that female clones began cycling at about 10 months of age, and exhibited estrous behavior by 12 months of age, within the normal range for Holstein heifers. Ten female clones were bred by AI to a non-clone bull, and all conceived and produced live, apparently normal progeny. Birth weight of progeny was 43.9 ± 4.1 kg, and gestation length was 281.1 ± 3.9 days, within the normal range for Holstein cattle.

Pace et al. (2002) reported that clone Holstein heifers (some transgenic) reached puberty at approximately 10 to 11 months of age, within the normal range for their breed. They also reported that all 22 clone heifers were inseminated and diagnosed pregnant (Table V-6). The heifers calved between 23 and 25 months of age, within the recommended range for first parity Holsteins; no details were reported in this study.

Enright et al. (2002) reported on the same set of animals as Govoni et al. (2002) and Savage et al. (2003). Four non-transgenic Holstein heifer clones of a single donor animal were compared to age and breed matched heifers derived from AI. The heifer clones reached puberty at a later age than controls (314.7 ± 9.6 days vs. 272 ± 4.4 days), and had higher body weights at first estrus (336.7 ± 13 vs. 302.8 ± 4.5 kg). No differences were noted between clones and controls in estrous cycle length, development of ovarian follicles, or profiles of hormonal changes. Three of the four clones and all four control heifers became pregnant following AI. Daily hormone profiles of lutenizing hormone (LH), follicle stimulating hormone (FSH), estradiol-17 β , and progesterone were similar between clones and controls. The cause of reproductive failure in the

⁶⁶ Pregnancy rate is defined as the fraction of animals confirmed pregnant per total number of animals in the breeding group (*i.e.*, animals available for breeding). Because so many dairy cattle are bred by AI, another useful term is of conceptions per insemination, where conception rate is defined as the fraction of animals conceiving per number of animals inseminated. Because most beef cattle are bred naturally, the number of times a cow is bred is seldom recorded, so the more general term “pregnancy rate” is used. In this risk assessment, “pregnancy rate” is used for both dairy and beef cattle, regardless of method of breeding.

one clone could not be determined. Reproductive hormone profiles in this heifer were similar to the other animals in the study, and no physical abnormalities could be found upon veterinary examination, although poor signs of estrus were observed. The later age and higher weight of clones at time of puberty relative to the controls may have been under genetic control as all four animals were derived from the same cow, although no records of age at puberty were kept for the source cow. Average age and weight were higher in clones compared to the comparator animals in this study, although they were similar to ranges previously reported for conventional Holstein heifers (Murphy et al. 1991, Radcliff et al. 1997).

Study	Transgenic Status	Clone Pregnancy Rate (fraction)	Comparator Pregnancy Rate (fraction)	Comments
Edwards (unpublished)	None	6/6 (1.00)	NP	Two of six pregnancies resulted in live calves
Enright et al. 2002	None	3/4 (0.75)	4/4 (1.00)	Cause of pregnancy failure in one clone not determined
Heyman et al. 2004	None	10/10 (1.00)	NP	
Kuhn et al. 2006	NA	NA	0.57	Holstein heifers only; based on data from 362,512 heifers
Knobil and Neill 1998	NA	NA	0.90 beef 0.95 dairy	Based on average pregnancy rates for replacement heifers
Lanza et al. 2001	All	24/24 (1.00)	NP	Conception rate to first AI 87.5%, remaining pregnant after second AI
Pace et al. 2002	Some	22/22 (1.00)	NP	
Panarace et al. 2007	None	74/74 (1.00)	NP	Reported for primi- and multiparous clones; results for one or two inseminations by AI or NM.
Wells et al. 2004	None	25/30 (0.83)	9/10 (0.90)	Conception rates to two AI services
NP = not provided NA = not applicable				

In a study of a cohort of transgenic cattle clones, Lanza et al. (2001) reported that 24 heifer clones of various breeds reached puberty between 10 and 12 months of age, with body weights ranging from 318 to 365 kg. Twenty-one of the 24 heifers conceived with the first insemination, and the remaining three conceived at the second insemination. At the time of publication, two calves had been born, and were reported as healthy.

A larger study (Panarace et al. 2007) reported breeding success for 74 heifer and cow clones bred by AI or natural mating, and stated that all 75 animals conceived after one or two inseminations/matings. The authors reported three spontaneous abortions at five and seven months of gestation and two cases of dystocia among the 74 pregnant clones. As of date of publication, 42 clones had given birth to viable calves. In the same study, 21 heifer clones were enrolled in a superovulation program for embryo transfer. The clones averaged 4.5 viable embryos/flushing, and were not different from control heifers. Also, these authors report that 54 bull clones were evaluated for semen quality (progressive motility, morphology and concentration) and were judged acceptable according to standard requirements. Table V-6 indicates pregnancy rates for cattle clones and comparators.

Forsberg et al. (2002) reported that a bull clone had matured into a “*healthy, fertile bull that has sired calves by artificial insemination and in vitro fertilization,*” although data were not provided. Kato et al. (2000) report that one of the clones derived from a Holstein cumulus cell was artificially inseminated, conceived, and gave birth to a normal calf.

In a recent study, Shiga et al. (2005) reported on the semen quality of two clones of a 12 year old Japanese Black bull. Semen was collected beginning when the bulls were 12 months old, and the study followed these animals through 16 months of age. Ejaculate volumes were similar between the two bulls (2.34 and 2.76 mL) but were lower than the range for conventional Black bulls (5-8 mL). However, sperm concentration (1,202 and 834 x 10⁶/mL), pH, and pre-freezing motility (71.4 and 66 percent) were within established ranges for conventional bulls of the same breed. Fertility testing using *in vitro* fertilization was performed, comparing the two clones to the donor bull. Development of embryos to blastocyst was not different between the clones and their donor (23.4 and 28.4 vs. 30.9 percent). Because few cows were available for breeding, only one clone was used to compare pregnancy rates to AI with the donor bull. Only 22 cows were inseminated by the clone, compared to 102 cows inseminated by the donor. Nonetheless, pregnancy rates were similar between the clone and the donor bull (54.5 vs. 62.7 percent). Two of the twelve resulting pregnancies to the clone ended in spontaneous abortion in mid-pregnancy, compared to five spontaneous abortions (5/64) for cows pregnant by the nuclear donor.

Similarly, Wells et al. (2004) reported that rates of development to blastocyst for embryos fertilized *in vitro* by sperm from six bull clones were similar to blastocyst rates for four non-

clone bulls (range 10-25 percent for clones vs. 13-30 percent for comparators). In the same report, Wells et al. stated that pregnancy and calving rates of heifer clones following two rounds of AI were 83 percent (25/30), and were only slightly less than for a small group of conventional heifers (9/10, 90 percent). Gestation length was slightly longer for a group of 16 clones compared to nine comparators (287 ± 3 vs. 281 ± 3 days), but still within the range for the breed (Friesian). The heifer clones calved spontaneously with only minor assistance. Clones exhibited normal maternal behavior, and bonded with their calves.

Tecirlioglu et al. (2006) also evaluated semen quality and fertility of three clones and their Holstein-Friesian nuclear donors. All three clones were derived from skin fibroblasts. Two of the clones (A-1 and A-2) were derived from a six-year-old bull (donor A) that was euthanized shortly afterward due to a spinal injury. The third clone was derived from a second donor (donor B, an eight-year-old bull) by SCNT. The embryos were frozen and thawed before transfer to recipient heifers. The donors and their clones were housed under similar conditions. Semen collection and freezing was performed at the same commercial facility for all five bulls according to routine procedures established by the facility. Semen was evaluated for volume, concentration, total and progressive motility, total, primary and secondary abnormalities. Semen was also used to evaluate *in vitro* fertilization (cleavage rate, blastocyst rate, inner cell mass, trophectoderm⁶⁷ cell count, and total cell count). Differences in semen volume were inconsistent, with Donor A and Clone B-1 having lower volumes than Clones A-1, A-2 and Donor B. However, sperm concentrations and motility measures were similar among clones and donors. Sperm morphology (measures of abnormalities) was within acceptable limits for all five bulls (> 80 percent morphologically normal sperm). *In vitro* fertilization rates (expressed as cleavage rate by 48 h) were higher for clones than for their donors; however, there were no differences in rates of blastocyst formation between clones and donors, except Clone A-2, which had a higher development to blastocyst than the other four bulls. There were no differences in embryo quality (as measured by cell counts of inner cell mass (ICM), trophectoderm (TE) and total cell number) between embryos of clones and donors. Pregnancy rates and pregnancy losses were compared only between Donor B and Clone B-1. Individual IVF embryos from Donor B (n=40) and Clone B-1 (n=37) were transferred to synchronized heifers. Heifers were examined for pregnancy at 30, 120 and 240 days of gestation. Number of live calves and calf survival per sire were also reported. Pregnancy rates at day 30 were similar between donor (16/40) and clone (17/37), as were pregnancy losses measured from 30 to 240 days of gestation (2/40 vs. 5/37). All heifers pregnant at day 240 delivered live calves, and there was no postnatal mortality. The authors noted that there were no phenotypic abnormalities among the progeny, even though all the calves were produced via IVF and delivered by C-section.

⁶⁷ In the blastocyst stage of development, embryonic cells segregate into two types: the inner cell mass is destined to become the fetus, while the trophectoderm cells become the placenta.

In a companion study to Chavatte-Palmer et al. (2004), Heyman et al. (2004) reported on reproduction and lactation of 17 Holstein clones compared to age-matched, AI-derived half-siblings maintained under the same conditions. Only preliminary data from three females were available at the time of publication. The authors reported that milk yields were similar for the three clones compared to the non-clone comparators (9341 ± 304 vs. 8319 ± 1800 kg for 305 day lactation). Somatic cell counts (SCC, an indicator of mammary gland health) were assessed monthly throughout the first lactation in clones and comparators. Mean SCC for clones and comparators were $116 \pm 103 \times 10^3$ vs. $113 \pm 50 \times 10^3$ cells/mL, and were not statistically different. These values are well below the point at which animals would be judged to have subclinical mastitis ($1,000 \times 10^3$ cells/mL).

Tian et al. (2005) also reported first lactation milk yields and SCC for four clones and their non-clone comparators, indicating that lactation curves were similar for both groups. Total milk production for the first lactation was not different between clones and comparators (8646 ± 743.8 vs. 9507.8 ± 743.8 kg). One clone gave birth prematurely to a stillborn calf, did not have complete udder development, and produced approximately 30 percent less milk during her first lactation compared to her clone mates. Typically, the udder develops during the last month of pregnancy, so lack of complete development following premature delivery is not unusual. Overall, SCC was low for both clones and comparators (based on Figure 2b of the paper: $\sim 40 \times 10^3$ vs. 35×10^3 cells/mL).

Heyman et al. (2004) also reported on the fertility and reproduction of male clones. Three clones of an eight year old bull were enrolled in an AI center at approximately 12 months of age, and semen collection was initiated when the clones were between 13 and 15 months of age. Clones and their semen were handled the same as other bulls at the center. Sperm from the three clones and the nuclear donor were compared. Percentages of normal sperm were not statistically different between the nuclear donor and the three clones (See Table V-7). Preliminary IVF trials using the sperm of these four animals indicated that a lower concentration of sperm from the clones was needed to achieve a similar fertilization rate to that of the nuclear donor. Given the age of the nuclear donor (approximately 9 years old at the time of this phase of the study), this is not surprising. Cleavage rate and development to blastocyst were not statistically different between the nuclear donor and the clones (see Table V-7). Only one of the clones (Clone #2) was used for comparison of AI with the nuclear donor. Overall pregnancy rate for the clone was 65 percent (41/63 inseminated). Two pregnancies were lost by day 90 of gestation, representing a 5 percent loss, similar to that reported by Thompson et al. (1998) for IVF embryos. Only 26 of the pregnancies were allowed to go to term, resulting in 25 live calves, and one premature stillborn. Average birth weight of the progeny was 36 ± 2 kg, below average for the breed. The authors attributed this to the fact that the nuclear donor was considered an “easy calving” bull, indicating that he was selected because of his ability to produce smaller calves.

Table V-7: Results of IVF for nuclear donor and three clone bulls.			
Animal	Normal sperm (%)	Cleavage rate¹	Blastocyst rate
Donor	86.5	273/363 (75.2)	108/273 (39.6)
Clone 1	85.0	244/386 (63.2)	93/ 244 (38.1)
Clone 2	77.5	207/381 (54.3)	72/207 (34.8)
Clone 3	67.0	196/359 (54.6)	80/196 (40.8)

¹ Cleavage rate and blastocyst rate are expressed as number of embryos cleaved or forming blastocysts/ total number of embryos. Percentages are in parentheses.

ii. Unpublished data

Data on two clone bull cohorts were submitted directly to CVM for use in this risk assessment. These are discussed in Appendix G. Briefly, one dataset represents semen characteristics of four bull clones collected by a commercial reproduction service over a two month period. Age of the bulls at time of collection was not provided. Data were not consistently collected, in that some of the values, such as sperm motility, were not recorded for all bulls at all collection points. Two of the bulls appeared to produce acceptable semen, one bull produced poor to marginal semen, and one bull produced poor quality semen. The second dataset provided breeding soundness data on three bull clones aged 18 to 24 months old. As with the previous dataset, some data were missing. However, the available data indicated that scrotal circumference and semen quality for these bulls were acceptable. Semen from one of the clones was used in both IVF and AI programs to assess fertility. Fertilization rate for IVF was 75 percent for this clone, and overall pregnancy rate to AI was 65 percent, with a five percent pregnancy loss.

In the data submitted by the Edwards laboratory (Appendix G), information on reproductive function was provided for six clones. Of these six, two delivered healthy, normal calves, but the other four pregnancies resulted in stillbirths.

e. Developmental Node V: Post-Pubertal Maturation and Aging

i. Peer-reviewed literature

There are limited data on concerns related to aging and longevity of conventionally-derived cattle. As mentioned previously, McEvoy et al. (2000) noted that IVP-derived calves diagnosed as suffering from LOS at birth had abnormally large hearts compared to *in vivo* derived calves when slaughtered at 13 months of age, although the LOS calves appeared to grow normally and were not larger than *in vivo* controls. This group questioned whether the *in vitro* process could affect long-term animal health and longevity. No further details were available to indicate

whether the enlarged hearts were functionally abnormal, nor were any follow-up studies identified.

Wells et al. (2004) conducted a retrospective analysis of cattle clones that were generated through SCNT at AgResearch in New Zealand to determine their long-term survival. They found that 133 (13 percent) calves were born from 988 SCNT embryos transferred into recipient cows. Sixty seven percent of these calves (89 animal clones) survived to weaning (3 months of age) and 81 percent of the calves (72 animal clones) survived post-weaning, with the oldest animal being 4 years of age at the time of publication of their article. They estimated the annual mortality rate in cattle cloned from somatic cells to be at least 8 percent. The reasons for death were variable, including euthanasia due to musculoskeletal abnormalities (4 animals), bloat (2 animals), ryegrass staggers (2 animals), misadventure (2 animals) and one case each of anemia, heart failure, kidney failure, ruminal acidosis, lungworm, clostridia, and overfeeding on grain supplement. Some of these deaths were preventable. The musculoskeletal abnormalities included animals with severely contracted flexor tendons and those displaying chronic lameness, particularly in milking cows. In surviving cattle clones, blood profiles and other indicators of general physiological function such as growth rate, reproduction, rearing of offspring, and milk production were all within the normal phenotypic ranges.

Wells et al. (2004) also reported on hematology and clinical chemistry of nine heifer clones and their progeny at two years of age. Heifer clones were compared to nine non-clones and a published reference range. Of the 13 hematological values measured, clones were within the range of their contemporary comparators for nine values. White blood cell counts (4.57 ± 0.48 vs. $6.91 \pm 1.16 \times 10^9/L$), lymphocytes (3.13 ± 0.31 vs. $4.93 \pm 0.92 \times 10^9/L$), and eosinophils (0.11 ± 0.09 vs. $0.32 \pm 0.18 \times 10^9/L$) were lower for clones than contemporary comparators, but within the published range. Basophil counts were higher for clones than comparators (0.08 ± 0.07 vs. $0.01 \pm 0.03 \times 10^9/L$), but still within the reference range. Of the 15 clinical chemistry values analyzed, 14 of the values for clones were within the range of their contemporary comparators. Creatine kinase, the only value that was outside the comparator range (191 ± 136 vs. 112 ± 76.4 IU/L) was within the published reference range (0-370 IU/L).

The Chavatte-Palmer et al. (2004) study reported that of the 38 clones surviving the juvenile period (which they defined as past six months of age), 36 were still alive among the older clones (aged 15 months to four years). Cause of death was reported for only one of the clones that died: apparent heat stress during an unusually hot summer, approximately one week after her second calving. Twenty of the clones are currently enrolled in a long term health study with 20 AI comparators. This study has not been completed, but preliminary reports indicate that so far the only observed clinical sign has been fungal lesions of the skin, which have occurred in both

clones and controls. The authors state that they have not yet evaluated whether the lesions occur more frequently in one group or the other.

Batchelder (2005) reported the sudden death of a Holstein heifer clone at 25 months of age. On necropsy, the heifer was diagnosed with severe trace mineral (selenium and copper) deficiency. Other cattle grazing the same pasture were clinically normal. As a young animal, this clone was reported with frequent, mild left-sided bloat. Clones in this and other studies (Wells et al. 2004; Cyagra 2003: Appendix E) have also been reported with bloat and other gastro-intestinal tract problems. Two other clones in the Batchelder (2005) study were reported as healthy at 19 months of age. These two surviving animals required little supportive care at birth; however, both animals required umbilical surgery.

Panarace et al. (2007) stated that eight adult clones (male and female, although proportions of each gender were not specified) died of various causes. Only three causes of death were specifically cited: uterine torsion, broken leg, and back problems. It is not clear from this report how many animals died from each cause or what other causes were identified.

Many questions have been raised regarding the immune function of clones and their ability to resist or recover from disease, yet few studies have examined this issue directly in bovine clones. Most information regarding immune responsiveness in clones have been derived from data on WBC counts and differentials, and more general reports on long-term health and viability of cattle clones. One such study (Tanaka et al. 2006) found no differences in proportions of granulocytes, monocytes, B cells, total T cells, and most T cell subsets between six lactating Holstein clones and five non-clone comparators. A difference was detected in proportions three T cell subpopulations ($CD4^+$, $CD8^+$, and $WC1^+\gamma\delta$) in clones in early lactation compared to non-clones, although the differences disappeared in mid-lactation. Milk yield of the clones was higher than yield of non-clones throughout lactation, but the difference was particularly noticeable during early lactation, with clones reaching peak milk yield at approximately 45 kg/day, whereas non-clones peaked at approximately 32-35 kg/day. It is expected that high yielding cows may experience some immune suppression due to the stress of high production, so the slight differences noted in this study between certain T cell subsets between a group of high producing clones compared to much lower producing comparators may be more of a reflection of the differences in production, rather than a function of being clones. The study would have been more informative had the production levels of the clones and comparators been more similar.

One interesting study was found recently which may shed some light on the immune function of adult cattle clones. Theoret et al. (2006) examined the reaction of three 18 month old male cattle clones to skin grafts from each other and an unrelated bull. The report does not specify whether the clones were bulls or steers. The clones were all derived from fibroblast cells of the same

Holstein X Zebu fetus, but oocytes were derived from three different Holstein cows. Thus, the clones had the same nuclear DNA, but mitochondrial DNA (mtDNA) was a mixture of mitochondria from the fetal cell and oocyte donors. The primary purpose of the study was to test the role of mitochondrial proteins in host/graft rejection. Mitochondrial DNA from the fetal cell donor was determined to contribute only 1 to 2.6 percent of mtDNA to the skin of the clones; thus the majority of mtDNA was contributed by the oocyte. Each animal received an autograft (a graft from their own skin) and a graft from each of the other animals, such that each clone received grafts from its fellow clones and from the unrelated bull. None of the four animals rejected the skin grafts during the acute phase; however, the unrelated control bull rejected grafts from clones within four weeks, whereas the clones rejected grafts from the unrelated control bull after six weeks. The clones appeared to accept grafts from each other for the duration of the study (13 weeks).

ii. Cyagra Data: 6-18 Month Cohort

Both the veterinary examination and laboratory data indicated that clones in the 6-18 month cohort were healthy and normal, and that in general, laboratory values for this age cohort were more similar to the Cornell laboratory reference range than the younger age groups (see Appendix E).

iii. Unpublished data

Certificates of Veterinary Inspection and results of serological testing and hematology for two heifer clones aged approximately 14 months were submitted directly to CVM in response to our request for data. These results and the table of values for clinical chemistry and hematology for these two heifers are presented in Appendix G. Briefly, red cell distribution width was the only value outside the laboratory reference range for these heifers. All other values were normal.

The data submitted by J.L. Edwards (Appendix G) indicated that of the Jersey clones surviving the juvenile period, one died at one year of age and a second was euthanized at 2.5 years of age following periodic scours that were unresponsive to treatment. Five additional clones were described as healthy, and no unusual pathological findings were noted at necropsy following euthanasia at four years of age (as part of a herd reduction program). Seven clones in the Edwards cohort are listed as still alive and healthy at four years of age.

f. Progeny of Bovine Clones

Many clones have been bred and have been at least reported as having given birth. Lanza et al. (2000) reported that a transgenic cow clone had given birth, and her offspring was growing

normally. Both the University of Connecticut (Enright et al. 2002) and Infigen groups (Pace et al. 2002) have reported breeding and subsequent calving of several of their cow clones, but no information on the health status of these progeny has been made available.

In a presentation to CVM in 2003, Galli et al. (unpublished; see Appendix G) provided limited data on three progeny of bull clones. The three calves (two female and one male) were born following normal gestations (271 to 280 days) and were within the normal birth weight range for Holstein cattle (42 to 45 kg). The presenters stated that they observed no abnormalities in the progeny of the bull clones.

Shiga et al. (2005) reported on 10 calves of a Japanese Black bull clone. The clone produced three female and seven male calves by AI. Female progeny weighed on average 33.2 ± 2.0 kg, and male progeny weighed 32.3 ± 4.1 kg at birth, within the normal range for Japanese Black cattle, and similar to the birth weights of calves of the nuclear donor (30.7 ± 3.5 kg for females and 34.0 ± 4.9 kg for males). No additional information was available on the health and development of the progeny in this study.

Wells et al. (2004) produced 52 progeny of clones via natural mating or AI with conventionally bred bulls. According to their report, 85 percent of these calves were alive at 24 hours after birth, compared to 84 percent (27/32) for contemporary comparators. All progeny calves were described as “phenotypically normal.” Wells et al. noted that this was similar to survival rates of clones within 24 hours after birth, but the progeny required less care than clones. Only one progeny animal was lost after the first 24 hours. Hematology and clinical chemistry of 15 progeny between the ages of one and three years were compared to conventionally derived age-matched cattle and the same published range that was used for comparing two-year-old clones. One limitation of these data is that only three contemporary comparator animals were used, thus possibly failing to capture the natural variability among cattle. Nine of 13 hematology values for progeny of clones were within the range of the three comparators. The four values that fell outside the range were MCV (44.6 ± 5.39 vs. 49.7 ± 1.53 fL), MCH (16.1 ± 1.92 vs. 17.7 ± 0.58 pg), MCHC (363 ± 11.5 vs. 354 ± 2.00 g/L), and eosinophils (0.22 ± 0.23 vs. $0.31 \pm 0.13 \times 10^9/L$), however, these differences were small. All hematology values for progeny of clones fell within the published range. For a discussion of the diagnostic value of these measurements, see Appendices E and H. Eleven of 15 clinical chemistry values for progeny fell within the range of contemporary comparator values. Creatine kinase (136 ± 98.1 vs. 168 ± 133 IU/L), AST (45.7 ± 7.49 vs. 36.7 ± 6.43 IU/L), and GDH (8.40 ± 6.99 vs. 3.33 ± 0.58 IU/L) were higher in progeny of clones, and creatinine (101 ± 37.3 vs. 138 ± 11.5 μmol) was lower in progeny than contemporary comparators. All clinical chemistry values for progeny of clones were within the published range. Interestingly, the average creatinine value for comparators was slightly higher than the published range (138 ± 11.5 vs. 55-130 μmol).

Similar to Wells et al. (2004), Heyman et al. (2004) reported that 25 live-born progeny of a clone bull were physiologically normal at birth. Progeny calves and their placentae did not exhibit any of the phenotypic defects sometimes noted for clones. Progeny of 10 female clones bred by AI to a non-clone bull weighed on average 43.9 ± 4.1 kg at birth, within the range of birth weights for their breed. All progeny calves were alive and appeared normal. Additionally, three female clones were bred by natural service to a male clone and also produced three apparently normal calves.

Ortegon et al. (2007) reported on the health and development of 30 progeny calves (19 females and 11 males) of Starbuck II, a Holstein clone bull used to breed conventionally produced Holstein cows by AI. The calves were monitored for the first 12 months of life and compared to age- and breed-matched calves of conventional ancestry. Heart rates, body temperatures, respiration rates of the progeny of clones were determined to be normal, as were mucous membranes, lymph nodes, cardiac and respiratory sounds on auscultation. The only illness reported was diet-induced diarrhea in two progeny heifers; however, the same condition was reported to occur more frequently and with greater severity in comparator calves, although the paper does not discuss specifics of these cases. Both groups achieved puberty between 10 and 12 months of age and within the expected weight range (318 – 365 kg). Genitalia were reported as normal based on visual inspection and rectal palpation, and female progeny had similar levels of plasma progesterone compared to conventional heifers. Five clone-sired heifers were bred by AI to the same commercial bull, and two were confirmed pregnant 33 days after insemination.

Kasai et al. (2007) described growth performance in a heifer calf produced by a mating in which the dam and sire were both clones. The calf was produced by mating the female cow clone to the male bull clone by artificial insemination. Comparators in this study were seven full sibling sisters (daughters of the nuclear donors) produced by mating the female nuclear donor to the male nuclear donor by artificial insemination. Comparator embryos were flushed from the nuclear donor, frozen, and later transferred to recipients.

Following a 292-day gestation (11 days longer than the average for comparator pregnancies), the female clone cow delivered the calf of the clones without assistance. Birth weight, clinical examination, hematology, serum biochemistry and telomere length of the calf of the clones were within the ranges measured for the comparator calves. Growth parameters from birth to 12 months, as measured by body weight and shoulder height, were similar in the calf of the clones and the comparators. No serious health problems were observed in the calf of the clones. At 18 months of age, the calf was artificial inseminated and was four months pregnant at the time of publication. To the authors' knowledge, this was the first report demonstrating normal growth performance in a calf derived from mating two clones. These results are consistent with those of

Heyman et al. (2004), who did not observe LOS in offspring produced by mating either pairs of clones or clones to non-clones.

g. Summary for Health of Bovine Clones and Their Progeny

Based on a review of the literature, the SCNT process in cattle is associated with increased incidences of early pregnancy loss or later-term spontaneous abortion of clone embryos and fetuses. Identified hazards for surrogate dams of bovine clones are hydrops and dystocia. The risk of developing either of these complications appears to be both species- and laboratory/cell line-dependent. Not all cases of hydrops in clone-bearing pregnancies develop into a significant complication or threat, but severe, and possibly even moderate, hydrops conditions, when not diagnosed early, may result in the death of the surrogate dam and the clone. Large Offspring Syndrome increases the risk of dystocia (difficult birth), and may be related to the development of hydrops (abnormal fluid accumulation (edema) in one or more compartments of the placenta and/or fetus).

Neonatal death rates for cattle clones currently average approximately 20 percent. Dystocia may be the most influential factor on calf mortality, due to trauma of difficult labor and emergency C-section; however, abnormal organ and musculoskeletal development also appear to play important roles. Congenital organ defects are observed in some neonatal clones that do not survive; these also occur spontaneously at much lower frequencies in conventionally bred cattle and other farm animals (Szabo 1989). There is some evidence to indicate that organ abnormalities in calf clones may be the result of aberrant epigenetic reprogramming leading to dysregulated expression of genes that control organ formation and development.

During the juvenile period (up to approximately six months of age), bovine clones continue to be at an increased risk of morbidity or mortality compared to animals produced by natural service or ARTs. Estimates of mortality during this period range from 14 to 42 percent. These deaths appear to be sequellae of the initial developmental abnormalities noted in the perinatal node that persist into the juvenile period (*e.g.*, musculoskeletal defects, prolonged recumbency, enlarged umbilicus, respiratory distress, poor thermoregulation, cardiovascular failure, gastroenteritis). It is likely that these developmental abnormalities result from faulty epigenetic reprogramming. Most of the approximately 58-86 percent of calf clones that are not affected by fatal abnormalities during the first six months of life appear healthy, demonstrate normal patterns of growth and development, and complete the juvenile period without further complication.

The available data on reproductive performance of cattle clones suggests that there are no adverse effects on their reproductive health. Of seven reports of reproduction in clone heifers,

five studies indicated 100 percent pregnancy rate; pregnancy rates in the other two studies were within the range of historical values (≥ 75 percent). Similarly, several studies have demonstrated that clone bulls are fertile, and that the quality of their semen and rates of pregnancy and calving following AI are similar to comparators (either nuclear donor or conventional bulls).

Data on post-pubertal maturation and aging indicate that as surviving clones approach maturity, they are as healthy as their non-clone comparators. Among older clones that die or are euthanized, health problems appear to be related to pre-existing conditions (musculo-skeletal defects, GI tract problems) already identified during the perinatal and juvenile periods.

Progeny of cattle clones produced by normal sexual reproduction appear to be normal and healthy. Offspring of clones do not exhibit LOS and appear to grow and develop normally. These observations support the idea that epigenetic reprogramming errors in clones are reset during gametogenesis, thus precluding transmission of anomalies from clones to the next generation and are consistent with the observations of Tamashiro et al. (2002) in mice, which indicate that anomalies present in clones are not transferred to their sexually reproduced progeny.

2. Swine

Survival of live-born swine clones from various studies is summarized in Table V-8. As with cattle, relatively few studies included contemporary comparators, so historical data from various references and data bases were also incorporated into the table to provide context.

Reference	Transgenic Status	Surviving/Total Live-Born Clones (fraction)¹	Surviving /Total Live- Born Comparators (fraction)	Comments
Bethhauser et al. 2000	None	4/4 (1.00)	NP	
Bondioli et al. 2001	All	2/2 (1.00)	NP	Described in Chapter VI
Boquest et al. 2002	None	1/2 (0.50)	NP	
De Sousa et al. 2002	None	1/1 (1.00)	NP	
Lagutina et al. 2006	None	11/14 (0.79)	NP	
Onishi et al. 2000	None	1/1 (1.00)	NP	

Table V-8: Survival Rates of Live-Born Swine Clones and Comparators				
Reference	Transgenic Status	Surviving/Total Live-Born Clones (fraction)¹	Surviving /Total Live- Born Comparators (fraction)	Comments
Polejaeva et al. 2000	None	5/5 (1.00)	NP	
Walker et al. 2002	None	27/28 (0.96)	NP	
Yin et al. 2002a	None	8/8 (1.00)	NP	Described in Chapter VI
USDA/NAHMS 2001 (6/00 – 7/00) ³	NA	NA	0.89	Historical data from animals produced by AI and natural service in commercial operations

¹ Survivors through the Juvenile Period/Live births
² NP = not provided; data not available
 Transgenic Status: All = All of the clones cited in the publication are derived from transgenic donor cells, Some = Some of the clones cited in the publication are derived from transgenic donor cells, None = None of the clones cited in the publication were derived from transgenic donor cells.
³ <http://nahms.aphis.usda.gov/>

a. Developmental Node 1: Pregnancy and Parturition

i. Pregnancy

In swine, a litter-bearing species, at least four viable embryos are needed during early gestation for the sow to carry a pregnancy to term (Polge et al. 1966). Fetal death in conventionally bred swine was reported in one study as occurring between days 35 of gestation and term (van der Lende and van Rens 2003). Peaks in swine fetal mortality appeared to coincide with changes in placental growth at approximately day 35, from days 55-75, and again around day 100 of pregnancy (van der Lende and van Rens 2003). Overall fetal mortality in this study was 9.2 percent, with 46.9 percent of gilts (102/192) having some evidence of dead or mummified fetuses at farrowing (birth or parturition). Some fetal loss is expected in swine, and may be a function of uterine capacity (the available room in the sow's uterus) (Vonnahme et al. 2002). No health problems were reported for sows in these studies. Typically the sow is able to resorb or expel non-viable embryos and fetuses without ill-effects.

A study of a commercial swine herd found that uterine capacity becomes limiting at approximately day 36 of pregnancy (Vonnahme et al. 2002). Uterine capacity and fetal survival to term were more dependent on placental size and efficiency than the size of the fetus (Vonnahme et al. 2002). In this study, smaller placentae were associated with larger numbers of

viable piglets born per litter, while individual fetuses with large, less efficient placentae generally did not survive.

It is difficult, then, to draw conclusions regarding fetal loss in clone-bearing swine pregnancies. Betthausen et al. (2000) reported the birth of four live pigs from two sows (two pigs per sow) following transfer of 100 to 300 clone embryos plus 100 IVF embryos per sow. Similarly, Onishi et al. (2000) reported the birth of a single clone pig after transfer of 110 clone embryos per recipient, and Polejaeva et al. (2000) reported the birth of 5 clone pigs in one litter following transfer of 100 clone embryos per recipient. None of the studies reviewed indicated health problems in the surrogate dams.

Most of the studies regarding early embryonic development of swine clones and the establishment and maintenance of pregnancy compare differences in survival of embryos generated by varying culture media or activation protocols (Miyoshi et al. 2006b, Lee GS et al. 2005a, King and DeSousa 2006, Lagutina et al. 2006, Kim S et al. 2006, Im et al. 2006). Most of these studies only discuss methodology or *in vitro* development of embryos, and do not provide data on actual pregnancies or pregnancy outcomes. One study (Lagutina et al. 2006) provided more detail regarding pregnancy outcomes for NT embryos cultured to blastocyst stage in defined media (SOF plus non-essential amino acids) prior to implantation in surrogate sows. Four sows received between 40 and 80 NT embryos each. Two pregnancies went to term and resulted in eight and six pigs per litter, respectively, with a total of 14 pigs born. All newborn pigs and their placentae appeared normal at birth. One pig subsequently died of diarrhea, and two were killed when the sow stepped on them.

ii. Parturition

Studies in non-transgenic swine clones did not report any complications with delivery (Betthausen et al. 2000; Onishi et al. 2000; Polejaeva et al. 2000; King et al. 2002; Walker et al. 2002). During discussions with CVM, clone producers indicated that agalactia (failure to lactate) was noted in sows giving birth to piglet clones (see CVM Memorandum I at http://www.fda.gov/cvm/CloningRA_Memorandum_I.htm).

b. Developmental Node 2: Perinatal Period

i. Peer-Reviewed Publications

Incidence of illnesses from all causes for conventional pigs was greatest during the first 3 days after birth (45.2 ± 2.4 percent of all pigs in this age group), with scours (diarrhea) being the most

prevalent cause of illness (52.7 ± 4.9 percent of all reported illnesses) (USDA/NAHMS 1992⁶⁸). Prewaning mortality rates among pigs averaged 11.0 ± 0.3 percent of herds observed (USDA/NAHMS 2001). The principal cause of preweaning pig deaths was due to being laid on by the sow (52.1 ± 2.0 percent of all deaths). Causes of death that might be attributed to infection, such as scours (9.3 ± 1.4 percent of all deaths) and respiratory illness (3.0 ± 0.5 percent of all deaths) were less prevalent, possibly due to biosecurity measures employed at most swine operations (USDA/NAHMS 2001). Because swine are a litter-bearing species, dystocia is less common, and was not cited as a cause of pig death in the USDA study (USDA/NAHMS 2001).

Based on information from the peer-reviewed literature, success rates from swine cloning studies (as measured by number of viable offspring) are low even when compared to reports of cloning in other species. Most pregnancies fail to reach term, despite efforts to support surrogate sows hormonally or with co-transfer of IVP or parthenogenic⁶⁹ embryos. There are few detailed descriptions on health and vitality of neonatal non-transgenic swine clones available in the literature, although the few studies that report successful births claim that pigs are typically normal and healthy.

Bethhauser et al. (2000) reported the birth of four male pigs in two litters cloned from cultured fetal fibroblasts. In this study, 100-300 SCNT embryos and up to an additional 100 IVP embryos were co-transferred to each sow. The confirmed pig clones were reported as healthy, but no details were provided on what, if any, measurements were taken. Onishi et al. (2000) reported the birth of one confirmed pig clone after two separate attempts. None of the embryo clones cultured to blastocyst stage developed to term in this study. The single surviving clone pig was born after the second experiment, when two- to eight-cell stage embryos were transferred to sows. In both experiments, clone embryos were co-transferred with non-clone “helper” embryos. The single clone pig was reported as apparently healthy and normal, and weighed 1.2 kg at birth, within the normal range for its breed. The placenta for this pig weighed 0.3 kg and was reported to be anatomically normal. A more recent study (Lagutina et al. 2006) indicated greater success culturing swine NT embryos in defined media (SOF plus non-essential amino acids) to the blastocyst stage prior to implanting them in surrogate sows. Fourteen boar clones were born alive, and pigs and their placentae were reported as normal in appearance. One pig subsequently suffered diarrhea and died, and two others were killed by their surrogate dam. The 11 survivors were reported healthy at the time of writing, when they were approximately 10 months old, and three had proved fertile by that time. The paper does not state how many of the boars were actually tested, or at what age. Boars are generally considered sexually mature between six and

⁶⁸ <http://nahms.aphis.usda.gov/>

⁶⁹ Parthenogenesis is a form of reproduction in which an unfertilized egg develops into a new individual, occurring commonly among insects and certain other arthropods. Parthenotes typically do not develop into a viable fetus in mammals.

eight months of age; however, because semen production is related to body size, and boars do not achieve mature body size until after one year of age, they are typically not used for breeding until they are at least a year old (Pond and Maner 1984).

Polejaeva et al. (2000) reported the birth of five pigs in a single litter following double SCNT (re-cloning)⁷⁰ and hormonal support of pregnancy. Average birth weight of the clones was 1.24 kg, which was 25 percent lower than average birth weight for non-clone pigs of this same line (1.64 kg). Clone pigs were delivered by C-section, although no explanation was given for choosing this method of delivery.

Walker et al. (2002) reported greater success with hormone supplemented pregnancies and large numbers of transferred SCNT embryos per gilt. All transferred embryos were SCNT clones, as opposed to previous studies which co-transferred so-called “helper embryos.” In this study, four of five recipients carried pregnancies to term, producing litters of five to nine pigs each, for a total of 28 pig clones. Only one of these was stillborn, and one was reported to be born with anal atresia (absence of an anal opening), necessitating euthanasia. Actual birth weights of surviving pigs were not reported, but the authors mentioned that they were small at birth. The authors commented that variability in birth weight could be attributed to uterine effects, and that none of the pigs displayed signs of LOS. They also stated that no placental abnormalities were noted. Similar reports of low birth-weight pigs have been recorded by other researchers (Boquest et al. 2002), although actual birth weights were not presented.

Park MR et al. (2004 and 2005) reported the death of 22 of 35 live born SCNT clones within the first week of life. Several health problems were noted including cerebromeningitis,⁷¹ diarrhea, leg abnormalities, Leydig cell hypoplasia⁷² and unknown factors. Gestation length was similar for the clones (117.82 ± 1.94 days) and the comparators (115 ± 2.4 days). However, the authors noted low birth weights for the clones (0.80 ± 0.29 kg) relative to the comparator pigs (1.27 ± 0.30 kg). The authors attempted to characterize the causes of death of pig clones and noted evidence of problems with blood flow and cerebromeningitis. Many bacterial diseases already established in the swine industry can result in similar clinical signs. Although the investigators tested for the presence of 12 types of microorganisms, they did not detect any infections. However, these clinical signs are commonly noted with bacterial infections which were not

⁷⁰ Re-cloning, double SCNT, and serial SCNT, are synonyms for the process of generating a clone of a clone.

⁷¹ Inflammation of the membrane covering the cerebrum or anterior portion of the brain.

⁷² The Leydig cell is a type of cell found in the testes and is the primary source of testosterone. Leydig cell hypoplasia results in failure to generate sufficient testosterone for development of secondary sex characteristics, and affects spermatogenesis resulting in infertility.

tested in this study including *Actinobacillus suis*, *Escherichia coli* septicemia, *Haemophilus parasuis*, *Salmonella spp.*, and *Streptococcus suis*. Therefore, it is not possible to rule out the presence of these organisms as possible sources of the clinical observations noted in this study. Also, the authors noted that some of the dead pigs were born weak, which may have predisposed them to bacterial or viral diseases which may have contributed to the clinical signs observed in the study. Finally, the authors identified low birth weight as a possible contributor to the neonatal morbidity. This study identified several clinical observations in neonatal swine clones. However, all of the clinical observations noted have been associated with diseases commonly reported in the swine industry (Straw 1999).

More recently, Estrada et al. (2007) also reported an increase in low birth weight SCNT clones compared to pigs derived from artificial insemination. The litter size for the SCNT clones was smaller (6.2 ± 3.1 piglets) compared to A.I derived litters (11.5 ± 2.8 piglets). However, the number of pigs born alive, stillborns, and mummies did not differ from comparators when the data were adjusted for litter size. Additionally, the authors noted no increase in the number of large offspring as reported in other studies for cattle and sheep.

ii. ViaGen dataset

A complete discussion of the ViaGen dataset is provided in Appendix F. For neonatal swine clones (n=7) only birth weights were available. Swine clones in this dataset were smaller at birth than AI comparators (n=16) (1.12 vs. 1.73 kg). It should be noted that these swine clones were delivered by C-section following induced labor on or the day before expected parturition, while comparator pigs were farrowed following natural onset of labor. Low birth weight of swine clones has also been noted in previous studies (Polejaeva et al. 2000, Walker et al. 2002, Boquest et al. 2002).

iii. Unpublished data

Appendix G includes data from a commercial cloning company on birth weight and average daily gain (ADG) of three swine clones during the first three months of life, and on body temperature, respiration and heart rates of five pig clones for the first two days after birth. The five pig clones were born in two litters. Two pigs born in the same litter with low birth weight (1.0 kg) died within 48 hours, with no cause of death reported. Breed of swine was not provided, nor were any data on non-clone comparators provided, making interpretation of weight and growth data difficult. Available comparator data from the ViaGen dataset and published literature indicate that heart rate and ADG were normal for these pigs.

c. Developmental Node 3: Juvenile Development**i. Peer-reviewed literature**

Among conventional weaned pigs (greater than 21 days old), the total number of illnesses reported was 1,721/213,910 pigs weaned in the observation group (0.8 percent). The most commonly reported cause of illness among weaned pigs was nervous system disorders (12 percent of illnesses), followed by respiratory problems (10.4 percent). The number of weaned pigs dying was reported as 1,906/213,910 pigs weaned (0.9 percent), with most common causes of death also attributed to nervous disorders (13.4 percent) and respiratory problems (16.6 percent) (USDA/NAHMS 1992⁷³).

Archer et al. (2003a) studied physiological and clinical chemistry markers of swine clones. Clones and age- and breed-matched comparators were evaluated at 15 and 27 weeks of age. Body weights of the animal clones at 27 weeks of age did not differ significantly when relative coefficients of variation in body weights were compared. Body weights of all the animals overlapped and were within the normal range for the age and breed, with the exception of a single clone that was small at birth, and never attained the size of its littermates. Teat number was the same for all (6, 6 distribution) except one clone piglet (6, 7 distribution). One of the clones also exhibited an unusual hair growth pattern (*e.g.*, longer and sparser), which the authors state prompted an examination of the histology of the skin. Results of that investigation indicated that with one exception, skin morphology showed no unusual variations among the pigs. The exception was a clone that exhibited morphology indicative of hyperkeratosis. Whether this was the same pig as the one exhibiting the unusual hair pattern is not specified.

Hyperkeratosis, also referred to as parakeratosis, occurs in naturally bred and AI pigs between the ages of 6 and 16 weeks, and is generally associated with zinc and essential fatty acid deficiency or excess dietary calcium or phytate (naturally occurring compounds in grain that bind certain minerals). Gastrointestinal disorders may also affect zinc absorption, and contribute to the development of this condition (Cameron 1999). Other possible causes of hyperkeratosis include heredity, and other, non-specific causes of skin inflammation (Blood and Radostits 1989). Dermatitis vegetans is the inherited form of this disease in swine, and is a semi-lethal recessive gene (Blood and Radostits 1989). The inherited form of the disease generally presents before the pig is three weeks old.

Results of blood clinical chemistries for clones were similar to those of age-matched controls. In addition, changes in alkaline phosphatase, globulin and A/G ratio between 15 and 27 weeks were also similar among clones and controls, and are age appropriate. Cortisol levels were more

⁷³ <http://nahms.aphis.usda.gov/>

variable among clones and controls, and across time periods compared to other measurements, but no consistent trend could be identified. Differences in cortisol concentrations may result from individual variation among animals in their response to handling-induced stress.

For this set of animals, with one exception, no anomalies are present that would appear to have any direct impact on animal health. Nutrition-related parakeratosis has been known to result in reduced growth and appetite, diarrhea, and vomiting when severe. Mortality from this disease is uncommon. Dermatitis vegetans, the hereditary form of this disease in swine, may result in death of the pig, or the pig may recover completely (Blood and Radostits 1989). This is the only known report of hyperkeratosis occurring in clones. The apparently normal status of the clinical measurements indicates that the clones in this study possess the same physiological functions as their sexually-derived counterparts.

In a companion study, Archer et al. (2003b) evaluated behavioral characteristics including food preference (to apples, bananas, saltine crackers, and carrots), temperament (as judged by time to remove a towel placed on the pig's head and attempts to escape mild restraints (being placed on their backs and being lifted off the ground)), and time budgets (the amount of time spent engaged in a particular activity in their pens). The results of this study indicated that the behaviors of pig clones were no more homogenous than the behaviors of their comparators. The relevance of the study to an evaluation of the health of swine clones is that the animals behaved in much the same manner as conventional animals, and displayed no behavioral anomalies at the times tested (15-16 weeks of age for the food trials, 8-9 weeks and 14-15 weeks for the towel test, 7 weeks for the restraint tests, and 13-15 weeks for the time budget tests).

Carroll et al. (2005) examined the acute phase response in piglet clones. This response occurs in the early phases of infection, and serves as a defense mechanism that enables the body to control infection while the adaptive immune response is being developed (Janeway et al. 1999).⁷⁴ Carroll et al. (2005) hypothesized that an impaired acute phase immune response might provide an explanation for the increased mortality observed in clones in some studies during the perinatal and early postnatal periods. To test this hypothesis, they compared the acute phase response to a lipopolysaccharide (LPS)⁷⁵ challenge in clones from two genetically identical cell lines (cell line C1, n=2; cell line C2, n=7) and related AI-derived comparators (n=11). All pigs were raised under the same conditions, were apparently healthy, weaned at 21 days, and challenged when they were 26 – 30 days of age. The authors measured changes in plasma cortisol and two cytokines, tumor necrosis factor α (TNF α), interleukin-6 (IL-6), every 30 minutes over a six hour

⁷⁴ Immediately upon exposure to bacteria, immune cells in the blood release the proinflammatory cytokines tumor necrosis factor α (TNF α) and interleukins-1 and 6 (IL-1 and IL-6). These cytokines stimulate the liver to produce acute phase proteins which act like antibodies by binding to bacteria, leading to their destruction.

⁷⁵ LPS is a component of the cell wall of gram-negative bacteria.

period from two hours prior to LPS challenge through four hours following challenge. Prior to LPS challenge, clones had lower levels of cortisol and TNF α compared to AI controls. Following LPS challenge, clones of cell line C2 demonstrated lower cortisol, TNF α and IL-6 responses compared to AI controls. In clones of cell line C1, there was a tendency for TNF α and IL-6 responses to be lower compared to controls. From these results, the authors concluded that cortisol and proinflammatory cytokine profiles associated with the acute phase response are altered in clone piglets, and that the cell line from which clones are derived may determine the nature of this response.

ii. ViaGen dataset

Clones weighed less at slaughter and took 27 days longer to reach slaughter weight than their contemporary comparators. This may be due to the fact that clones spent the first 50 days of life in highly biosecure conditions before being moved to a conventional swine facility for the start of the experiment. This would have presented swine clones with a significant immune challenge that likely would have slowed growth as they adapted to their new environment.

Three clones were described as “poor-doers:” animals that exhibited slow growth rates and other health problems. All three of these animals suffered from periodic or chronic scouring along with other health problems (see Appendix F). On average, organ weights as a percentage of body weight were lighter for clones than for comparators. Overall, swine clones had lower IGF-I and estradiol-17 β levels at slaughter compared to non-clone comparators. Other blood values were variable among animals, and did not indicate any consistent trends. One clone was diagnosed with a lung adhesion at slaughter.

d. Developmental Node 4: Reproductive Development and Function

Semen collected once a week from a boar clone between 10 months to 14 months of age and evaluated in a study conducted by Martin et al. (2004) was reported as having motility, sperm concentration and ejaculate volume similar to those of non-clone boars. Clone gilts in the same study were reported to show first estrus at 215 ± 4 days and 200 ± 0.6 days for the two genetic lines in the study. Five clone gilts (four transgenic, one non-transgenic) were inseminated with the semen from the clone boar and all became pregnant and farrowed without incident. Gestation length, litter size, proportion of pigs born live and birth weights were similar between litters from the clone pigs and litters from non-clone pigs. Sixty-five pigs were born as a result of the matings, three of which were stillborns (4.5 percent). By way of comparison, the mating of five non-clone females to a non-clone boar resulted in 60 pigs born with four stillborns (6.7 percent). All live-born pigs born to the clone parents were normal except one pig which had contracture of

the flexor tendon (arthrogryposis) of both hind limbs. The authors reported that the frequency of arthrogryposis was similar to reported estimates for commercial swine in Australia. Survival to weaning was similar for both groups with 58 of the 62 live born clone offspring pigs surviving (94 percent) and 53 of the 55 non-clone offspring pigs surviving (96 percent).

No other peer-reviewed reports have been identified to date on puberty and reproduction in male or female swine clones. However, as part of a large dataset submitted to CVM by ViaGen, Inc., four clone boars and three comparator boars (one nuclear donor and two AI-derived sons of a nuclear donor) were examined for semen characteristics and fertility (see Appendix F for the full report). There were no differences between clones and comparators in sperm concentration, total sperm count, percent total motility, percent progressive motility, or number of sperm abnormalities. Farrowing rates were higher for clones than for comparators (73.5 vs. 62.5 percent), although this difference may be due to the age of one of the comparators, a five-year old Hamline⁷⁶ boar used as the nuclear donor for three of the clones in this study. Litter size was more variable for clone sires than for comparators, and mean litter size for litters sired by clones was slightly smaller than for comparator boars (10.94 vs. 11.76 pigs/litter), but were similar to the average cited for U.S. commercial swine production (10.66 pigs/litter).

e. Developmental Node 5: Post-Pubertal Maturation and Aging

No reports on aging and maturity in swine clones were identified.

f. Progeny of Swine Clones

Martin et al. (2004) reported that progeny of male and female clone pigs were born with comparable birth weights to non-clones. One offspring of the mating was reported to have contracture of the flexor tendon in both hind limbs. The frequency of this abnormality was reported by the authors as similar to reported estimates for the Australian swine industry. Survival rates to weaning were similar between the offspring of the clones and the non-clone offspring (94 percent and 96 percent respectively).

In a follow-up to the study reported by Archer et al. (2003a) for swine clones, Mir et al. (2005) reported on the body weight and blood profiles of female swine clones and progeny of swine clones up to 27 weeks of age. To produce progeny for this study, nine clone and five comparator gilts were bred to the same non-clone boar. All gilts gave birth naturally (spontaneously, vaginally). All pigs were housed under the same conditions, and groups were penned together

⁷⁶ “Hamline” refers to a specific crossbred line of swine used by ViaGen, Inc. This line was developed by crossing various breeds, including Duroc, European Landrace, Pietran, and European Large White swine.

according to age. Blood samples were collected at 15 and 27 weeks, and pigs were weighed at 27 weeks of age. Although litter sizes for clones and comparators were small compared to industry standards (7.78 ± 2.6 and 7.40 ± 3.0 pigs/litter for clones and comparators, respectively), there were no differences between clones and contemporary comparator gilts in this study. There were no differences in body weight at 27 weeks between clones, comparators, or progeny of clones and comparators. As with the Archer et al. (2003a) study, the ranges in blood values between clones and contemporary comparators overlapped for the variables measured. Significant differences in blood urea nitrogen (BUN) at 15 weeks and alkaline phosphatase (ALP) at 27 weeks were noted between clones and comparators as well as clone progeny and their comparators. The authors note that other blood variables found to be different between clones and comparators (creatinine, phosphorus, and calcium) were not different between progeny of clones and their comparators.

No other peer-reviewed reports have been published to date on progeny of swine clones. However, a large study of progeny of swine clones was submitted by ViaGen, Inc. The study included data from 402 progeny of swine clones and 300 age-matched, genetically-related comparator pigs. For a full description of this study, see Appendix F. All progeny in this study were farrowed and raised to slaughter under similar conditions. The percentage of animals reaching slaughter age was lower for progeny of clones than for comparators (295/402, 73.4 percent vs. 243/300, 81 percent); however, much of this difference can be attributed to the loss of a single litter of clone progeny. When data from this litter is excluded, the percentage of neonatal deaths was similar for progeny of clones and comparators, and was similar to the averages for commercially raised U.S. swine. Abnormalities noted among pigs in this study (e.g., anal atresia and spraddle legs) have been documented in the commercial U.S. swine population at similar rates. There were no consistent differences between progeny of clones and comparators for blood clinical chemistry or hematology, and the few minor differences noted did not indicate any health concerns. Growth rates were similar between groups in this study, also.

g. Summary for Health of Swine Clones and Their Progeny

Swine carrying clone pregnancies do not appear to experience hydrops and dystocia. With the exception of one pig clone born with anal atresia, no other reports of frank deformities have been noted for this time period in non-transgenic swine clones, although birth weights may be lower in swine clones relative to non-clone comparators. The single study reporting high mortality rates in non-transgenic swine clones reported clinical signs that may be related to various causes, including infectious disease, which cannot be ruled out based on the available

data. Swine clones grew more slowly and weighed less at slaughter than sexually-derived comparators, although this difference may have been the result of immune challenge when clones were transitioned from a biosecure environment to a more conventional rearing facility (ViaGen 2005, Appendix F). Three clones in the ViaGen study were described as “poor doers,” with periodic or chronic scouring and other health problems that resulted in poor growth. One clone was diagnosed with a lung adhesion at slaughter. Comprehensive data on immune function in swine clones is lacking. Results of one study conducted during the juvenile period indicate that the acute phase immune response may be altered in piglet clones. The overall implications of this study for the health of swine clones is unclear. Because the test animals were sacrificed at one month of age, it is unknown whether this effect, seen in only two piglets, persists throughout the life of the animals. Further, since these clones were otherwise healthy, there is no reason to assume that their overall immune function had been compromised. Because this alteration was observed in a very small number of clones created from a single cell line, and has not been confirmed in other studies, further work is needed to determine whether these results apply to swine clones in general.

Reports from Martin et al. (2004) and ViaGen, Inc. (Appendix F) indicate that fertility is normal in boar and gilt clones. No reports on aging of swine clones are currently available.

Available reports from the literature and the ViaGen Inc. dataset suggest that progeny of swine clones are not different from pigs derived through conventional breeding. The few reports of health problems in progeny of swine clones indicate they are not different either in quality or frequency from conventionally bred swine.

3. Sheep

Compared to other food animal species which have been cloned, data on sheep clones and their surrogates are sparse. Table V-9 presents a summary of survival of live-born sheep clones from the available literature.

Table V-9: Survival Rates of Live-Born Clones and Comparators				
Reference	Transgenic Status	Surviving/Total Live-Born Clones(fraction)¹	Surviving /Total Live- Born Comparators (fraction)	Comments
Edwards et al. 2002	None	6/15 (0.40)	NM 6/6 (0.00) IVF 4/4 (0.00)	Total of 43 pregnancies to term. Thirty three stillborn or “died immediately after birth”
Loi et al. 2006	None	0/9 (0.00)	IVF 48/51 (0.94) NM 159/165 (0.96)	
Peura et al. 2003	None	1/8 (0.13)	NP	
Wells et al. 1998b	None	3/10 (0.30)	NP	
USDA/NAHMS 2002 (2/01 – 4/01) ⁴	NA ³	NA	0.98	Historical data from animals (mostly natural mating) in commercial operations

¹ Survivors through the Juvenile Period/Live births
² NP = not provided; data not available
³ NA = not applicable
 Transgenic Status: All = All of the clones cited in the publication are derived from transgenic donor cells, Some = Some of the clones cited in the publication are derived from transgenic donor cells, None = None of the clones cited in the publication were derived from transgenic donor cells.
⁴ <http://nahms.aphis.usda.gov/>

a. Developmental Node 1: Pregnancy and Parturition

i. Pregnancy

Little information is available on embryo or fetal loss in sheep following natural service or ART pregnancies. Powell et al. (2006) fed embryo donor ewes (n=64) 30 g/kg supplemental urea two weeks prior to superovulation and breeding by AI as part of an ET experiment to test the effects of high plasma urea concentrations in donor ewes on subsequent embryo development. The control group received only hay prior to superovulation and breeding. Embryos were collected from donors 36-42 hours after insemination and cultured in synthetic oviductal fluid (SOF) medium with or without steer serum, and transferred to surrogate ewes on day 6 of culture. Pregnancies were terminated at 125 days gestation, and the uteri and fetal tissues were surgically

removed from surrogate ewes. Considerable variability in fetal weight was noted among all treatment groups, such that no effect of treatment could be determined statistically in this study. Pre-existing plasma urea levels in embryo donor ewes prior to initiating experimental diets was determined to be most closely related to fetal weight in this study.

Similar to research in cattle *in vitro* embryo production, recent attention has been focused on the use of serum products in culture media and length of time in culture for *in vitro* sheep embryos (Rooke et al. 2007, Walmsley et al. 2004). Rooke et al. (2007) added serum at different times during *in vitro* culture (first two days and last two days of six day culture), and noted differences in IVF embryo development. The comparison groups in this study included IVF embryos cultured without any serum and fetuses produced by AI. A total of 198 IVF embryos were transferred to an unspecified number of surrogates, and 26 AI derived fetuses acted as a control group. Pregnancies were terminated at 125 days gestation in this study. Including serum during the first two days of embryo culture retarded embryo development, and increased weights of the pregnant uterus, placenta, fetus, and fetal heart and liver. In contrast, adding serum only during the last two days of culture increased the number of blastocysts formed by day 6, while fetal weight, heart and liver weights in this group were not different from control (AI) fetuses or fetuses cultured without serum. The authors report, however, that even in the group cultured without serum, some oversized fetuses compared to controls were noted. Concentrations of IGF-II in serum of 125 day fetuses that had been cultured *in vitro* were lower compared to the AI group. This is consistent with the findings of Young et al. (2001), who noted that IGF2R, a protein which binds IGF-II, in the serum of SCNT lambs was related to the development of LOS.

In the study by Walmsley et al. (2004) IVF sheep embryos (n=235) were transferred after two and six days of culture, respectively, to estrous-synchronized ewes (n=79). A total of 83 lambs were born in this study. Number of lambs surviving was not reported in this study. Fewer lambs were born in the early transfer group (n=38) compared to later transfer (n=45). The shorter time in culture did not prevent LOS, although the incidence of LOS in both groups was too low to draw any conclusions relative to the effect of time in culture (two vs. six days) on LOS incidence.

As noted for cattle, abnormal development of the placenta in clones of both embryonic and somatic cell origin is one cited cause of mid- and late-term spontaneous abortion in sheep (Wells et al. 1998a; Fletcher et al. 2007). Further, Wells et al. (1998a) cite too few and/or abnormal cotyledons in placentae of sheep clones. Increased fetal weight was not associated with increased placental weight in studies of sheep IVP fetuses by Sinclair et al. (1999), although these investigators did not examine placental morphology in their study. They hypothesized that fetal overgrowth during the last trimester of pregnancy in sheep, with associated hypoxia (lack of oxygen) and accumulation of lactic acid, was the cause of hydrops in IVP pregnancies.

Similar to Wells et al. (1998a), Fletcher et al. (2007) noted reduced number of placentomes in placentae of clones (n=11) compared to controls (n=40). They also noted that the placentomes of clones were larger than placentomes of controls at two gestational ages (105-134 days and 135-154 days). There was no difference in placental weight between the two groups, although the average weight of individual placentomes was greater among clones than controls. In the earlier group, two of six fetal clones had already died *in utero*, while the remaining four were showing signs of failure as indicated by blood gas measurements. Similarly, in the later gestation group, one of the five fetal clones was dead, while the remaining four were displaying signs of failure. No differences in fetal weight between the two groups were noted, in contrast with Sinclair et al. (1999). Fletcher et al. noted the presence of shed trophoblast tissue and hemorrhagic cotyledonary villi among the clones, but not in controls, which they speculate led to failure of the clone pregnancies. One of the drawbacks of this study is the lack of description of how control pregnancies were established. The authors do not state whether control ewes were bred by natural mating or some form of ART.

Loi et al. (2006) reported similar blastocyst rate and implantation rate following embryo transfer for SCNT (n=93) and IVF (n=123) embryos in a recent study, but noted what they termed “dramatic losses” of SCNT pregnancies after 30 days gestation. Twelve of the 93 (13 percent) transferred SCNT embryos developed to term, compared to 51 out of 123 (41.6 percent) IVF embryos. Macroscopic and microscopic examination of the placentae from clones revealed a marked reduction in vascularization in the villi of the placentomes, as well as lack of differentiation in the trophoblastic tissue.

ii. Parturition

(a) Dystocia

A similar relationship between dystocia, birth weight, and parity of conventional sheep dams was reported by Dwyer (2003) as for cattle (Nix et al. 1998). Ewes (female sheep), however, tend to carry twin pregnancies more often than cows, and ewes bearing single lambs were more likely to experience dystocia and require assistance during labor than twin-bearing ewes. Overall, the incidence of dystocia requiring assistance in Suffolk and Scottish Blackface ewes in the Dwyer study was 10.2 percent for twin-bearing ewes requiring assistance, and 31.0 percent among single-bearing ewes requiring assistance.

LOS has been described in sheep derived from IVP pregnancies as well as in SCNT-derived pregnancies (reviewed by Young et al. 1998). The incidence of LOS in lambs is difficult to estimate, due to the few studies of cloning in this species, the small numbers of animals in individual studies, the lack of comparator information, and the variability among breeds for birth

weight. In a study by Peura et al. (2003), 8/11 clone lambs were more than 20 percent above the average birth weight for their breed at time of delivery, and 5/8 large lambs were delivered by emergency C-section. Only one of the eight live-born lambs survived (Table V-9). This study did not record whether the surviving lamb was delivered vaginally or by C-section.

A recent and fairly detailed study by Loi et al. (2006) noted three of 12 sheep clones delivered at term were stillborn with degenerate placentae, and five clone surrogate ewes developed hydroallantois. The lambs of these five ewes died within 24 hours of birth by C-section, and had degenerative lesions of the liver and kidney which the authors considered to be the result of hydroallantois (see discussion in section 2, a. ii. a. for discussion of the relationship between hydrops and liver and kidney defects). An additional set of twins were born following assisted vaginal delivery from a non-hydropic ewe, but died within 24 hours due to respiratory distress. The remaining two SCNT bearing ewes were delivered by C-section. Their lambs survived for one month.

In another study comparing cloning procedures with other ARTs, an increase in assisted deliveries was observed for ewes carrying clone and IVP-derived pregnancies compared to AI or natural service pregnancies (Ptak et al. 2002). Delivery was assisted because of a lack of adequate uterine contractions and general lack of preparedness for delivery in the ewes carrying clone and IVP-derived lambs.

(b) Other complications

Ptak et al. (2002) reported that normal maternal behavior was impaired in ewes carrying both IVP and clone-derived pregnancies. Ewes carrying IVP or clone embryos did not show common signs of labor (increased activity, bleating, contractions), and delayed licking neonatal lambs (to bond with lambs, and to stimulate lambs to breathe, stand and nurse). Ptak et al. (2002) also reported a lack of expected prepartum changes such as cervical dilation and swelling of the vulva in ewes carrying clone pregnancies. In such cases, delivery was assisted by administering hormones to induce more typical labor, or by C-section.

b. Developmental Node 2: Perinatal Period

Among lambs, the mortality rate for all causes was 2.2 ± 0.2 percent (USDA/NAHMS 2002), although the report did not indicate the age at which losses were most prevalent. According to the USDA report, the principal cause of lamb death was predation (killed by predators) (44.1 ± 1.1 percent for all operations). However, in one large study comprising 4,511 lambs and their dams of various breeds (Christley et al. 2003), factors that had the greatest effect on neonatal mortality were birth weight and blood immunoglobulin concentrations. In this study, the

mortality rate among neonatal singleton lambs was increased in both high and low-birth weight lambs, with the lowest death rate associated with a birth weight of about 5.5 kg. The authors suggested that the increased mortality rate with increasing birth weight may be attributed to the increased risk of dystocia. The relationship appeared to be breed-dependent in this study, with single lambs of Suffolk sheep at greater risk of dystocia than multiple lambs, while multiple lambs of Dorset sheep were at increased risk of dystocia compared to single lambs of this breed. Also in this study, increased serum immunoglobulin levels in lambs were associated with reduced risk of death in lambs 2 to 14 days old.

Studies involving IVP and cloning in sheep report lambs born with many of the same clinical signs as noted for cattle clones, including LOS (reviewed by Young et al. 1998). Mortality rates were elevated relative to lambs produced by natural service in IVP-, BNT-, and SCNT-derived lambs (Campbell et al. 1996, Edwards et al. 2002, Ptak et al. 2002, Walmsley et al. 2004, Loi et al. 2006).

In the study by Walmsley et al. (2004) on length of time in culture for IVP embryos, eight lambs were born with congenital defects: three lambs from a group of embryos cultured for two days and two lambs from a group of embryos cultured for six days, and three lambs whose parentage (culture group) was undetermined. These last three lambs may have been the stillborns noted later in the paper, although the authors do not specify. Congenital defects noted in LOS lambs included cleft palate and maxillary deviation; tendon contracture; enlarged kidneys with fluid-filled renal pelvices and immature connective tissue; pulmonary adenomas; hyperplastic thyroid; and dilation and fibrosis of the hepatic portal and central veins; and right heart failure (the last two were noted in three stillborn lambs). The number of viable lambs appeared to be associated with the oöcyte donor, while incidence of LOS appeared to be related to both the oöcyte and the semen donor in this study.

Unlike cattle, however, there were no differences noted in mortality of lambs produced by IVP and nuclear transfer (NT) in the Ptak et al. (2002) study. Mortality for IVP and NT produced lambs was significantly higher compared to lambs produced by AI and natural mating in the Ptak et al. study. Ptak et al. (2002) compared different glucocorticoid treatments in perinatal lambs in an attempt to improve survival. Although mortality of untreated NT-derived lambs in this study was higher (~30 percent), mortality of lamb clones treated with glucocorticoid (betamethasone) was around 20 percent (based on Figure 3 of Ptak et al. 2002). Actual numbers of lamb clones in each treatment group was not provided, although a total of 22 SCNT-derived lambs were born alive. Another recent study (Peura et al. 2003) looked at nutritional status of the oöcyte donor to determine if it had an effect on embryo development and lamb survival. Although SCNT embryos from donors on a high plane of nutrition had a higher rate of pregnancy initiation, pregnancy loss and neonatal lamb mortality was high in this study for both treatment groups.

Pregnancy rate to term was 17.6 percent (9/51) for the high nutrition group and 5.4 percent (2/37) for the low nutrition group. Of the 11 pregnancies that went to term, eight lambs were born alive. Four of these lambs died within the first 24 hours. Three more lambs died or were euthanized prior to 30 days. The single surviving lamb, originating from the “high” nutrition group, was reported as “thriving” at 15 months of age.

In contrast to Ptak et al. (2002), Edwards et al. (2002) noted all IVF lambs (n=4) and lambs generated by natural mating (n=6) survived and were healthy for the duration of their study, although number of these control animals were small. Stillbirth and neonatal mortality among clones, however, was high. Of 43 pregnancies that went to term, 33 lambs were either born dead or died immediately after birth. The authors did not specify numbers of stillbirths vs. numbers of neonatal deaths, nor do they state the numbers of singleton, twin or triplets that survived to term, although they state that between one and three embryos were transferred per ewe. Fifteen lambs survived the first 24 hours, but only seven of these lived more than 10 days. Necropsy reports on lambs attributed stillbirth and neonatal deaths to either respiratory distress or bacterial infection. Two more lambs died at 25 and 29 days of age due to bacterial infection.

Similar to Edwards et al. (2002), Loi et al. (2006) noted no differences in survival of lambs produced by IVF compared to NM, while seven of nine clones born alive died within 24 to 96 hours of birth. Five of these seven lambs were born to ewes diagnosed with hydroallantois, and on necropsy revealed enlarged renal pelvis and ureters, with blood clinical chemistries indicating hyperkalemia, hypernatremia, and elevated levels of ammonia and urea, but low protein; the authors related all these clinical signs to hydroallantois. An additional two lambs born to non-hydropic ewes suffered respiratory distress and also died after 24 hours.

c. Developmental Node 3: Juvenile Development

Very little information is available on either conventional sheep or sheep clones for this age group. Wells et al. (1998a) noted that BNT lambs that survived the neonatal period and were raised under varying conditions (indoors or outdoors, winter and spring lambing) were apparently healthy, based on blood urea levels (an indicator of kidney function, protein metabolism, and hydration) and daily live weight gains. Loi et al. (2006) noted that two SCNT lambs that survived the neonatal period suffered respiratory dysfunction and died at one month of age due to secondary bacterial infections. These two lambs had apparently normal placentae at birth. Similar to Loi et al., Edwards et al. (2002) reported the deaths of two lambs at 25 and 29 days due to respiratory infection.

The Edwards et al. (2002) study is the only one we encountered that detailed birth weights, growth and some endocrine function in lamb clones, but the study is very small, with only four clones, four lambs derived by IVF, and six lambs from natural mating, and terminated when the lambs were approximately 28 days old. No differences in gestation length or birth weights among the three groups of lambs, although the range of birth weights (1.9 to 11 kg) was greater among clones compared to the two control groups. Similarly, there were no differences among the groups for abdominal circumference or crown-rump length at birth. Growth rates, as measured by changes in body weight, crown-rump length and abdominal circumference averaged per day of study, also were not different among groups. Also, no differences were detected in plasma glucose or insulin levels among the three groups, and plasma insulin decreased with age in all three groups over the course of the study. Differences were noted for plasma ACTH and cortisol among the groups. Clones had the highest ACTH levels throughout the study, and lambs from NM were lowest, while IVF lambs were intermediate between the other two groups, and the range of values for IVF lambs overlapped the ranges of clones and NM lambs somewhat. Cortisol, which is released from the adrenal gland in response to ACTH, followed a similar pattern to ACTH: elevated in clones, lowest in NM lambs, and intermediate in IVF lambs. This may indicate differences in the levels of stress experienced by the different groups of lambs. Given that two of the clones died from infection during the study, it is likely that their cortisol levels were increased in response to the infection. Individual blood levels of ACTH and cortisol were not provided in this study.

d. Developmental Node 4: Reproductive Development and Function

In the Wells et al. (1998a) study, two BNT ram clones were allowed to mate naturally with an unspecified number of ewes for proof of fertility. Eight ewes became pregnant and produced a total of 15 lambs, which were delivered without assistance. Male lambs were reported to weigh 5.2 ± 0.5 kg and females weighed 4.6 ± 0.5 kg, and were not different from lambs sired by a non-clone control ram of the same breed mix as the BNT-derived rams. No similar data on SCNT-derived sheep were identified.

e. Developmental Node 5: Post-Pubertal Maturation and Aging

With the exception of “Dolly,” the first adult SCNT sheep, there is very little information available on post-pubertal maturation and aging in sheep clones. At one year of age, Dolly’s telomeres were 10-15 percent shorter than other sheep of a similar age (Shiels et al. 1999). However, extensive health screens performed failed to identify any abnormality that would suggest premature aging at that time,⁷⁷ despite the reports in the popular press that at 5 years of

⁷⁷ <http://www.roslin.ac.uk/publicInterest/wasDollyOldAtBirth.php>

age that Dolly developed premature arthritis (Dyer 2002). At 6 years of age, Dolly contracted sheep pulmonary adenomatosis (SPA or Jaagsiekte), an incurable, contagious disease that is thought to be caused by a respiratory retrovirus, which causes tumors in the lungs of infected animals.⁷⁸ The disease is endemic in flocks in the region of Scotland where Dolly was housed, and losses due to SPA in infected flocks in Scotland are generally 2-10 percent (Sharp 1991). Before Dolly's diagnosis, two other sheep clones at the same facility had succumbed to SPA, so it is likely that Dolly was infected through contact with those sheep. Dolly was euthanized when tumors were detected in her lungs at the age of 6 years.⁷⁸ Other than SPA, necropsy revealed no other major health problems (Powell 2003b).

Recent reports in the popular press have recorded the death of a relatively young sheep clone in Australia (Arlington 2003), although the cause of death for this animal has not been reported. Under ideal conditions, sheep may live to 15 years of age.

f. Progeny of Sheep Clones

Wells et al. (1998a) reported that progeny of a male ram clone (BNT) were born healthy and within the expected weight range for their breed mix. No information on progeny of SCNT clone sheep is available.

g. Summary for Health of Sheep Clones

Data on sheep SCNT clones is scarce and, except for anecdotal reports and Dolly, do not extend beyond the early juvenile period. Existing data for Developmental Nodes I and II suggest that surrogate ewes and neonatal lamb clones experience similar problems as cattle clones and their surrogates (hydrops, dystocia, LOS). However, given the very few studies that have been conducted and the few animals involved, it cannot be determined whether the frequency of these abnormalities are elevated compared to other ART in sheep. One study (Ptak et al. 2002) indicated that the incidence of LOS in lamb clones was not different from IVP lambs, although actual numbers of lambs with LOS for each ART method was not reported in this study. Data for Developmental Node IV and progeny are only available for BNT clones, and only from one study. The only information available for Developmental Node V is from the death of Dolly and three other sheep clones of unknown ages.

4. Goats

As with sheep, relatively few studies have been conducted with goat clones, and many of these have used transgenic clones. Unlike sheep, however, several of the goat studies are fairly

⁷⁸ <http://www.roslin.ac.uk/publicInterest/DollyFinalIllness.php>

detailed, and provide a more complete picture of the health of the animals involved at most developmental nodes. Table V-10 provides survival data for live-born goat clones from the six studies which reported this information. Similar data for conventionally bred goats is not currently available.

Table V-10: Survival Rates of Live-Born Clones		
Reference	Transgenic Status	Surviving/Total Live-Born Clones (fraction)¹
Baguisi et al. 1999	All	3/3 (1.00)
Chen et al. 2007	None	5/6 (0.83)
Keefer et al. 2001	Some	1/4 (0.25)
Keefer et al. 2002	None	7/9 (0.78)
Lan et al. 2006	None	3/3 (1.00)
Reggio et al. 2001	All	5/5 (1.00)
¹ Survivors through the Juvenile Period/Live births ² NP = not provided; data not available Transgenic Status: All = All of the clones cited in the publication are derived from transgenic donor cells, Some = Some of the clones cited in the publication are derived from transgenic donor cells, None = None of the clones cited in the publication were derived from transgenic donor cells.		

a. Developmental Node 1: Pregnancy and Parturition

i. Pregnancy

A study of 515 healthy, conventionally bred dairy goats (Engeland et al. 1997) noted that the does that spontaneously aborted or delivered stillborn kids did not show any signs of clinical illness. Age of the doe, number of fetuses/doe (twins or other multiples), social status (position in the herd hierarchy) and previous history of pregnancy loss were the factors most closely associated with spontaneous abortion in dairy goats in this study. Does more than three years of age and those which had previously lost pregnancies were more likely to lose a pregnancy during the study compared with younger does and does with a history of successful births. Does carrying three or more fetuses were more likely to lose a fetus than does carrying only one or two fetuses, possibly due to limitations in uterine capacity. Does with a low status in the herd were more likely to lose their pregnancy than does with moderate or high status. The authors suggested that this last factor may be related to stress.

In general, cloning-related problems similar to those noted for sheep and cattle have been reported in only one study on goat clones (Lan et al. 2006). Because there are relatively few reports of goats bearing clone pregnancies (Keefer et al. 2001, Reggio et al. 2001, Baguisi et al.

1999 (the latter two reporting on transgenic clones), Lan et al. 2006, Chen et al. 2007), and the number of animals involved in individual studies is small, CVM could not determine whether the overall lack of complications reported in this species was the result of differences in methodology, species-specific differences, or simply an artifact of the small numbers of animals involved and small number of published papers.

In the Lan et al. (2006) study, three different cell lines: adult skin fibroblasts (SFC), cumulus cells (CC) and fetal fibroblasts (FFC). In addition, FFC were sourced from two different passages: passage 3-5 and 20-25. All CC nuclear donors were from early passage (1-2), and all SFC donor cells were sourced from passages 10-15. None of the SFC embryos survived to 90 days of gestation. One fetus from the CC group was spontaneously aborted on day 80, and one FFC fetus was aborted on gestation day 127. The authors state that the placenta of the aborted CC fetus lacked cotyledons, but it is unclear whether it was completely lacking, or had a reduced number. The aborted FFC fetus presented with an open abdominal wall, exposed organs and hypertrophic heart. The remaining two CC pregnancies resulted in apparently normal, healthy kids. The one term FFC pregnancy resulted in a healthy kid that was apparently normal at birth, but was noted to have a small abdominal hernia at 60 days of age.

ii. Parturition

Data on effects on surrogate dams are not currently available.

b. Developmental Node 2: Perinatal Period

Although few reports on goat clones appear to have been published, the results of these trials contrast with those of sheep and cattle. None of the studies reported cases of LOS or related perinatal clinical signs in goat clones. One paper (Lan et al. 2006) reported the spontaneous late-term abortion of an abnormal fetus, but all term pregnancies resulted in clones that were apparently healthy at birth.

Keefer et al. (2002) reported deaths of two goat clones during delivery of two twin pregnancies, but causes of these deaths were not reported. Keefer et al. (2001) reported normal birth weights in transgenic male Nigerian Dwarf goat clones compared to historical records for the same breed at the same facility (average 2.35 kg), and noted no placental abnormalities. In this study, three young goat clones died from respiratory infections of bacterial origin, one at one day of age, the other two at later times (one month old and three months old). As mentioned previously, respiratory problems of various causes are the most commonly reported clinical sign in ruminant clones (Table V-3). As respiratory ailments, including pneumonia, are common in the general

goat population (Pugh 2002, Merck Veterinary Manual Online 2005⁷⁹), it is not possible to tell from this study whether the infections in these clones were potentiated by the SCNT process. Also, as noted earlier, young ruminants are dependent on passive immunity transferred through colostrum. The Keefer et al. studies (2001, 2002) provided no details on source or quality of colostrum provided to the goat clones after birth, or on IgG levels in kid serum.

Other papers on cloning in goats employed transgenic cells as donors (Baguisi et al. 1999; Reggio et al. 2001). Although transgenesis may have increased complications in studies of SCNT in cattle and swine (Hill et al. 1999, Carter et al. 2002, Lai et al. 2002), studies in transgenic goat clones noted no perinatal morbidity or mortality (Baguisi et al. 1999, Keefer et al. 2001, Reggio et al. 2001). Birth weights of transgenic goat clones were within the expected range for their breed in these three studies.

c. Developmental Node 3: Juvenile Development

Agricultural statistics for conventional goats of this age range were not available. As presented in more detail in Chapter VI, Keefer and her colleagues (Gauthier et al. 2001, Keefer et al. 2001, Keefer et al. 2002) reported on the life history, with particular emphasis on reproductive function, in a small cohort of goat clones. No adverse outcomes were noted in this group, and development appeared to parallel non-clone comparators. In a study of transgenic prepubertal goats, Reggio et al. (2001) reported that transgenic goat clones weighed on average 20.9 kg when weaned at 90 days of age (normal weight and age for weaning dairy goats), and were apparently healthy at 12 months of age.

In the only study encountered to date that included data on hematology and clinical chemistry of goat clones (Behboodi et al. 2005), a group of seven transgenic clones were compared to age-matched comparators and to published values (See Appendix D). Hematology values were similar between clones and comparators, and all hematology values fell within the published range (Pugh 2002). For clinical chemistry, 18/24 values were not significantly different between clones and their age-matched comparators. Of the 19 clinical chemistry values for which published ranges were available, 18 of the values for clones and comparators fell within the published range. The one value out of the published range was creatine kinase (CK) (244.6 vs. 204.4 IU/L for clones and comparators). However, values between clones and comparators were not statistically different. It is unclear whether or not the comparators in this study were also transgenic, whether they were the same breed as the clones, or how they were generated (AI, natural mating, IVP, or ET). The study also does not specify the age of the goats at time of blood sampling, so it is difficult to interpret the high values for CK in these animals compared to the published range.

⁷⁹ <http://www.merckvetmanual.com/mvm/index.jsp>

The only study reporting an abnormality in a goat clone (Lan et al. 2006) noted a small abdominal hernia that was not detected until 60 days of age. Abdominal hernias are not uncommon in young farm animals, and may result from separation of the umbilicus to close to the abdominal wall, or from other injury, and may not be detectable immediately. Umbilical-related problems, including hernias, have been reported in a number of cattle clones (Cyagra 2003: Appendix E). This is the first report of this abnormality in a goat clone. Although the paper does not discuss the fate of this clone, given the small size of the hernia, surgical correction would be an appropriate response.

d. Development Node IV: Reproductive Development and Function

Gauthier et al. (2001) studied sexual maturation and fertility of Nigerian Dwarf goat clones. Four bucks produced by AI were used as controls in this study. Average age at first semen collection for controls was 141 ± 22 days (approximately 20 weeks of age), with the earliest age at first collection 103 days. In buck clones, the earliest age at first collection was 125 days, but average age at first collection for three SCNT-derived bucks was not different from the mean of the control bucks (142 ± 8 days). First semen collection volume for all bucks was small (<0.1 ml). Subsequent collections were made at different ages for clones and controls, and thus are not appropriate to compare. Sperm motility did not appear to be different between clones and controls. The study did not mention whether there were any differences in sperm quality or morphology between clones and controls. Semen from two of the SCNT-derived bucks was used to inseminate six does. Five of the six does were determined to be pregnant, and all five delivered a total of nine healthy kids. Birth weights of progeny of the clone bucks ranged from 1.25 to 2.30 kg, and were not different from non-clone Nigerian Dwarf kids born at the same facility.

Reggio et al. (2001) reported only that five female transgenic SCNT-derived goats demonstrated estrus, were bred by natural mating, and produced kids. Age at puberty, number of services to conception, and details of the parturition and lactation were not reported in this study.

e. Developmental Node V: Post-Pubertal Maturation and Aging

No reports on aging and maturity in goat clones were identified.

f. Progeny of Goat Clones

Gauthier et al. (2001) reported that progeny of male goat clones were born healthy and within expected weight ranges for their breed. Reggio et al. (2001) similarly remarked that five

transgenic goat doe clones were bred and produced kids, and that the kids were continuing to grow as expected.

In one study, progeny from goat clones were found to have shorter telomere length in testicular biopsies compared to conventionally derived animals and the telomere lengths were intermediate to the values obtained for their clone fathers' and age-matched control testes (Betts et al. 2005). This suggests that there was incomplete telomere elongation in the offspring of clones, although as mentioned above it is uncertain whether telomere length is a predictor of longevity.

g. Summary for Health of Goat Clones

Although few studies have been performed on goat clones, some data is available for four of the five developmental nodes, and some limited information on progeny is also available. Unlike cattle and sheep, goat clones do not appear to develop LOS. Likewise, there have been no adverse reports of pregnancy in surrogate goat does (i.e., hydrops and dystocia). Although three goat clones were reported to develop respiratory problems, it could not be determined from the study (Keefer et al. 2001) whether this was related to cloning or not. Goats appear to grow and mature normally and produce normal progeny. The potential effect of shortened telomeres in one report on progeny of goat clones cannot be estimated at this time. No data on post-pubertal maturation are available for goats at this time.

D. Conclusions

Studies performed to date indicate that health problems observed in pregnancies carrying animal clones are not unique; similar problems are well documented in pregnancies produced by IVP and ET, and the same birth defects are sometimes seen in animals that are naturally bred or produced by AI.

Early embryo loss may be related to *in vitro* culture conditions, which may cause abnormal development and early embryo/fetal death in both SCNT and IVP pregnancies; however, embryo/fetal loss appears to be higher in SCNT compared to IVF. Failure of epigenetic reprogramming may also play a role in these losses for SCNT embryos (see Chapter IV). The impact of such events on the health of the dam is dependent on the stage of pregnancy when loss occurs. Losses due to defects in the embryo or failure to implant do not pose a hazard to the dam in early stages of pregnancy, whereas mid- and late-term spontaneous abortions may pose a health hazard to individual females if they are unable to completely expel the fetus and its associated membranes.

As with pregnancy data, information from the perinatal period indicates that cattle and sheep clones are at the greatest risk of morbidity and mortality, compared with goats and swine. Also as observed in the pregnancy data, the abnormalities noted in animal clones are not unique to animals derived by SCNT; similar outcomes have been observed in other ARTs, albeit at lower rates. Most of the information on neonatal mortality comes from cattle.

The major clinical finding associated with these observed outcomes appears to be a complex of clinical signs collectively known as LOS, which has been described in calves and lambs produced by ET, IVP, BNT, and SCNT. Some of the clinical signs reported (*e.g.*, contracted tendons, cranial malformations) may be directly related to fetal oversize, constraints of the surrogate's uterine capacity, and dystocia during labor. Other signs, such as respiratory, cardiovascular, hepatic and renal (kidney) abnormalities, enlarged umbilicus, reduced immune response, and GI tract problems do not appear to be related to these effects, and may occur even among calves within the normal range of birth weights for their breed, but are considered part of the syndrome due to the frequency of co-occurrence. Placental insufficiency, arising from as yet unknown causes, but may be related to early stage epigenetic reprogramming errors (see Chapter 4) is implicated in many of the observed health problems. Hence, LOS may be a misnomer, but the term has become familiar to scientists working in the ART field. The causes of LOS remain unclear, but may be related to *in vitro* culture conditions and other factors, such as incomplete reprogramming of the somatic cell nucleus (see Chapter IV).

Most prepubertal cattle, swine and goat clones appear to grow and develop normally following the early neonatal period as demonstrated by reports on health status and laboratory measurements presented in the available published data and other reports on health status supplied by private companies. However, some reports indicate that congenital abnormalities and neonatal health problems in cattle and possibly sheep may result in death or require humane euthanasia of the animals during the juvenile and later periods if they are chronic or become acute at a later time.

Based on the biological assumptions and molecular data reviewed in Chapter IV, progeny of clones are expected to be normal. Based on empirical observations, data regarding the health status of the progeny of both cattle and swine clones indicate no increased risk of health problems compared to conventional animals.

Three traits that may be genetically caused were identified (cryptorchidism in three calves derived from the same cell line, parakeratosis in one swine clone, and sensitivity to endophyte toxicity in two cattle clones). These may pose health risks to the animals, and are certainly economically undesirable. Healthy clones appear to behave similarly to sexually-derived comparators or, where the information was available, to their genetic donor.

Insufficient time has elapsed since the first domestic livestock clones were born to make any reliable observations on maturity, aging, or the lifespan of these animals. Reports on telomere lengths in animal clones are highly variable, appear to be tissue dependent, and may not be reliable predictors of lifespan. As most female food animals are not maintained to old age, the risk of increased health problems or decreased longevity, if any exist, would be primarily to male animals kept as breeding stock.

This component of the Risk Assessment has compared SCNT with other ARTs with respect to effects on animal health. It has not been possible to perform a strict quantitative analysis of the risk of SCNT to the health of animals involved in cloning for two fundamental reasons: with the exception of cattle, the number of animals that have been studied and reported upon is small, and the rates of adverse outcomes are variable among studies. Therefore, rather than evaluating relative rates (percentages) of adverse outcomes in small, individual studies of cloning and other ARTs, the outcomes from cloning studies should be considered within the context of the actual number of animals involved within a study. Further, a weight of evidence approach, looking for commonalities across multiple studies rather than relying on one or a few small studies, is the most appropriate and supportable approach to use in this analysis.

The conclusions from this assessment of the risks of cloning to animal health may be summarized as follows:

- Cows and ewes used as surrogates for SCNT-derived pregnancies appear to be at increased risk (e.g., incidence) of late gestational complications such as hydrops, as well as dystocia at parturition, that occur, but at a lower frequency, with other ARTs such as IVP. The risk to surrogate swine and goats bearing clones does not appear to be increased compared to the general population; however, the limited dataset in these species increases the uncertainty associated with this conclusion.
- There is an increased risk (e.g., incidence) of mortality and morbidity in perinatal calf and lamb clones compared with calves and lambs produced using other ARTs. In cattle and sheep, the increased risk appears to be a function of LOS. Survival of these clones appears to be a function of both the severity of the clinical signs and neonatal management. The available information suggests that morbidity and mortality is not increased in perinatal swine and goat clones; however, the limited dataset in these species increases the uncertainty associated with this conclusion.
- During the juvenile period (up to approximately six months of age), bovine clones continue to be at an increased risk of morbidity or mortality compared to animals

produced by natural service or ARTs. These deaths appear to be sequellae of the initial developmental abnormalities noted in earlier developmental nodes that persist beyond the perinatal period (*e.g.*, musculoskeletal defects, prolonged recumbency, enlarged umbilicus, respiratory distress, poor thermoregulation). It is likely that these developmental abnormalities are the result of faulty epigenetic reprogramming.

- Clone calves that survive the perinatal period and are not affected by congenital abnormalities appear to be healthy and demonstrate normal patterns of growth and development. Swine and goat clones do not appear to be at increased risk of morbidity or mortality during the juvenile period compared to animals produced by natural service or ARTs. Insufficient information exists to assess the risk in the juvenile developmental node for sheep clones.
- No increased risk of adverse effects on reproductive capacity has been observed in bovine clones. The available studies, although limited, indicate that reproductive function in swine clones is also normal. Insufficient information exists to assess the risk in this developmental node for sheep or goat clones.
- Insufficient data exist to assess the risk of adverse health effects to mature and aging animal clones. The available information indicates that there are no new risks to the health of maturing animals from cloning, although, as previously mentioned, chronic health problems arising from congenital abnormalities may persist. Drawing empirical conclusions regarding longevity in domestic livestock clones is difficult due to the relatively short time that the technology has existed, and the common practice of culling livestock once productivity declines.
- Progeny of clones are normal, healthy and not at increased risk of health problems compared to other sexually reproduced animals.

Chapter VI: Food Consumption Risks

Chapter VI: Food Consumption Risks

A. Potential Hazards and Food Consumption Risks Associated with Food Products from Animal Clones and their Progeny

1. Assumptions

This Chapter of the Risk Assessment is focused on food safety concerns, and assumes that any clones or their products would be subject to the same local, state, and federal laws and regulations as conventional food animals or their products. These assumptions exclude animals with gross anomalies that would not enter the human food supply (although they might be rendered). It also assumes that any hazards arising from the consumption of products derived from animal clones would result from epigenetic dysregulation of the genome of the developing animal, as described in Chapter IV.

The focus of this analysis is the identification of potential subtle hazards in otherwise healthy-appearing animals. Using the *Critical Biological Systems Approach* (CBSA), we evaluated animal health data on as fine a level of resolution as possible. This includes individual animals or even individual analytes per animal in order to have a sensitive screen for adverse outcomes (and thus food consumption risks). Thus, although some of the data in this chapter reprises information previously addressed in Chapters IV and V, the methods by which the data were evaluated differed. Although Chapter V addressed risks to the health of animals involved in the cloning process, the focus of Chapter VI is to identify whether any adverse outcomes observed in clones provide insight into the identification of food consumption hazards. The emphasis here is not on macroscopic adverse outcomes observed in clones and animals produced using other ARTs, but on the search for potential subtle food consumption hazards, which can be thought of as alterations in the physiology of normal and healthy-appearing clones that may indicate food consumption risks. A second step involves determining whether any of the adverse outcomes noted differ from those identified in conventionally bred animals to determine whether cloning poses any unique food consumption risks.

The search for these subtle hazards requires analysis of physiological parameters in blood and tissues (*e.g.*, clinical chemistry, hematological measurements, hormone levels). The working assumption is that these subtle hazards, which are likely the result of inappropriate or incomplete genetic reprogramming, could lead to alterations in the expression of key proteins that could affect the nutritional content of food.

Chapter VI also describes the Compositional Analysis (See Chapter III), which includes all of the information that we could identify on the composition of meat or milk from clones or their progeny. Much of this information has been published or made available since 2005, and tends to evaluate very similar compositional components; much of it is on animals for which physiological data are also available.

2. Critical Biological Systems Approach to Clones of Cattle, Swine, Sheep, and Goats

Chapter V and VI review the health outcomes reported for clones of cattle, swine, sheep, and goats. Over 2,000 references were identified in our literature searches; closer examination revealed that approximately 400-450 papers were useful to the understanding of the subject, and a smaller fraction of those actually cited papers were cited for information on the health of clones or the composition of their food products. Many of these reports are on the same cohorts of animals, but concentrate on different measurements or life stages. Several are reviews of adverse outcomes that have been observed in individual animals or cohorts of animals, but do not provide new data. As indicated previously within the Risk Assessment and detailed in Appendix D, some of the animals on which reports are provided are somatic cell nuclear transfers of transgenic cells, thereby actually being reports on transgenic animal clones. These have been included in the food consumption risk assessment when they provide corroborative information, and the transgenic status of the animals has been indicated when that information is available.

The following section reviews the available information on animal cloning by species, sorting the information into developmental node-specific groupings. This approach was most applicable to bovine clones, where there is significantly more information compared to other species. For those species where information is very limited, such as sheep, the available information is presented as a single unit.

a. Bovine Clones

The largest number of publicly available publications and data sources address clones of dairy and beef cattle. Many reports on effects noted in the Cell Fusion/Reprogramming, Embryo/Fetal, and Perinatal periods tend to come from the early cloning experiments. Others test hypotheses regarding some component of the SCNT process (*e.g.*, cell cycle, cell source, culture conditions, epigenetic reprogramming (see Chapter IV)), and either do not result in live births, or result in very few live births. Very few systematically evaluate the health of the animals, many simply state that “animals appear normal and healthy” or that “no differences were observed between clones and controls.” CVM has extracted as much information as possible from these studies, and has incorporated its findings into the appropriate Developmental Nodes. Some of these

papers, particularly those addressing methodological developments, do not provide information directly applicable to the risk assessment; these have not been cited.

During the course of preparing this risk assessment, clone producers shared information on various cloning outcomes with CVM. The most comprehensive dataset was generated in response to preliminary presentations of the risk assessment methodology by FDA staff at various scientific meetings. In particular, one clone producer, Cyagra, Inc., attempted to gather information on all of the cattle clones that it had produced, including animals that did not survive or that were culled for various reasons. In some cases, this has proved impracticable due to the dispersal of clones to their ultimate owners. The Cyagra dataset is the most comprehensive survey of the health status of cattle clones that has been assembled, and this information has been incorporated into this Risk Assessment. Many of these data have been compiled with data on other clones produced by Cyagra in different parts of the world, and have been published in summary form in a peer-reviewed publication (Panarace et al. 2007), discussed previously in Chapter V, and at the relevant points in this chapter. Details on the animals, the methods used to collect and interpret the data, and the actual data themselves can be found in Appendix E. Cyagra also collected data on the composition of meat from several clones; these data are also in the Appendix.⁸⁰

The information provided by Cyagra differs from that presented in the peer-reviewed literature for several reasons:

- The data were collected specifically to address issues raised in this risk assessment, and thus are not part of a hypothesis-testing study, or written to provide examples of novel or unusual events;
- They have not been peer-reviewed outside CVM (to the Center's knowledge);
- They include individual animal data; and
- They are far more extensive with respect to the number of clearly non-transgenic animals evaluated (n=78 surviving and tracked animals), and the number of observations on individual animals than any other study or series of studies from a particular laboratory.

i. Cell Fusion, Nuclear Reprogramming, and Embryonic and Fetal Development in Bovine Clones⁸¹ (Developmental Node 1)

⁸⁰ ViaGen, Inc. has also developed an extensive dataset on the health and composition of swine clones and their progeny. This is the most comprehensive dataset on the health of swine clone progeny and the composition of their meat. Similar to the Cyagra dataset, these data and their detailed analyses are found in Appendix G, and are summarized within the text of this Chapter.

⁸¹ This Chapter emphasizes the morphological changes observed in this Developmental Node, unlike Chapter IV, that summarized molecular findings.

SCNT is a relatively inefficient process. “Successful” event estimates can be based on the number of fused cells, implanted blastocysts, or pregnancies confirmed at a specific day of gestation, and range from one in one thousand (usually based on fused cells) to one in four (confirmed pregnancy at gestation day 60). The former estimates include the earliest reports of SCNT, as well as studies testing various methodological variables, and reflect the “technology development” nature of the reports. Cloning efficiency in cattle, defined as the number transferred embryos resulting in live calves, has been estimated as 9-13 percent (Wells 2005, Panarace et al. 2007). In a large, 5-year study of bovine clones, Panarace et al. (2007) noted that cloning efficiency can vary from 0 to 45 percent depending on the cell line used for SCNT. When measured from the detection of an established pregnancy in the surrogate dam, the success rate can range from 1-2 percent (as reviewed in NAS 2002b) to approximately 20-25 percent as related to CVM by commercial cloning ventures.

Lack of success at the cell fusion stage is likely due to several factors, the most significant of which are technological (*e.g.*, damage to the oöcyte or donor cells) or biological (*e.g.*, incorrect reprogramming of the genome of the donor cells (Chapter IV) or possible lack of synchrony between donor cell and oöcyte). An alternative justification proposed by Hochedlinger and Jaenisch (2002) among others, is that the extremely low success frequency is a reflection of the inability of all but “stem cells” of various degrees of pluripotency to be reprogrammed, and the serendipitous outgrowth of such cells selected at random for use as donor cells. Regardless of the explanation, few fused donor/oöcyte pairs survive to divide or to become established as pregnancies in surrogate dams.

The following overview of methods that may affect success rates of SCNT are included to allow the reader to understand that there are many different components that may influence cloning efficiency. It is important to remember, however, that the goal of this chapter of the risk assessment is to identify and characterize potential subtle hazards in clones and to determine whether they pose food consumption risks.

(a) Peer-reviewed Publications

The following section provides summaries of studies that contribute to identifying some of the factors that may contribute to successful nuclear transfer at the earliest developmental node. It is intended to be illustrative, and not comprehensive.

Effect of the Zona Pellucida. The importance of the *zona pellucida* in embryo development is not clear, and there are conflicting outcomes in different studies evaluating its role. Dinnyes et al. (2000) compared developmental rates of cattle oöcytes subjected to SCNT, parthenogenetic activation, or *in vitro* fertilization. For the oöcytes undergoing SCNT (n=106), 74 percent fused,

90 percent of fused embryos cleaved by Day 2, and 29 percent of cleaved embryos developed to blastocysts. Eighty-one percent of parthenotes⁸² (early embryos arising from parthenogenetic activation) (n=47) incubated in 5 percent CO₂ in air cleaved by Day 2 of the experiment, but only 17 percent developed into blastocysts. Parthenotes (n=98) incubated in 5 percent O₂, 5 percent CO₂ and 90 percent N₂ had a 79 percent cleavage rate on Day 2, and a 32 percent survival to blastocyst stage. By comparison, *in vitro* fertilized oocytes (n=98) had a 69 percent cleavage rate by Day 2, and 35 percent developed to blastocysts. Because parthenotes are “clones” that have not undergone nuclear transfer, the *zona pellucida* of the embryo is not disrupted. This disruption has been hypothesized to be a possible cause of early embryo failure in nuclear transfer (NT) embryos. The lack of difference in development to blastocyst between SCNT, parthenotes and IVF embryos cultured under the same conditions suggests that disruption of the *zona pellucida* may not be an important factor in early loss of SCNT embryos. Conversely, Ribas et al. (2006) noted no difference in development to blastocyst in *zona*-free vs. *zona*-intact IVF mouse embryos, although the authors stated that *zona*-free blastocysts were smaller and more irregular than *zona*-intact embryos. None of the embryos in this study were transferred to recipients for gestation, however, so further development could not be assessed. In another study involving IVF-derived sheep embryos, Ritchie et al. (2005) transferred eight *zona*-free embryos to four surrogate ewes. One of these pregnancies progressed to term and resulted in a live lamb.

Cell Culture Conditions. Several laboratories have attempted to optimize culture conditions to improve cloning efficiency (Kubota et al. 2000; Li GP et al. 2004; Park ES et al. 2004b; Du et al. 2005). These manipulations have included addition of various compounds to culture media, co-culture with “feeder cells,” and serum starvation. Results of these studies have been mixed, as described below.

In order to study the influence of culture conditions of donor cells used for SCNT, Kubota et al. (2000) used fibroblasts derived from a skin biopsy obtained from a 17 year old Japanese Black beef bull. Donor cells for nuclear transfer were obtained from cultures that had undergone 5 (n=570), 10 (n = 269), or 15 (n = 264) passages.⁸³ All cultures were serum starved prior to nuclear transfer, except that cells from passage number five were divided into two groups, one of which was serum-starved (n=288), and the other was not (n=282). There were no differences among groups for fusion or cleavage rates, but development to blastocyst stage was lower in cells from Passage 5, relative to cells from the higher passage rates, regardless of whether or not

⁸² A form of reproduction in which an unfertilized egg develops into a new individual, which occurs among crustaceans and certain other arthropods. Parthenotes, unlike somatic cells, do not need to be reprogrammed, as they are already in an undifferentiated state.

⁸³ A passage is a cell culture process in which culture vessels that are full of cells are diluted to lower cell densities. This allows the cells to overcome the growth inhibition that comes with limited space. Each dilution is referred to as a passage, so that a culture that has been passaged five times has started with low cell density, grown up to high cell density, been diluted, and had that process repeated four more times.

the cells were serum starved. A total of 54 blastocysts were transferred to 36 recipient cows. Fifteen cows were diagnosed pregnant, of which nine spontaneously aborted between 39 and 123 days of pregnancy. All three of the pregnancies derived from Passage 5 cell cultures spontaneously aborted. Six calves derived from the two more extensively passaged cultures were delivered at term, two from cultures that had undergone 15 passages, the other four from cells that had undergone 10 passages. Two calves derived from Passage 10 donor cells died shortly after birth. In this study it appears that cells that have been more extensively passaged make better donors than less extensively passaged cells. The biological basis for this is not clear, unless cells that have been passaged more extensively in culture somehow become more amenable to epigenetic reprogramming.

In another study of culture conditions, Li GP et al. (2004) compared development of SCNT embryos co-cultured with bovine cumulus cells or with one of two different types of serum (fetal calf serum (FCS) or bovine serum albumin (BSA)) for seven days. The rates of cleavage, morula and blastocyst formation were similar across treatment groups. Fewer blastocysts in the FCS group exhibited normal chromosomal ploidy compared to the BSA group (24/41 or 58.5 percent vs. 24/35 or 68.6 percent), but both of the serum supplemented groups performed poorly compared to the cumulus cell co-culture group, in which 34/42 (80.9 percent) of blastocysts had normal ploidy.

Park ES et al. (2004b) noted that although not effective in improving embryo development alone, the combination of β -mercaptoethanol (ME) and hemoglobin (Hb) enhanced the rate of development of NT embryos to the morula stage compared to unsupplemented media (19/57 vs. 55/85). Development to blastocyst, however, was similar between untreated controls and either the combined treatments or ME or Hb supplementation alone (16/57 vs. 18/99, 15/95, and 40/104 for control, Hb, Me and Hb + ME, respectively). Similarly, Du et al. (2005) found no beneficial effect of adding phytohemagglutinin-L (PHA) to culture media for survival, cleavage or blastocyst formation of NT embryos. From a total of 324 fused embryos, three live calves were born: two from the PHA group and one from the untreated group.

Heterogeneity of Fusion Components. Hiendleder et al. (2004b) studied how differences between nuclear and oöplasm sources can influence SCNT outcomes by using three breeds of cattle (Brown Swiss, Dwarf Zebu, and two varieties of Simmental) as oöcyte sources and granulosa cells from a Brown Swiss cow as the source for somatic cells. Four groups of SCNT embryos were produced. All pregnancies were terminated at 80 days gestation and uterine contents collected to determine the number of viable fetuses. Details on individual fetuses were not discussed, but the authors noted that SCNT fetuses in general were heavier, had a larger thorax circumference, and a reduced crown rump length: thorax ratio (a standard measure of body size) compared to AI fetuses. The proportion of viable fetuses was significantly affected by

source of oöplasm, and was higher for fetuses produced using Dwarf Zebu oöplasts than the other three sources. The lowest viability was noted for one, but not both, of the Simmental sources. Interestingly, the difference between the two Simmental sources for viability was significantly different. No details regarding the oöcyte donors, other than breed, were provided, so there is no way to determine if other factors (e.g., age of the oöcyte donor cows, nutritional status, health history, or size of follicles collected) might have influenced fetal viability. The authors also compared mitochondrial DNA sequences between the two Simmental oöcyte sources, and noted extensive polymorphism in coding and non-coding regions of the two mitochondrial genomes. Although there has been speculation that mitochondrial dimorphism may affect development of SCNT embryos, only one study was identified that looked specifically at mitochondrial effects on embryo development (Takeda et al. 2005). Also of interest, when fetal morphology was compared in the Hiendleder et al. study, hybrid fetuses (reconstructed using either Zebu or Simmental oöplasm) were not significantly different in size compared to AI fetuses of the same gestational age; however, fetuses produced using the same breed as source of both oöplasm and nucleus (Brown Swiss) exhibited fetal overgrowth. The Brown Swiss cows that were used as sources of oöcytes were different individuals from the Brown Swiss donor of the nuclear DNA. The authors do not report whether they compared mitochondrial DNA of the nuclear donor with that of any of the Brown Swiss oöcyte donors.

Source of Donor or Recipient Cells. Tissue source of nuclear donor cells can also affect development and survival of NT embryos. Galli et al. (1999) used bovine blood lymphocytes as nuclear donors. Lymphocytes, involved in the immune system, must undergo rearrangement of their DNA in order to produce immunoglobulins. Panelli et al. (2004) examined tissues of four aborted NT fetuses and the chondrocytes of the single surviving clone from the Galli et al. experiments. The results were compared to chondrocytes from three non-clone bulls (how the comparator bulls were generated is not described). The aborted fetuses exhibited DNA rearrangement in brain cells that was typical of terminally differentiated lymphocytes, but the surviving clone showed no rearrangement in chondrocytes isolated from his sperm, similar to chondrocytes collected from non-clone bulls. Based on this small dataset, the authors suggested that although terminally differentiated cells can sustain development through the late fetal stage, cells more amenable to reprogramming (dedifferentiation), such as stem cells, were more likely to result in live clones.

Xue et al. (2002) reported on the relative success rates associated with generating clone embryos from three different tissues collected from a 13 year old Holstein cow. In their hands, ovarian cumulus cells had the highest rate of development to blastocyst (57 percent, n=92), compared to skin fibroblast cells (34 percent, n=110) and mammary epithelial cells (23 percent, n=96). Six term pregnancies resulted following transfer of ovarian cumulus nuclear transfer (NT) embryos to recipient cows (5.5 percent, n=109), and four (7 percent, n=57) term pregnancies resulted

from skin fibroblast NT embryos. None of the embryos generated from mammary epithelial cells resulted in a term pregnancy when transferred to recipient cows (n=34). The expression of X-chromosome linked genes in various tissues from deceased animals and conventional controls, and from the placentae of surviving clones was also investigated. Results indicated that X-chromosome inactivation occurred normally in the surviving female clones, but was incomplete in the clones that died. Embryo samples were taken to determine if there were differences in cell counts in embryos from parthenotes and SCNT-derived embryos at the same stage of development. Cell numbers for NT embryos were lower compared to parthenotes at all stages examined (Day 5 morula: 35.1 ± 1.1 , n=48 for NT vs. 43.5 ± 1.5 , n=58 for parthenotes; Day 7 blastocyst: 81.0 ± 3.7 , n=46 for NT vs. 93.8 ± 5.6 , n=48 for parthenotes). The importance of differences in cell numbers is not clear from this study, as mammalian parthenotes generally do not develop to term. Cell counts of IVF embryos, which would have been a more informative comparison, were not provided.

Gong et al. (2004b) compared granulosa cells from adult cattle of two different breeds (Holstein and Chinese red-breed yellow cattle), skin fibroblasts from two individual Holsteins and a Holstein fetus, and oviductal cells from a Holstein fetus for development and survival through the birth of clones. The rate of blastocyst formation was lowest for one of the two adult skin fibroblast sources (253/906 blastocysts/fused couplets or 27.9 percent), although the other adult fibroblast cell line was comparable to the fetal fibroblast cell line (52/132 or 39.4 percent vs. 1294/3412 or 37.9 percent). Fetal oviductal cells had the highest rate of blastocyst formation in this experiment (456/1098 or 41.5 percent). A total of 346 Day 7 blastocysts were transferred to 171 recipients. Pregnancy rate at day 60 was 34.5 percent (59/171), with 25 surrogates carrying 27 calves to term. Because of the small numbers of calves delivered at term, no differences could be detected among donor cell sources for live birth. Of the 27 calves born, eight died during the perinatal period, and another seven died at later stages. Seven of the calves died of causes associated with LOS (hepatic, cardiac, or gastro-intestinal defects, respiratory distress), and eight animals apparently died due to management errors. It is not clear what portion of the perinatal deaths were due to birth defects/respiratory failure or management errors. Birth weights of calves were not reported.

Some authors have suggested that the stage of the cell cycle may also influence cloning outcomes. However, results in different laboratories (Wells et al. 2003b; Urakawa et al. 2004; Ideta et al. 2005) using cells in different stages have been mixed. Wells et al. (2003b) compared putative G₀ cells (cells that apparently were not dividing) to G₁ phase (cells that had begun dividing) cells for SCNT. They noted high early pregnancy losses, but no losses after 120 days of gestation, and no reported hydrops in the G₀ group. In contrast, G₁ phase cells had higher losses to term (21/43 pregnancies lost after 120 days gestation) and higher incidence of hydrops (18/43 (42 percent) of pregnancies), but higher post natal survival than clones from G₀ cells. In contrast

to the Wells et al. study, Urakawa et al. (2004) reported success using fetal fibroblast donor cells in the G₁ phase. However, it should be noted that Urakawa et al. used only G₁ phase cells, and did not compare to other stages of development. Two cell lines were used, derived from fetuses with the same dam but two different bulls. Ten blastocysts were transferred into ten recipients, resulting in nine live calves. According to the authors, calving was “uneventful.” Differences were noted between cell lines, in that three calves resulting from one of the lines tended to be heavier at birth than the six calves of the other cell line used (actual birth weights not provided). One of these three heavy-weight calves died after two days without standing. The authors do not report on the health or survival of the remaining eight calves beyond the first six days of life. Ideta et al. (2005) compared development of embryos constructed with G₁ or M phase fetal fibroblasts, and noted that G₁ SCNT embryos had higher rates of development to blastocyst than M phase cells (31 vs. 16 percent). Only five surrogate cows received embryos in the Ideta et al. study, of which three were diagnosed pregnant on day 30 of gestation, and one live calf was delivered. All of the transferred embryos were developed from G₁-phase somatic cells. The single calf died two days after birth. Health of the surrogate dams, method of delivery, and birth weight of the single calf were not reported in this study. Based on these studies, two of which used only embryos developed from G₁ phase cells, it is not possible at this time to determine the influence of the stage of the donor cell cycle on subsequent development of the embryo/fetus.

It has been proposed that cellular quiescence facilitates reprogramming of the somatic cell nucleus (Wilmot et al. 1997; Wilmot and Campbell 1998) and thus may increase the efficiency of SCNT. To test this postulate, Lawrence et al. (2005) used serum starvation to induce quiescence in bovine granulosa cells prior to their use as donor cells for SCNT. No developmental differences were found *in vitro* between embryos derived from serum-starved and serum-fed cells. More heifers receiving clones from serum-starved cells were confirmed pregnant (9/13, 67 percent vs. 11/25, 44 percent), but embryonic loss between days 29 and 50 of pregnancy was greater in the serum-starved group (88 percent vs. 36 percent). In this study, only one fetus from the serum-fed group survived to term.⁸⁴ Most of the remaining fetuses were lost due to complications from hydroallantois, and two died ≤ 6 d after Caesarian delivery (C-section). The authors concluded that the use of serum-fed granulosa cells was associated with a high incidence of losses during the third trimester due to hydroallantois and fetal overgrowth.

Embryo and Fetal Development. Early pregnancy failures in bovine clones are thought to be a function of incorrect reprogramming of the donor cell that manifest as lethal developmental defects (see Chapter IV). Some of those developmental defects may manifest as difficulties in placentation. For example, Hill et al. (2000b) noted that placentae from gestation day 40-50

⁸⁴ No abnormalities were reported for this calf, and post-natal growth was normal from birth until the calf died suddenly at 9 months of age. The cause of death was acute enterotoxemia associated with Type A *Clostridium perfringens* (Lawrence et al., 2005)..

clone embryos were hypoplastic (low cell density), and had poorly developed cotyledons. (In ruminants, the cotyledon is the fetal part of the junction between the maternal and fetal sides of the placenta where nutrients and wastes are exchanged.) Additional placental anomalies in first trimester aborted fetal clones may include decreased numbers of placentomes (the junction of maternal and fetal components of the ruminant placenta that serve to transport nutrients into and waste out of the fetal environment), and poor formation of blood vessels in the placenta. In contrast, Lee RS et al. (2004) noted that although fewer cotyledons were present in SCNT placentae compared to AI and IVF placentae at day 50 of gestation, vascularization was very good, and appeared more developed in SCNT compared to AI or IVF placentae. Edwards et al. (2003) also studied this phenomenon in transgenic and non-transgenic bovine clones and observed that approximately 50 percent of transferred embryo clones established a pregnancy when measured by the presence of a heart beat between gestational days 29-32. This rate was compared favorably to that observed for non-clone IVF embryos. Edwards et al. (2003) noted that 50-100 percent of embryo clones spontaneously aborted between 30–60 days of pregnancy. Dindot et al. (2004) have noted more than 80 percent of hybrid bovine clone pregnancies (*Bos gaurus* X *Bos taurus*) were lost between gestational days 30 and 60. Evaluation of the early placental structures at gestational day 40 indicated an absence of cotyledons in each clone pregnancy, while the control (AI) fetuses had between 4 and 25 cotyledons per pregnancy). Pace et al. (2002), in a study that included transgenic clones, noted that the fetal abortion rate prior to gestational day 60 was 67 percent. A comparison of the crown-rump length of calved and aborted clone fetuses with AI-generated fetuses from gestational day 25 to gestational day 70 indicated that prior to abortion fetuses grew at the same rate.

Later pregnancy failures are thought to be a function of developmental defects, including placentation abnormalities. Heyman et al. (2002a), for example, compared pregnancy loss between gestation day 90 and calving among clones derived from adult somatic cells, fetal somatic cells, blastomere nuclear transfer (BNT), and *in vitro* fertilization (IVF) animals. They noted that the somatic cell clones showed a pregnancy loss incidence of approximately 44 percent and 33 percent, while BNT clones were lost in only 4 percent of the pregnancies, and the IVF control group lost no pregnancies.

Abnormal placentation can, however, result in the birth of a viable clone (Hill et al. 2000b). In this case, one of six transgenic fetal clones detected at 40 days of gestation continued to develop to term, and when delivered vaginally weighed 37.7 kg, within the normal weight range for Holstein calves (35 to 45 kg). The calf was considered normal based on physical examination at birth. It suckled normally, and at the time of publication, was two years old and considered to be normal. The placenta was similar in weight for term Holsteins (4.3 kg vs. mean expected weight of 5.6 kg). Its structure, however, was highly abnormal, with only 26 cotyledons present, of which only 12 were judged to have been functional. These were enlarged, and the authors

hypothesized that this increased size allowed the normal development of the calf. The authors also note that pregnancies resulting from IVF have also been reported to contain fewer placentomes⁸⁵ than those of conventional cattle. As discussed in Chapter III, the role of transgenesis in the development of this pregnancy cannot be determined. Batchelder (2005), however, working with non-transgenic clones, also noted fewer and larger placentomes in placentae of eight live-born clones compared to AI and ET comparators.

To gain a better understanding about the relationships between fetal and placental growth in bovine clone pregnancies complicated by hydroallantois, Constant et al. (2006) examined placentae from pregnant cows between days 180 and 280 of gestation. Placentae were obtained from 10 AI pregnancies, 6 IVF pregnancies, and 18 SCNT pregnancies. All surrogate dams bearing SCNT-derived pregnancies in this study were diagnosed with hydroallantois and were humanely slaughtered within 1-2 weeks of diagnosis. No differences in morphology or stereology were found between AI and IVF pregnancies, so these groups were combined into a single control group for purposes of statistical evaluation. One of the key qualitative findings of this study is that placental overgrowth in clone pregnancies preceded fetal overgrowth. Before day 220, fetal weights were similar in SCNT and control pregnancies despite evidence of abnormal placental development in SCNT pregnancies (fewer placentomes and higher mean placentome weight). After day 220, fetal weights were significantly higher in SCNT pregnancies and the ratio of fetal weight to total placentome weight was decreased compared to controls. No major histological abnormalities were observed in SCNT placentomes, but after day 220, growth of the fetal component of the placenta, particularly fetal connective tissue, was favored over the maternal component. Based on these findings, the authors suggest that placental overgrowth in SCNT pregnancies is due to factors inherent to the placenta and is not simply driven by fetal overgrowth. The authors also suggest that the term “large placenta syndrome” may be better than “large offspring syndrome” to describe the complications commonly associated with SCNT pregnancies.

The molecular mechanisms underlying placental overgrowth in SCNT pregnancies are unclear at this time. Based on studies in the mouse, it has been suggested that placental overgrowth associated with nuclear transfer may be caused by inappropriate epigenetic reprogramming (DNA methylation), leading to dysregulated expression of genes that regulate placental development. This hypothesis is consistent with the suggestion of Constant et al. (2006) that placental overgrowth associated with SCNT in cattle is mediated by placenta-specific factors.

(b) Summary for the Embryonic/Fetal Developmental Node in Bovine Clones

⁸⁵ The structures involved in connecting the fetal and maternal tissues consisting of a cotyledon and a caruncle in the cotyledonary placenta. The cotyledons or chorionic villi are of fetal origin and "plug into" the caruncles or receptacles in the maternal uterine wall.

(Developmental Node 1)

This period manifests the highest degree of risk for the developing clone. The probability of an SCNT-embryo implanting, and the subsequent likelihood of an implanted clone embryo surviving and continuing to develop appropriately are low. Various investigators have attempted to understand the role of various components of the donor/recipient/cell culture system that comprises the “cloning unit” to improve efficiency with different sources of nuclear or oocyte donors or by manipulating the culture conditions. These studies have been met with mixed results. Lack of success can be attributed to failure of the genome to be reprogrammed (Chapter IV), including failure of the embryo to begin dividing and implant in the uterus, and failure of development in the first trimester (likely due to defects in reprogramming that manifest as poor placentation or other defects that do not allow the fetus to develop), or physical damage to the early embryo. Some may be due to the types of cells used as donors for nuclear transfer. Difficulties that may persist in later pregnancy are largely associated with placentation anomalies that may co-develop with Large Offspring Syndrome (LOS) (see Chapter V). Nonetheless, some of these early embryos do divide, implant, develop, and give rise to live animals, as discussed in the subsequent Developmental Nodes.

ii. Perinatal Development in Bovine Clones (Developmental Node 2)

In the early studies of the technology, relatively high perinatal losses were reported. Deaths generally resulted from phenomena associated with LOS, including poor development of the respiratory and cardiovascular systems. (For a more complete description, refer to Chapter V.) In general, animals with LOS tend to have high birth weights (ranging from 20-50 percent greater than breed averages), poorly developed and sometimes edematous (fluid-filled) lungs and other tissues, and heart malformations and malfunctions. These animals may also have kidney and liver anomalies, and may initially exhibit difficulties in maintaining homeostatic functions such as body temperature and glucose metabolism. The latter are discussed in more detail later in this section. As the expertise develops, however, more animals are either born with no apparent defects, or have perinatal supportive care and survive to grow into healthy cattle.

(a) Peer-reviewed Publications

Most of the adverse outcomes that have been reported result in loss of the fetus before birth, although there is another period of loss after delivery, usually within the first few days of life. Reproducible sets of adverse outcomes have been observed, including LOS and gross morphological abnormalities that may result in pregnancy loss either early in gestation or late in gestation. For example, contracture of tendons has been noted in some clones. None of the abnormalities noted in animal clones are unique to animals derived by SCNT; all have been

observed in natural reproduction, as well as in ARTs such as AI and IVF (reviewed by Cibelli et al. 2002, Pace et al. 2002, Farin et al. 2006, and in Chapter V).

Despite the initial frequency of publications describing adverse outcomes of SCNT, two classes of successful outcomes actually predominate at birth. The first includes animals that may require assistance with delivery and immediate post-natal support in maintaining oxygenation and body temperature. Among others, Cibelli et al. (2002) noted that adverse effects associated with abnormal placental functions in the birth of a group of transgenic clones can be mitigated by intensive veterinary care immediately following birth. One bull clone described by Hill et al. (2000a) required considerable veterinary support immediately after birth due to respiratory problems (immature lungs and pulmonary hypertension), lack of suckling reflex, apparent Type I diabetes, and other health problems. According to this report, the calf improved rapidly, and the diabetes resolved (the calf was able to maintain normal blood glucose and insulin levels) by two months of age. This animal has fully recovered, and is reported to be a vigorous and healthy bull (PIFB 2003).

The second set of successful outcomes consists of those animals born with relatively little assistance (due to the high cost of developing animal clones, most are delivered via planned C-section, and may require more supportive care than animals derived from more conventional breeding techniques), and appear to be normal and healthy (see especially the Cyagra database (Appendix E)). Although many reviews attribute the difference in birth weight to various degrees of LOS, higher birth weights may also be due to the greater care afforded surrogate dams carrying animal clones relative to standard husbandry of conventional animals. Alternatively, birth weight may be related to genetics of the nuclear donor. No data were found on birth weights of nuclear donors, but studies indicate that birth weight is heritable (Knight et al. 2001; Chapter V).

Forsberg et al. (2002) reported the production of 103 cattle clones, of which 47 were produced from non-transgenic cells and 56 from transgenic cells, including a Holstein bull calf generated by recloning an embryo derived from genital ridge cells. Of five pregnancies initiated from that recloning, two aborted prior to gestational day 30, one pregnancy was terminated at gestational day 203 due to hydrops, one set of twins died at birth due to the surrogate dam's ketosis, and the fifth gave rise to "Gene," the first cattle clone not produced from an embryonic cell line.⁸⁶ Little further information on Gene's birth status, growth, or development is found in the peer-reviewed literature, except that as of the end of 2001, when the Forsberg et al. manuscript was accepted for publication, Gene had matured into "*a healthy, fertile bull.*" In a separate recloning trial

⁸⁶ The first publication describing the production of cattle SCNT clones appeared in Science in 1998 (Cibelli et al. 1998). Gene's gestation overlapped with Dolly's and due to species differences in length of pregnancy, Dolly became the first SCNT clone born alive.

described in this report, fibroblast cell lines derived from another fetal clone were used as donors to generate 28 blastocysts that were then transferred into 14 surrogate dams. Nine pregnancies were initiated. Four of those pregnancies went to term, and five calves (three singletons and one set of twins) were produced.

Forsberg et al. (2002) also used cells from adult animals as donors for SCNT. Ear cells from a bull (age and breed not specified) were used to generate 32 embryo clones that were transferred into 17 surrogate dams, of which 10 became pregnant. Five pregnancies were lost prior to gestational day 60, and two more were terminated due to hydramnios or hydroallantois (these conditions are also referred to as hydrops). Three live animals were born, but one was euthanized at 11 days of age due to a heart defect. In a separate trial described in the same paper, cumulus cells from an *in vivo* matured oocyte from a 17 year old cow were used to initiate 11 pregnancies, from which three calves were born. Although information on the health status of many of these animals is not available, 15 of these animals were bred, gave birth, and their milk studied by Walsh et al. (2003) (See Compositional Analysis Method - Section 3).

In addition, Pace et al. (2002) of the same group reported on the development of 117 cattle clones from the reconstructed embryo stage through to lactation. These animals were born between January 1998 and February of 2000. Some of the cell lines from which these animals were developed were transgenic (Forsberg et al. 2002), and 75 percent of the resulting clones were transgenic. Because this report does not distinguish individual animals by cell source, it is not possible to determine which of the animals are transgenic. Interpretation of adverse outcomes should therefore be considered within the context of the discussion of transgenic animals in Appendix D. Of the 117 clone births, 106 were born alive, and 82 remained alive at the time of publication. Birth weights of the surviving clones ranged from 11-72 kg, with an average birth weight of 51 ± 14 kg. The distribution of birth weights was skewed in excess of birth weight ranges for conventional Holsteins.

Pace and his colleagues (2002) divided the calf clone deaths into preventable and non-preventable causes (summarized in Table VI-1). Of the 24 animals that did not survive, 12 died between post partum days 1-5, nine died between days 6-122, and three died at more than 123 days of age. Many of the animals appear to have experienced complications resulting from enlarged umbilici, and three of the deaths were directly related to this condition. For subsequent births, this condition was managed by prophylactically tying or clamping off the umbilical arteries. Difficulties with the umbilicus were also observed at levels apparently higher than in conventional animals by Kishi et al. (2000); Gibbons et al. (2002); Cyagra (Appendix E); Edwards et al. (2003); and Batchelder (2005).

Table VI-1: Summary of Causes of Death of Calf Clones
(adapted from Pace et al. 2002)

Non- Preventable Deaths				
Physiological System Involved	Calves (n)	Age at death (days)	Birth Weight (kg)	Observations
Multiple dysfunctions	3	1-2	11-63	Failure of most systemic functions
Placental	2	1	50-59	Apparent premature separation of placenta
Respiratory	1	3	62	Lung immaturity, meconium aspiration at birth
Digestive	2	78-122	52-60	Chronic diarrhea (n=1); Intussusception of small intestine with obstruction (n=1) ¹
Circulatory	1	42	52	Congenital heart defect
Nervous	1	154	51	Hydrocephalus
Musculoskeletal	1	298	44	Developmental orthopedic disease
Preventable Deaths				
Physiological System Involved	Calves (n)	Age at death (days)	Birth Weight (kg)	Observations
Placental	3	1	53-69	Extensive internal bleeding from enlarged umbilicus
Respiratory	3	1-5	48-66	Developed pneumonia (n=2); Premature induction of labor 16 days early, immature lungs (n=1)
Digestive	5	5-90	59-72	Clostridial infection (n=1); Developed abomasal ulcers ² from eating wood chips (n=2); Bloat (n=2)
Musculoskeletal	1	328	42	Injury, dislocation of patella
Urinary	1	112	59	Pyelonephritis ³ probably secondary to umbilical infection
<p>¹ Intestinal intussusception is the collapse of one portion of the intestine into another, like a telescope, often resulting in the obstruction of the intestine.</p> <p>² The abomasum is the fourth compartment of the stomach of cattle, similar to the human stomach in function.</p> <p>³ Pyelonephritis is an inflammation of the kidney brought on by bacterial infection.</p>				

Chavatte-Palmer et al. (2002) described the gross pathology of 16 abnormal (LOS) SCNT-produced fetuses and stillborn calves. (This report also compares the clinical, hematological, and endocrine characteristics through two months of age in 21 apparently normal cattle clones with the same parameters in calves produced by IVF or AI; detailed discussion of these data can be found the Juvenile Development Node.) The study included 11 abnormal fetuses and 1 normal SCNT fetus between 154 and 245 days of gestation, recovered either at slaughter or after spontaneous abortion, and five abnormal stillborn SCNT calves obtained at term. Three IVF-produced calves, one abnormal calf recovered at 242 days of gestation and two normal calves recovered at term, served as control material. All 16 of the abnormal SCNT fetuses (and placentae) showed some degree of edema due to hydrops. Pregnancy outcomes and gross pathological findings in the clone calves are summarized in Table VI-2.

Table VI-2: Summary of Pathologies observed in Abnormal Fetuses and Stillborn Calves produced by SCNT (adapted from Chavatte-Palmer et al. 2002)		
Pregnancy Outcome	IVF Controls	SCNT
Abnormal Fetuses or Stillborns	NR	<p>11/12 exhibit “pathological gestation;” 1 animal sacrificed for control.</p> <p>5 term stillborn (gd* 274.4 ± 2.6).</p> <p>Abdominal ascites and edema.</p> <p>7 fetal membranes show large edematous cotyledons, and lower mean number of placentomes.</p> <p>Mean and median weight of placentomes higher than for normal pregnancies and controls.</p> <p><i>Kidney defects:</i> Fetus: 1 enlarged Stillborn: both autolyzed. 1 apparently normal fetus had “seemingly small kidneys.” 1 large fatty liver in fetus; seemingly large amount of fat surrounding abdominal organs in “several” fetuses (number not specified). No other gross morphologic abnormalities in other organs.</p>

In a follow-up study by this same group, including animals from the 2002 study (Chavatte-Palmer et al. 2004), the authors noted a 76 percent survival rate (44/58) among clones following the first week after birth. Causes of death during the neonatal period included hyperthermia, umbilical hernia, respiratory problems, ascites (abnormal fluid accumulation) in the chest and abdomen, fatty liver, limb deformities, various digestive tract problems, and abnormal or degenerating kidneys.

Reports from research groups noting no differences between clones and naturally bred animals provide very few details about the health status of the clones. For example, Kubota et al. 2000 reported that although 30 blood measurements were taken on four clone calves, and that they observed no differences between the clones and their age-matched peers, neither the nature nor the numerical values of the measurements were provided.

Matsuzaki and Shiga (2002) evaluated the potential link between endocrine status and perinatal difficulties in Japanese Black clone calves delivered via C-section (selected by the investigators on the basis of a comparison of fetal size and maternal pelvic diameter, or rapidly expanding hydroallantois) relative to clones delivered vaginally, or Japanese Black calves produced via AI, and IVF calves born via spontaneous vaginal delivery. Birth weight, plasma cortisol levels, adrenocorticotrophic hormone (ACTH), and components of the insulin-like growth factor signal transduction pathway (IGF) were evaluated. Average birth weights of clones delivered by C-section were heavier than AI controls; average birth weights of vaginally delivered clones and IVF animals were intermediate compared with C-section clones and AI control animals. Clones delivered by C-section had lower cortisol and IGF-1 levels than AI and *in vitro* produced controls, similar ACTH levels, and had more IGF binding protein-2 (IGFBP-2) relative to controls. The authors concluded that in C-section delivered clones the expected parturition rise in plasma cortisol did not occur, and that these animals failed to initiate the switch to extra-uterine IGF-1 system during late gestation. Four of five C-section delivered clones died within the first week following birth; one of the eight vaginally delivered clones died in that same time period of unspecified causes.

In their first study, Kato et al. (1998) reported that eight of 10 blastocysts derived by SCNT from a Japanese Black beef cow completed gestation and were born. Seven were delivered vaginally, while one was delivered by emergency C-section due to dystocia. Two of the calves were born prematurely. Four of the eight calves died. No abnormalities were noted, and the authors attributed the deaths to “environmental factors” as described in Table VI-3.

In a second publication, Kato et al. (2000) reported the production of 13 surviving clones of 24 deliveries of Japanese Black and Holstein donor cells. Pregnancy duration was approximately equivalent to that of the donor cell breed, except that “a few” recipient cows had shorter gestations. Calves were either born vaginally or delivered via C-section; no criteria were given for the decision to perform C-section. Seven animals were either stillborn or died at delivery.

Table VI-3: Summary of Clone Outcomes
(source Kato et al. 1998)

Calf	Gestation Length (days) ¹	Vaginal (V)/ Cesarean (C) Delivery	Birth Weight (kg) ²	Status at Publication	Cause of Death
1	242	V	18.2	Alive	NA ³
2	242	V	17.3	Alive	NA
3	266	V	32.0	Dead (day 3)	Pneumonia apostematosa from heatstroke
4	267	V	17.3	Dead (day 0)	Inhalation of amniotic fluid
5	267	V	34.8	Dead (day 0)	Inhalation of amniotic fluid
6	276	V	23.0	Alive	NA
7	276	V	27.5	Alive	NA
8	287	C	30.1	Dead (day 0)	Dystocia and delayed delivery

¹ Average gestation length for Japanese Black cattle: 286.6 ± 0.9 days
² Average weight of Japanese Black calf at birth: 27.0 ± 0.8 kg
³ NA = not applicable

Two clones died during C-section due to dystocia, but presented no gross abnormalities. One clone born appeared normal at birth but died 19 days later from septicemia. Six dead clones had significant morphological abnormalities of the kidney or outer extremities, including severe tendon contracture. One clone was born disemboweled, and another had a “warped” face. All of these abnormal births were attributed to infection with Akabane virus, a known teratogen (birth defect inducer), as antibodies to the virus were detected in the serum of afflicted animals. Mean body weights of clones were higher than those of controls,⁸⁷ with nine clones exceeding the mean body weight of controls by >40 percent. Interestingly, Kato et al. report on the unusual appearance of some male clones derived from a bull that was 10 years of age when cells were taken for donors in the SCNT process. At birth, the bull calves were reported to exhibit “*an adult appearance, displayed as many wrinkles in the skin, thick bone structure and rough hairs resembling those of adult males.*” They speculate that these might result from mutations in the donor cells that increase with age or to telomere length.

In the Kubota et al. (2000) study of clones from the 17 year old Japanese Black bull described in the Cell Fusion/Fetal Developmental Node (Developmental Node 1), two calves died shortly after birth, one of which was diagnosed as having Akabane Virus. The other died due to complications following a difficult delivery (dystocia). Four others survived, and were reported to be healthy and normal. The average gestation periods for the clone pregnancies was 294 days (range of 291-299 days), which was nine days longer than the breed average of 285 days. Average birth weight of the clones was 36 kg (range of 30.7 to 42.5 kg), approximately 20 percent heavier than the breed average of 30 kg.

⁸⁷ Mean body weights of Holstein calves at term were 40 kg for females and 47 kg for males; for Japanese Black cattle, mean female calf birth weight at term was provided as 27 kg, and male at 38 kg.

Kishi et al. (2000) used fibroblast cells from ear punches of Holstein or Japanese Black cattle, and somatic cells isolated from the colostrum of mammary gland epithelial (MGE) cells from Holstein cows as SCNT donors. Of the 45 embryos implanted into 31 recipients, three pregnancies were confirmed on gestation day 60, and two calves were born from colostrum derived MGE cells. One clone was delivered at 279 days of pregnancy by C-section and weighed 44 kg; the other was vaginally delivered after induction of parturition at 280 days of gestation and weighed 45 kg. For the fibroblast-derived clones, 43 embryos were implanted into 37 recipients. Five pregnancies were confirmed on gestational day 60, and 2 calves were born (one Holstein and one Japanese Black). The clone derived from the Japanese Black fibroblast died six hours after birth due to internal hemorrhage of the umbilical artery. Two of the Holstein clones (the origin of the cells is unclear) received blood transfusions due to anemia at some unspecified time after birth. The three remaining Holsteins (presumably including the post-transfusion clones) were reported as “*normal and healthy.*”

A series of papers (Taneja et al. (2000); Tian et al. (2000); Xu and Yang (2001); Enright et al. (2002); Govoni et al. (2002); Xue et al. (2002); Savage et al. (2003)) has been published on a group of female Holsteins cloned from a 13 year old cow by the laboratory of X. Yang at the University of Connecticut. Most of these studies report on the birth and later development of these calves, and are discussed in the sections appropriate to those developmental nodes.

In a meeting abstract, Taneja et al. (2000) described the premature delivery of 10 Holstein clones and the supportive care that they required. Normal gestation length for a Holstein averages 282 days (range 280 to 285 days). All the calves born in this study were premature (average gestation length 266.6 ± 2.0 days), regardless of whether labor was induced or occurred naturally. Three cows initiated labor spontaneously at 263.0 ± 3.8 days gestation. Twin calves born to one surrogate dam were stillborn, with one requiring manual delivery. One of the calves in the spontaneous labor group was delivered by C-section, showing signs of stress and hypothermia (body temperature $<100^{\circ}\text{F}$). This calf was hospitalized after 36 hours, when it began running a fever. A chest x-ray revealed immature lung development, and blood gas measurements indicated low blood oxygen concentration. The calf also underwent surgery for an umbilical abscess and for patent urachus (the canal connecting the bladder with the umbilicus) on day 6, after which it recovered and survived. The last calf born in the spontaneous labor group was delivered vaginally with some assistance, was diagnosed with immature lung development and low blood oxygen concentration; it died within 12 hours of birth. Necropsy of this calf indicated bacterial infection and septicemia, as well as immature lung development. The remaining five surrogate dams were treated with dexamethasone 17 hours prior to planned C-sections. Four single calves and a pair of twins were born in the induced labor group. Two calves were delivered vaginally without assistance at 8 and 15 hours post induction treatment. The first calf

(born after eight hours) was healthy and did not require supportive care. The second calf (born after 15 hours) died three hours after birth; necropsy revealed that it had died of hypoxia and immature lungs. A set of twin calves and another single calf were delivered by C-section. One of the twin calves and the singleton survived, while the other twin and another single calf died soon after birth. Necropsy revealed that they had inhaled meconium (the first intestinal discharge that normally occurs after birth that can appear in the amniotic fluid if the fetus is distressed) and the lungs failed to inflate completely. All but one of the surviving calves required supportive care ranging from supplemental oxygen to surgery. The four surviving clones were the subject of additional studies by this lab, including Enright et al. (2002) and Govoni et al. (2002). In the study by Xue et al. (2002) comparing the relative effectiveness of different cell types as donors for SCNT, four of the six calves from the ovarian cumulus group survived the perinatal period; all four of the calves born from donor skin fibroblast cells died. All deaths occurred within 24 hours of birth due to respiratory distress.

Batchelder (2005, 2007a, 2007b) reported on the birth of eight clones (three Hereford and five Holstein) and nine comparators produced by AI (n=3) or ET (n=6). She noted an interaction between outcome and cattle breed, such that Hereford clones were heavier (range 50.0 to 71.0 kg; n=3) than their breed-matched ET comparators (range 31.5 to 48.0 kg; n=3), while Holstein clones had similar birth weights to their breed-matched ET comparators (37.1 vs. 39.4 kg). Neonatal clones had lower values for RBC, WBC and hematocrit at birth and for the first hour, but values were similar to comparators thereafter. Mean rectal temperatures were similar between clones and comparators at birth and declined rapidly within 1 h. This decline was more pronounced in clones (mean 1-h rectal temperatures were 101.4 F° in clones vs. 102.9 F° in comparators). After 6 h, thermoregulation was similar between clones and comparators. Extensive clinical chemistry analyses indicated similar values between clones and comparators for most of the parameters measured, with the exception of measures of carbohydrate metabolism. Clones exhibited lower blood glucose and lactate levels than comparators during the first 24 hours, but were similar to comparators by 48 hours. Plasma concentrations of fructose in clones were higher than comparators, resulting in a nearly 2-fold higher ratio of fructose to glucose in clones both at birth and 6 h later. The authors postulate that hyperfructosemia in newborn cloned calves reflects higher fructose concentrations *in utero* and may be related to the abnormal placental morphology (fewer total placentomes with a flattened shape and increased mass and surface area) observed in this and other studies of SCNT calves. No differences were noted between clones and comparators for plasma protein parameters, electrolytes and minerals, acid-base parameters, blood gases, or parameters indicative of kidney, liver and muscle function. In both clones and comparator calves, plasma immunoglobulins were undetectable at birth and increased rapidly after calves consumed colostrum, indicating that passive transfer of immunity occurred normally in the clones.

Although Batchelder noted several clinical signs often associated with LOS in both Holstein and Hereford clones (delayed time to suckle and stand, hypoglycemia, forelimb flexor tendon contracture, enlarged umbilicus, patent urachus, and respiratory distress), many of the same signs were noted in the AI-derived comparator group in this study (see Chapter V for more details). During the first 48 h, spontaneous urination was rarely observed in clones; manual stimulation was needed to evacuate urine. In this study all clones survived the first 48 hours after birth, but two clones were lost between 72 hours and six days of age. All comparator calves survived.

Wells et al. (2004) reported that a total of 133 clone calves were delivered as a result of 988 embryo transfers of somatic cell nuclear transfers (SCNT) using adult and fetal donor cells. Embryonic cloning resulted in 27 delivered clone cattle from 210 embryos derived from embryonic blastomeres (ENT). Both techniques were reported to result in a live birth success rate of 13 percent. Approximately two thirds of these calves survived to weaning (3 months of age).

Yonai et al. (2005) reported on the growth, reproduction, and lactation of clones whose nuclear donors were a high milk performance 13 year old Holstein and a six year old Jersey that had previously been used for embryo transfer. These animals had previously been characterized as having shortened telomeres, but are otherwise indistinguishable from cattle of presumably normal telomere length (Miyashita et al. 2002). (Discussions of growth and reproductive and lactational performance of these clones are found in Developmental Nodes 3, 4, and Compositional Analysis, respectively). Table VI-4 summarizes the success rates for the two breeds of dairy clones. All embryos, regardless of the breed of the donor cows, were implanted into multiparous Holstein surrogate dams. One of the recipients of Holstein embryos had twin calves. The overall success rates, as measured by surviving calves as a function of embryos implanted were approximately 5 and 10 percent for the Holsteins and Jerseys, respectively.

The authors state that although there is an approximately two-fold difference in the production rates between breeds, this difference is not statistically significant due to the low numbers in the study. The abortion rate in the surrogate dams carrying Holstein clones was approximately two times higher than the Jersey group (68.4 percent v 31.8 percent). No dystocia was noted in surrogates carrying Jersey clones; incidence of dystocia in the surrogates carrying Holsteins was not reported. The authors attribute the differences in outcomes to the smaller size of the Jersey fetuses relative to the Holstein fetuses. Gestational periods and birth weights were reported as being within normal ranges for dairy cows of these breeds. Although there was more variability in birth weights of the Holstein clones than the Jerseys, no symptoms of LOS were noted in these two clone cohorts. The authors note that although cell culture conditions have been implicated as a potential source of large calves, the two cell lines used for nuclear transfer were cultured under

identical conditions, implying that differences between the cell lines (i.e., heredity) was likely responsible.

	Jersey Embryos	Holstein Embryos
Recipients	22	63
Embryos Transferred	37	124
Pregnancy Detected at 40 - 60 days	7 (31.8%)	18 (28.6%)
Failure to Reach Term	1 (14.3%)	11 (61.1%)
Calves Delivered	6/22 (27.3%)	8/63 (11.1%)
Surviving Calves from Transferred Embryos	4/37 (10.8%)	6/124 (4.8%)
Production Rate from Recipients	4/22 (18.2%)	6/63 (9.5%)
Average Birth Weights \pm SD kg (ranges)	29.4 \pm 1.5 (27.5-31.0)	36.2 \pm 7.7 (27.0-47.0)

In summary, the survival rate of clones appears to be in the range of 5-18 percent, depending on how it is calculated. Many of the perinatal clones die of complications or sequellae of LOS. Newborn cattle clones may be more physiologically fragile than their comparators, and differences between clones and comparators include body weight, body temperature, alterations in the amounts of circulating IGF-II, leptin, growth hormone, T4, and differences in mean erythrocyte volume either on the day of birth or shortly thereafter. None of the differences between clones and AI- or IVF-derived controls persisted through the longest observation period (up to three months) (Chavatte-Palmer et al. 2002; 2004), and most resolved within a week or two of birth (Hill et al. 1999 (for transgenic clones); Enright et al. (2002); Govoni et al. (2002); and Tian et al. (2001)) (See subsequent discussions in the sections on the appropriate developmental nodes).

(b) Cyagra Dataset: Perinatal Cohort⁸⁸

Of the 134 clones in the Cyagra dataset that were born or delivered, 103 animals (or 77 percent) were alive three days after birth. The remaining 31 were stillborn, died, or were euthanized within three days of birth. Details on health and survival of conventional, age-matched comparators (comparators) are not available. At the time that data were collected on these animals (late March 2003), 67 were alive (64 percent of those surviving to 48 hours, or 50 percent of those born or delivered). Eight animals died between 4 and 149 days of birth. The problems noted at the time of birth and the causes of death for those clones not surviving are

⁸⁸ Data from Cyagra and the Center's detailed analyses of the data are found in Appendix E: Cyagra Dataset. Summaries of the analyses are presented in the narrative of the Risk Assessment. Readers wishing to have the best understanding of the Cyagra Dataset are urged to read the entire Appendix prior to continuing with the summaries.

summarized in Table E-2 of Appendix E: The Cyagra Dataset. Some animals required supportive care immediately after birth (*e.g.*, glucose, warming, or supplemental oxygen), and many (n=26) received umbilical surgery after birth.

Blood was drawn for clinical chemistry and hematology for 10 clones within a few hours (or in some cases, minutes) of birth. The actual measurements provided by the Cornell Animal Health Diagnostic Laboratory are found in Appendix E, Tables E-100a (clinical chemistry), and E100b (hematology). Charts E-100, E-101, E-102, E-110, E-111, and E-112 compare these values with the comparator population reared on the same farms and the Cornell Reference Values and are also found in Appendix E, along with all of the data from which they were generated.

Ninety percent of the total clinical chemistry values of the clones were within the range of values exhibited by the comparators, and 90 percent of the hematology values were within the comparator range. Twenty-seven of the 33 analytes (substances that were measured, such as sodium, cholesterol, or liver enzyme activity) had either no differences or one difference relative to the comparators (Chart E-101). The remaining six analytes tended to be more variable between clones and comparators. Liver values (AST, GGT, cholesterol, bile acids (hBA)) were lower in several clones, for reasons likely related to the placental/umbilical abnormalities, or transitions from fetal to adult circulation. GGT levels were also low relative to the comparators, probably related to blood sampling prior to colostrum intake, whereas comparators were administered colostrum prior to blood draw. None of the out-of-range values of these analytes poses any particular concern for food safety, as they are relatively close to the comparator range.

Blood cell parameters in the neonatal clones were also very similar to those of the comparators. Fifteen of the 17 analytes had either no differences or just one difference between the two groups (Chart E-111). With the exception of one clone that was infected with rotavirus and subsequently died, all red blood cell parameters were within the range of the comparator group. Three clones had white blood cell counts that were lower than the comparator range. One clone was infected with rotavirus but survived, indicating that at least in that animal, the immune system was functioning appropriately. There did not appear to be an increased incidence of infection in these animals, except where infection was associated with umbilical difficulties, also indicating that the immune systems were functioning appropriately.

“Sentinel” markers were sought that might predict a successful outcome for perinatal clone calves. Based on the literature and the Cyagra data, it does not appear that any one analyte or analyte profile is predictive of whether a particular animal, or indeed, the entire cohort of animals will develop into normal, fully functioning, healthy animals. The laboratory data are consistent with the hypothesis that animals that look and behave normally are normal with respect to laboratory values, implying that consideration of the *complete* dataset on an individual animal is the best predictor of the health of that animal. Further, the seven surviving Cyagra

clones that were sampled twice (# 71, 72, 73, 78, 79, 119, and 132) provide the baseline data for a small subcohort of animals for which there are laboratory measurements at two different time points, as described more fully in the Juvenile section.

(c) Unpublished data

In response to requests by CVM, various groups involved in cloning submitted unpublished data. One such group, a commercial cloning company, submitted body temperature, pulse and respiration rates on 19 cattle clones (breed(s) and gender not identified) during the first 72 hours of life. These data has been discussed in greater detail in Chapter V. Body temperatures were elevated during the observation period (mean 103°F at birth; 102.7°F at 72 hours); heart rates appeared to increase (95.2 beats/min at birth; 138.6 beats/min at 72 hours); while respiration rates remained fairly constant (53.9 breaths/min at birth; 53.1 breaths/min at 72 hours). It is often difficult to evaluate data on heart rate and respiration in livestock, since the stress of handling tends to increase these rates. Body temperature in neonatal clones appears to be quite variable, with some studies reporting hyperthermia (Chavatte-Palmer et al 2002; Batchelder 2005) which may persist through the first 50 to 60 days of life and then appears to normalize.

Another cloning firm presented birth records on two Holstein heifer clones delivered by C-section. The calves weighed 45 and 47.7 kg at time of delivery, within the normal range for Holstein cattle; body temperatures were 100 and 102.6°F at birth, slightly below and above normal (101.5°F) for cattle. These two calves were otherwise normal, according to the veterinarian's notes and limited blood chemistry (See Chapter V for details).

(d) Summary for Perinatal Developmental Node in Bovine Clones (Developmental Node 2)

The combined information from the peer-reviewed literature and the Cyagra dataset indicates that, overall, the health of newborn clones tends to be more unstable than their comparators, with a higher incidence of perinatal death. Abnormalities noted among both dead and surviving clones include respiratory distress, organ malformations, flexor tendon contracture, and umbilical difficulties. Some animals succumbed to infection, but there does not appear to be a decrease in immune function in the population of clones at the perinatal stage. Despite the increased incidence of perinatal death and abnormalities in newborn clone calves, none of the adverse outcomes observed are qualitatively different from adverse outcomes that have been observed in natural breeding or other ARTs. It is therefore unlikely that any unique food consumption risks have been introduced into these animals.

iii. Juvenile Development in Bovine Clones (Developmental Node 3)

Most of the information on this developmental node has been extracted from publications that primarily address the perinatal period.

(a) Peer-reviewed Publications

For purposes of following the cohorts of animals, these reviews have been grouped by institution.

The Institut National de la Recherche Agronomique (INRA) Studies: Renard et al. 1999 and Chavatte-Palmer et al. 2002, 2004

Renard et al. (1999) reported one case of lymphoid hypoplasia in a clone generated from cells in an ear biopsy of an animal that had herself been the product of blastomere (or embryo) nuclear transfer (BNT). An echocardiogram performed on the animal immediately after birth revealed an enlarged right ventricle of the heart. The animal was treated with an angiotensin converting enzyme (ACE) inhibitor and given diuretics for one month, at which time the condition was reported to be resolved. Blood samples taken every two days after birth revealed relatively high reticulocyte counts and immature blood cells in the blood during the first three weeks of life. Lymphocyte (white blood cell) counts were also reported as normal for about a month after birth, but counts fell rapidly after that time. Hemoglobin levels in the animal also decreased at about day 40. On day 51, the animal died from severe anemia. Histological examination of the calf revealed hypoplasia (lack of development) of the thymus, spleen, and lymph nodes or global lymphoid aplasia (absence of lymphoid cells in all organs in which they would likely be found) that likely began at birth. No evidence for the endogenous synthesis of immunoglobulin G was detected. Bovine Viral Diarrhea virus, which has been known to induce thymic atrophy, was ruled out. SCNT was implicated as the cause of the lymphoid aplasia, possibly due to the selection of a cell with a mutation responsible for the expression of the portion of the genome governing lymphoid development, or lack of appropriate reprogramming of the somatic cell nucleus. In a follow-up study by this group (Chavatte-Palmer et al. 2004) an additional four clones were diagnosed with thymic aplasia. Histological examination of the thymus glands of these calves indicated abnormal tissue organization, suggesting the aplasia was the result of epigenetic errors. It is not clear from the late report whether these four clones were also the result of serial cloning. To our knowledge, this is the only laboratory reporting thymic aplasia in clones. Three other calves in this cohort died suddenly with few or no clinical signs: two died of diarrhea, and one died without any apparent cause.

To investigate if apparently normal calf clones share similar clinical and endocrine characteristics with conventionally-bred calves, Chavatte-Palmer et al. (2002) monitored 21 clones from birth to two months of age. Data from this part of the study are summarized in Table VI-5. (Some of the results of this study are discussed in the Perinatal Developmental Node). Blood samples were collected daily for the first week of life, weekly for the first month, and every two weeks thereafter.

For the first week after birth, the mean rectal body temperature was higher in clones than AI controls, and some temperature spikes (up to 41° C; normal temperature is considered to be approximately 38.5 °C in dairy cows) were observed. Elevated temperatures in the clones persisted for 24-36 hours, and were not sensitive to pharmacological intervention. Animals were cooled by wrapping in wet towels and providing ventilation, although they did not appear to be distressed during the temperature spikes. No bacterial infection was detected, and no changes in hematology or clinical chemistry were observed. The authors state that the mean temperature remained elevated for 50 days, although data are only provided for the first week.

Hematologic parameters evaluated in this study included red blood cell count (RBC), hematocrit (HC), hemoglobin (Hb), and counts of white blood cells (WBC), including differentials (counts of the distributions of populations within the overall category of white cells). Mean cell volume (MCV) was higher in clones than AI controls, and the neutrophil:lymphocyte ratio at birth was higher in clones than in AI controls. The authors state that this ratio reflects the normal increase in fetal cortisol production at birth that occurs as a result of adrenocortical maturation. As previously mentioned, one clone presented with lymphoid aplasia (Renard et al. 1999) and had decreased lymphocyte and RBC counts. Measurements of all other hematologic parameters in clones were similar to those in AI controls. Clinical chemistry values were within normal limits. Thus, with the exception of the aplastic clone (Renard et al. 1999), no clinically relevant findings accompanied these measurements over the time period of the study. (For a discussion of the nature and relevance of these tests, refer to Appendix F).

Thyroxine (T4) is an important, stimulatory regulator of metabolic rate. Because metabolic rate is (usually) correlated with body temperature, elevated T4 secretion may be implicated as a cause of hyperthermia in clone calves. However, plasma T4 levels were lower in clones than controls during the first two weeks of life, after which they were similar to levels in controls. Chavatte-Palmer et al. (2002) noted that lower plasma T4 levels coupled with elevated body temperatures in young calves are consistent with studies on brown adipose tissue (BAT) by Carstens et al. (1997b). Brown adipose, found in neonates of many mammalian species, serves to generate heat

Table VI-5: Summary of Clinical, Hematologic, and Hormonal Characteristics from Birth to Two Months of Age in Apparently Normal SCNT Clone Calves Versus AI and IVF Controls (adapted from Chavatte-Palmer et al. 2002)			
Outcome	AI or IVF Controls	SCNT	Comment
Live Births Total Caesarian delivery Vaginal delivery	n=176 not specified not specified	n=21; 7 fetal origin; 13 adult origin 20 (18 at term, 2 were 1 week before term). 1	Clones delivered via C-section when natural calving had not occurred by gd 282. All calves survived to at least 2 mo of age.
Body Weight at Birth (kg)	43.7± 2.7 n=176	55.1± 2.7; n= 26 Higher compared with and AI and IVF calves (P<0.01)	No significant difference between AI and IVF.
Body Temperature (BT) at birth	Lower than SCNT (approximately 38 to 39.5°C)	Mean rectal BT higher in SCNT than controls in 1 st week, and until 50 days. Data provided for only 1 st week. Peak temperature spike approximately 41° C. No accompanying clinical signs.	Comparison between n=10 NT and n=10 combined AI (8) and IVF (2). Not sensitive to NSAID; regulated by using wet towels and ventilation.
Hematologic Parameters RBC, HC, Hb, WBC, Differentials Mean cell Parameters	n=8 Mean cell volume (43.59± 0.60 fl). Neutrophil: lymphocyte ratio at birth 3.14 ± 1.1; higher than SCNT.	n=21 live clones. Mean cell volume (50.07 ± 1.29 fl) higher than AI. Neutrophil: lymphocyte ratio at birth 6.28 ± 0.9; higher than AI. 1 animal with lymphoid aplasia (Renard et al. 1999), sudden decrease in lymphocyte and RBC counts.	Measured only at birth.
Clinical Chemistry Urea Creatinine AST ALT	NR	All values within normal limits; individual data not provided.	Measured only at birth.
Thyroxine (T4)	n=4	n=7; lower than AI controls for days 1-15 (P<0.05). Approximate kinetics the same as AI (rapid decrease from birth to d 4, then constant low level (~15-25 pmol/l) to day 15.	Measured for 2 months to determine whether associated with hyperthermia.

Table VI-5: Summary of Clinical, Hematologic, and Hormonal Characteristics from Birth to Two Months of Age in Apparently Normal SCNT Clone Calves Versus AI and IVF Controls <i>(adapted from Chavatte-Palmer et al. 2002)</i>			
IGF-1	n=5; No diff. from SCNT.	n=7; no difference from AI.	Measured from day of birth until 80 days of age.
IGF-II	Lower than SCNT at birth and d 15.	Higher than AI at birth and day 15 (P<0.05).	
IGFBP	No difference from SCNT.	No difference from AI.	
Leptin	n=5; Lower than SCNT animals, and less inter-animal variability.	n=6; higher than controls during first week after birth (P<0.01). More inter-animal variability and changes in absolute response in SCNT animals. Levels revert to normal in amount and amplitude after one week.	Measured from day of birth until 28 days of age.
Growth Hormone	n=6; no difference from SCNT	n=5; no difference	Same as leptin assay.
Pre- and Post-Prandial Insulin and Glucose	n=6; No significant differences between AI and clones at 1 or 8 days of age.	Some clones presented with hypoglycemia and hypothermia during first 24 h post partum. No significant differences between clones and AI after the first 24 hours.	Measured at 1 and 8 days of age.
Cortisol (ACTH Induction)	n=2; C-sect, n=6; natural birth. Basal levels in C-section births lower than natural birth.	n=11; C-section. n=1; natural birth. No significant differences between clones and controls. Basal and stimulated levels in C-section births lower than natural birth.	Increase in plasma cortisol in response to ACTH stimulation reflects appropriate adrenal maturation and function. Lower basal cortisol values probably due to C-section and not NT or IVF.
AI = artificial insemination IVF = in vitro fertilization NR = not reported C-section = Caesarian delivery gd = gestational day NSAID = non-steroidal anti-inflammatory drug			

to keep the newborn warm during cold stress. Metabolism in BAT is stimulated by norepinephrine, and norepinephrine secretion is increased by stress (e.g. cold stress). In cattle, BAT usually disappears following the neonatal period (Blaxter 1989). The findings of Carstens

et al. (1997b) indicated that, in contrast to most tissues, thermogenesis in BAT is suppressed by elevated T4 (see discussion in Chapter V). It is therefore plausible that the hyperthermia observed in the calf clones by Chavatte-Palmer et al. (2002) and other investigators is independent of T4, and may instead be explained by increased BAT metabolism. However, in the absence of empirical data on BAT metabolism (e.g. norepinephrine levels as an indirect measure), this idea remains speculative.

Chavatte-Palmer et al. (2002) investigated levels of several hormones that regulate growth; insulin like growth factor (IGF)-1, IGF-2, IGF binding protein (IGFBP), leptin, and growth hormone. There were no differences between clones and AI controls in concentrations of IGF-I, IGFBP, or growth hormone. Levels of IGF-II in clones were higher than controls at birth, but were lower in clones on day 15. Leptin levels were higher in clones than controls during the first week of life, but were similar in clones and controls for the remainder of the study. As leptin is produced by adipocytes (fat cells), the authors speculate that increased leptin levels in clones may reflect greater amounts of adipose tissue, consistent with the higher body weight of clone calves, and supported by the authors' subjective postmortem observations of more intra-abdominal fat in clones compared to non-clone calves.

As a measure of possible metabolic disturbances possibly related to the increased size of clones, Chavatte-Palmer et al. (2002) assayed plasma concentrations of insulin and glucose, both before and after feeding, and one and eight days of age. There were no differences between clones and AI calves in either pre- or post-prandial concentrations of insulin and glucose.

Finally, Chavatte-Palmer et al. (2002) measured cortisol secretion, both basal and in response to challenge with adrenocorticotrophic hormone (ACTH, the pituitary hormone that induces the production of cortisol). In cattle, as well as many other mammalian species, secretion of cortisol may be increased by stress. In prematurely born animals, the cortisol responses to ACTH challenge are decreased. Thus, challenge with ACTH provides a measure of adrenal maturation and function. Relative to calves born vaginally, cortisol levels (basal and ACTH-stimulated) in calves born by C-section were lower in both clone and non-clone calves. At one, seven, and 30 days of age, all of the calves exhibited similar cortisol levels following challenge with ACTH, indicating that maturation of the adrenal axis was normal in clone calves. These results also provide evidence that overgrowth of clone calves *in utero* is not due to fetal adrenal dysfunction.

To summarize, the study of Chavatte-Palmer (2002) indicates that there are some physiological indices that differ between apparently normal clones and controls during the transition from the perinatal to the early juvenile period. However, most of these differences were resolved within the first 15 days of age. Of the parameters that were different in clones, even the most persistent, elevated body temperature, was resolved after 50 days. The authors conclude that, based on their

data, apparently healthy clones should not be considered “physiologically normal animals until at least 50 days of age.”

In 2004, Chavatte-Palmer et al. published results from the first year of a three-year study designed to prospectively address the health of clones. They noted that for the first 65 days after birth, clones (n=25) had slightly lower hemoglobin levels than AI comparators (n=19), although the hemoglobin levels of the clones were still considered within the normal range. The lower levels persisted for the first 65 days after birth before reaching the same levels as the AI comparators. This finding reinforced the opinion of the group at INRA that clones could not be considered physiologically normal for the first two months of life.

Additional results from the three-year study were published by the group at INRA in 2007 (Heyman et al.). Twenty-one heifer clones of four different genotypes and 19 controls were studied between 4 and 36 months of age. Findings during the juvenile period will be discussed here, while observations relevant to reproductive development and the post-pubertal phase will be discussed in the appropriate developmental nodes. All but one animal survived the juvenile period of the study; one control heifer was euthanized at seven months due to extreme laxity of the flexor tendon. Using daily gain and feed intake as indicators, no differences in growth rate were observed between clones and control heifers up to 15 months of age. The authors state that repeated clinical evaluations revealed no differences in cardiovascular, respiratory and locomotive functions, clinical biochemistry, or immune parameters (data not shown).

It has been hypothesized that clones may be more sensitive to stress than conventional animals. To address this hypothesis, the group at INRA measured concentrations of cortisol and catecholamine metabolites every two months in a group of juvenile clones (n=5) and a group of age-matched controls (n=5). They found that although cortisol concentrations were more variable in clones, blood cortisol concentrations were similar between clones and controls during the juvenile period (4 to 12 months of age). Similarly, no differences were found between clones and controls in urinary concentrations of epinephrine and norepinephrine. The authors cite these findings as evidence that the clones were not suffering from chronic stress.

The University of Connecticut Studies: Govoni et al. 2002; Enright et al. 2002; and Savage et al. 2003.

Govoni et al. (2002) investigated the degree to which the somatotrophic axis⁸⁹ in Holstein clone heifers (n = 4) developed normally compared to AI-produced age-, gender- and breed-matched

⁸⁹ The somatotrophic axis governs the growth and development of the body. Growth hormone releasing hormone (GHRH), produced in the hypothalamus, stimulates production of growth hormone (GH) by the anterior pituitary gland. GH modulates systemic concentrations of IGF-I by binding to liver cells and stimulating

controls (n = 4). Serum samples were collected monthly from five to 14 months of age. Normal pulsatile patterns of GH secretion were observed in both clones and controls. Averaged across all time points, concentrations of GH were similar between clones and controls, but the patterns of secretion over time were different. GH levels declined in controls between five and 14 months of age, but concentrations of GH did not change in the clones during the same period, resulting in higher GH concentrations in clones at 9, 10, and 11 months of age. Parallel increases in IGF-I were observed in both groups over time, but clones had lower IGF-I concentrations compared to age matched controls from 5 to 11 months and at 14 months of age. As IGF-I is involved in development of ovarian follicles and uterine growth (Le Roith et al. 2001), lower circulating IGF-I levels may be partially responsible for the later onset at puberty observed in this group of clones (Enright et al. 2002). It is important to note, however, that the concentration of IGF-I required for normal sexual development is not known.

Clones secreted 5-fold more GH in response to GHRH compared to controls. When GHRH and somatostatin were administered together, there was less inhibition of GHRH-stimulated GH release in clone heifers compared to controls. Therefore, the elevated GH concentrations in the clones in this study may be indicative of an increased responsiveness to GHRH.

IGF Binding Proteins (IGFBPs) are responsible for transporting IGF in the blood and extend the half life of IGF-1. Concentrations of IGFBP-2 were static and not different between clone heifers and controls, but levels of IGFBP-3 were lower in clones compared to controls. This result parallels the lower concentrations of IGF-1 in the clone heifers.

Govoni et al. (2002) note that although they observed differences between clones and comparators in concentrations of GH, IGF-1, and IGFBP-3, concentrations of all three of these endocrine parameters were within the ranges previously reported in conventional cattle at similar ages. The authors therefore concluded that the clones exhibited age-appropriate development of the somatotrophic axis and, overall, clones and comparator heifers were developmentally similar over time. The authors also point out that the clones in their study were derived from a single nuclear donor (cow) with superior genetic merit (high milk production). There is evidence that genetic differences in cattle may be identified by GHRH-stimulated GH secretion (Lovendahl et al. 1996), and bulls of genetically superior bulls secrete more GH in response to GHRH (Kazmer et al. 1992; Zinn et al. 1994). Moreover, cows with high milk production have greater concentrations of GH than do cows with low production (Hart et al. 1975). Therefore, Govoni et al. (2002) speculate that the differences they observed in GH, IGF-1, and IGFBP-3 levels may be explained by the difference in genetic merit between the identical clones and their comparators.

production of IGF-I. Somatostatin, also produced in the hypothalamus, suppresses GH synthesis which in turn causes a reduction in IGF-I production in the liver. Somatostatin production is stimulated by high levels of IGF-I (Le Roith et al. 2001).

Savage et al. (2003) evaluated the behavior of the clones and age-matched controls described by Govoni et al. (2002). Between 32 and 36 weeks of age, there were no differences in weight or height between the clones (205.5 ± 9.9 kg; 117.0 ± 1.8 cm) and controls (211.4 ± 7.4 kg; 119.5 ± 1.4 cm). All calves were raised together under the same management conditions. Based on a series of studies evaluating approach to other animals and novel objects, clones exhibited age-appropriate behaviors, but were reported to be more aggressive and inquisitive than controls, and spent more time grooming and socializing. Clones tended to spend less time in playful behavior than controls. Review of records on the cow that served as the donor for the clones indicated that she had displayed similarly aggressive and inquisitive behavior as a young animal, suggesting that at least some of these behavioral traits may be genetically controlled. Clones spent more time in proximity to adult animals in an adjacent pen (which also housed the nuclear donor), and in proximity to the feed bunk compared to control animals. In general, clones were reported to spend more time with each other rather than socializing with control animals, with the authors speculating as to whether clones exhibit some form of genetic kinship recognition. Nonetheless, the overall conclusion of this study was that the clones behaved normally.

Other Studies

Wells et al. (2004) and Wells (2005) followed the growth and maturity of cattle clones generated at their facility in New Zealand through approximately four years of age. Approximately 80 percent of the clones delivered alive at term survived the first 24 hours of live. They reported that two-thirds of the 20 percent that died was due to spinal fractures syndrome or to deaths from dystocia, associated with LOS (Wells 2005). Another 15 clones died in the time period before weaning, most commonly of musculoskeletal abnormalities, including tendon contracture and chronic lameness, and umbilical infections, attributable to complications of LOS. They also reported two clones dying as the result of bloat, and an unspecified number of clones dying due to endophyte toxicity after eating fungus-infected ryegrass. Bloat and other gastrointestinal disorders have been reported by others (Cyagra 2003, Appendix E; Batchelder 2005), but also may result from feeding or grazing management problems. Wells et al. use the phrase “clonal family” to refer to clones derived from a particular donor, and note that the bloat and susceptibility to endophyte toxicity was restricted to one clone family, and likely due to their genetics. Another clone family consisting of three clones (and five half-siblings produced by AI) survived with no health anomalies and at the time of reporting was 18 months old. Other health problems observed during the juvenile period included anemia, chronic heart failure, and degenerative nephrosis, problems that have also been noted by other researchers (Chavatte-Palmer et al. 2004). Additional deaths were categorized as being due to misadventure and accidental deaths due to clostridial disease, parasitism, and over feeding. Surviving animals from

this group were characterized with respect to general health and physiological measurements; these are found in the discussion of Developmental Node 5 (Post-Pubertal Maturation).

Similar to Chavatte-Palmer et al. (2002), Batchelder (2005) also noted periodic moderate to severe hyperthermia in Holstein and Hereford clones up to 60 days of age. As with the Chavatte-Palmer clones, the Batchelder clones also showed no indication of infection, were unresponsive to anti-inflammatory drugs, and their behavior was unchanged; the hyperthermia also resolved spontaneously.

In their study of Japanese Black beef cattle clones described in the section on the Perinatal Developmental Node, Kato et al. (1998) reported that all of the clones that survived the perinatal period were alive and healthy at 85 and 120 days of age. In the subsequent study (Kato et al. 2000) of 13 clones that survived the perinatal period, 12 clones were alive and healthy at 117-350 days, and one clone died at three months “for no clear reason.”

Kubota et al. (2000), in their study of four surviving clones of a 17 year old Japanese Black bull, reported that the clones were 10-12 months of age at the time of publication. Based on veterinary examinations, growth curves, and 30 blood parameters no differences were found between the clones and their age-matched peers. No data were provided in the publication. Other groups have also reported normal growth rates for cattle clones (Wells et al. 2004; Heyman et al. 2004).

Yonai et al. (2005) (previously mentioned in the Perinatal Developmental Node) studied the growth of Holstein (n= 6) and Jersey (n=4) clones with shortened telomeres. Clones were given at least two liters of warmed colostrum immediately after birth, fed colostrum twice a day for the first five days of life, and monitored for physiological functions until they stabilized. Clones were fed according to the guidelines presented by the US National Research Council Nutrient Requirements of Dairy Cattle (1989). From Day 5 through Day 45, calves were given milk replacer twice daily, and offered calf starter pellets, hay and water during this time. After Day 45, all calves (clones and comparators) were weaned from milk replacer, and their feed gradually changed from calf starter pellet to formula feed over a two week period. Calves were fed 2-3 kg/day of formula feed, hay and water from Day 60 until one year of age. For the first 45 days after birth, the clones were reared in individual calf huts, after which they were reared together with other calves produced by AI or embryo transfer. Calves were held in a large pen in mixed groups of clones and age-matched comparators during the weaning period. After weaning, groups of 10-20 animals were moved into pens, and after one year of age, all animals were moved to a free-stall barn for heifers. Table VI-6 summarizes the average daily body weight gain of the clones from birth to two years of age. Body weights were collected monthly from birth to one year of age, and every three months between 15 and 24 months.

Table VI-6: Average Daily Gain (kg/day) for Holstein and Jersey Clones (source Yonai et al. 2005)									
months of age									
	0-3	3-6	6-9	9-12	12-15	15-18	18-21	21-24	
Jersey Clones (n=4)									
Mean	0.49	0.73	0.67	0.53	0.49	0.56	0.51	0.40	
SD	0.02	0.02	0.11	0.06	0.05	0.17	0.16	0.18	
Holstein Clones (n = 6)									
Mean	0.72	1.17	0.82	0.85	0.90	0.97	0.68	0.58	
SD	0.14	0.12	0.08	0.11	0.10	0.26	0.11	0.27	
SD = Standard Deviation									

The authors report that the average daily gain for the clones was greater than that of the standard of each breed. For the Holstein clones, the average bodyweights conformed to the standard during the first three months of age, but exceeded the standard after five months, while the Jersey clones exceeded the body weight of the Japanese Feeding Standard for Dairy Cows throughout the measured time period. The Holstein clones' body weights were approximately equivalent to that of the donor animal until 18 months of age, but exceeded it thereafter. The Jersey clones exceeded the body weights of the donor from birth to two years of age. The animals were reported as healthy with normal growth throughout this time period. No deaths were reported after the perinatal period.

There are other reports of clones that appear to be healthy at birth but unexpectedly die some time later. Gibbons et al. (2002), for example, reported a clone dying at 60 days of age due to respiratory and digestive problems. As mentioned above, Kato et al. (2000) also reported the death of a clone at 3 months. Chavatte-Palmer et al. 2004; Wells et al. 2004; Batchelder 2005 have also noted early deaths, but their cause(s) have not been clearly linked to cloning. The degree to which these unexpected deaths in cattle are related to cloning, or some disease process that is independent of cloning, is not clear. Ogunuki et al. (2002) have noted shorter life spans in some of their mouse clones; the cause of death appears to be due to liver damage, pneumonia, or neoplasia. The relevance of mouse models to domestic livestock has been discussed in Chapter IV.

(b) Cyagra Data: 1-6 Month Age Cohort

The calves from the Cyagra dataset most closely correlating to the Juvenile Developmental Node are the 46 clones and 47 comparators found in the 1-6 month of age group. Tables E-200a and E-200b, and Charts E-200, E-201, E-202, E-210, E-211, and E-212 describe CVM's analyses of the information.

In general, these clones appeared normal, although some anomalies were noted on physical examination. These may be related either to cloning or to the genetics of the animal that was being propagated. None are unique to clones, although their frequency appears to be higher in clones than in calves produced using other forms of reproduction (see Chapter V). One of the clones was culled for poor conformation (the physical appearance of the animal), a matter of potential business importance to the producer, but likely having no impact on either food or animal health. Conventional animals with poor conformation are generally not used in selective breeding programs, and may be culled; it is likely that breeders will put similar limitations on clones as well. Several of the clones experienced serious problems resulting from umbilical abnormalities, including enlargement, excessive bleeding, and infection of the navel. These were resolved surgically. In addition, three cases of cryptorchidism (undescended testicle) were identified in calves from the same cell line. Although this condition is relatively uncommon in conventional animals, it is observed with some frequency, and is thought to be hereditary.

Interestingly, three clones derived from the same Jersey cow cell line presented with very different phenotypes. Clones # 87, 88, and 89 were within 10 days of age of each other when they were weighed and blood samples drawn (131-141 days old). All three required umbilical surgery. The oldest, clone #87, weighed 282 pounds. Clone #88, who was a day younger, weighed 197 pounds, and the youngest (at 131 days of age) weighed 215 pounds. Otherwise, the animals were healthy on physical examination. A fourth clone from this cell line died at birth from LOS-related complications.

Measurements of analyte levels in the entire 1-6 month old cohort were generally very close to those measured in the comparators (Chart E-201). In aggregate, 96 percent of the total analyte values for clones were within the range of the comparators. A few were out of range: glucose values were above the range of the comparators in six of the 42 likely valid measurements (four were considered artifactual). In order to determine whether the hyperglycemia was transient or sustained, urinalysis results were checked for the clones with elevated blood glucose levels. As none of those tests were positive for glucose (the renal threshold for glucose in cattle is approximately 100 mg/dl: *i.e.*, if blood levels of glucose exceed 100 mg/dl for any appreciable time, glucose spills over into the urine), it is unlikely that the higher blood glucose levels (88-123) had been sustained long enough to allow for spillover into the urine. Most likely, these were transient elevations resulting from proximity to a meal or as a short-lived response to stress (as in being restrained for blood draws).

The hemograms for the cohort did not reveal any significant health concerns. None of the clones were anemic, and there was no depression of cellular immune function. Some of the clones had individual values that were outside the range of the comparators, but these were not judged to

pose either an animal health or food consumption risk (see Appendix E for a more complete discussion).

It is important to note that although this time period appears to be relatively short, it spans an important developmental transition period for ruminants. Calves that are closer to one month of age are still primarily milk-fed, while those closer to six months of age have mostly transitioned to a more adult diet, and function as ruminants. The youngest animals are in a very rapid growth phase, while the older animals in the range, although still growing, are doing so at a slower rate. Because young animals are growing rapidly, measures of bone growth such as calcium, phosphate, and alkaline phosphatase might be expected to be higher in younger compared to older animals. Comparison of both the clone and comparator laboratory values to the Cornell Reference Range (which is derived from adult cattle) (Charts E-200 and E-202) indicates that many of the clones and comparators exhibit calcium, phosphate, and alkaline phosphatase levels that exceed the Cornell Reference Range. This finding is consistent with higher rates of growth in young calves relative to adults, and provides confidence that clones and comparators are exhibiting similar, normal physiological responses to growth stimuli. Review of Chart E-201 reveals that clone alkaline phosphatase values are almost entirely within the range of the comparators (38 of 46 values). Most of the clones whose alkaline phosphatase levels exceeded the comparator range were the youngest animals.

Another set of physiological parameters that varies with age can be seen in total protein, globulin, and albumin levels. These measurements reflect, among other things, the immune status of the animal. Immediately after birth, globulin levels, which are largely comprised of immunoglobulins, are derived almost entirely from colostrum (the antibody-rich first “milk” to be secreted by mammals). “Passive immunity” is conferred by the ingestion and intestinal absorption of immunoglobulin-rich maternal colostrum. In the two to four months after birth, the calf’s own immune system begins to develop its production of immunoglobulins, as the circulating supply of maternally-derived immunoglobulins in the calf’s blood wanes. This phenomenon can be observed in Charts E-200 and E-202 (Clones: Reference Range (1 to 6 months) and Comparator Population: Reference Range). Clone and comparator globulin values are low relative to the Cornell lab reference range because that reference range is derived from adult animals with fully functional endogenous immunoglobulin production. The clone and comparator calves in this cohort have not fully started to produce their own antibodies from their own B-lymphocytes. Review of Chart E-201 (Comparison of Clones to Comparator Population), however, indicates that there were few differences between the clones and the comparator population, reflecting the appropriate age-related lag accompanying the decrease in passive acquired immunity and endogenous immunoglobulin production. The globulin levels that are different between clones and comparators reflect this age-related physiological phenomenon. Clones #72 and 73 were among the youngest in the one to six month old group, and thus would

be expected to have lower globulin levels. Comparison of the globulin value for clone #100 (174 days of age, globulin of 4.6g/dL) with clone #72 (48 days of age and globulin level of 1.6 g/dL) clearly demonstrates the age-related changes in the analyte, and appropriately reflects the normal developmental increase in endogenous globulin production.

Sub-Cohort Analysis

Examination of the subcohort of seven clones (# 71, 72, 73, 78, 79, 119, 132) at two time frames (birth and the 1-6 months of age) allows the determination that appropriate age-related physiological changes are occurring in the clones on an individual animal basis, rather than on a population basis. For example, gamma glutamyl transferase (GGT) values appear low relative to comparators in “within 24 hours of birth” time period for four of these seven clones. This likely reflects the difference in timing between when the blood samples were drawn for clones and comparators (Clones had their blood samples drawn prior to colostrum administration, while comparators had their blood drawn some time after being fed colostrum). As colostrum has high intrinsic GGT activity, the difference between the two groups may be due to its effective absorption of GGT by the comparators. GGT values normalized by the time of the second blood draw for three of these animals, and were only slightly lower (4U/L vs. the comparator range of 5-32 U/L) in the remaining clone at Day 48.

At birth, some of the clones in this sub-cohort had measures of liver function out of the comparator range (lower AST, and low bile acid or cholesterol levels). Low cholesterol is associated with retained fetal circulation in the livers of young animals. Were these low cholesterol levels to continue into the next developmental node, there might be cause for concern, but given that they normalized at the time of the second blood draw, there is little reason to expect that the lower values in these very young clones pose a health risk. The low levels at birth are more likely a reflection of the changeover from fetal to neonatal circulation, possibly exacerbated by the clones’ unusually large umbilical vessels, which often required surgical correction. The lower bile acid and AST values observed would also be related to the transition from fetal to neonatal circulation, and are not indicative of any disease state. All of these values normalized by the second measurement, as did additional analyte levels that were out of range for individual clones perinatally (low CK, TIBC, and iron). These measurements reflect normal adaptive physiological processes and not pathologic or disease states, and instead provide evidence of the “normalization” of the clones as they matured.

A few laboratory measurements appeared outside the range of the comparators in some of the clones at the time of the second measurement, but these do not appear to have clinical relevance. Complete blood count information is only available for four of the seven clones measured at both

time points, and do not appear to be reflective of clinical problems. For a more complete discussion of these data, see Appendix E.

(c) Unpublished data

Full hematology and clinical chemistry screens on three pre-pubertal bull clones (aged 5 to 7 months old) were shared with CVM by a private veterinary clinic (Appendix G, Tables G-3a-3c). The clones were described as being clinically, physically and behaviorally normal, with normal growth rates and size. Blood samples were taken three times over a six week period. All of the clinical chemistry data, with the exception of one, were within normal published ranges or within the comparator range for the testing laboratory. Just as for the physiological data shared by Cyagra, the reference range for the testing laboratory was for an older cohort of animals (that were also female), and were not age-appropriate. The one analyte that fell outside a reference ranges occurred in a single sample in one bull clone, and was a low cholesterol value. All measurements in the subsequent sample from this bull clone were within normal ranges.

(d) Summary for Juvenile Developmental Node in Bovine Clones (Developmental Node 3)

Numerous studies have provided information about the physiology of bovine clones during the juvenile period. Some juvenile bovine clones exhibiting LOS at birth eventually succumb to its sequellae. Surviving clones, however, appear to grow and develop normally. Detailed review of laboratory results from several studies indicates that, overall, the physiology of juvenile clones reflects normal, appropriate responses to ongoing growth and developmental signals, and for most of the juvenile period, bovine clones are functionally indistinguishable from non-clones. A possible exception is the period of transition from Perinatal to the Juvenile node, during which some physiological differences between clones and conventional cattle have been identified. Almost all of these differences are transient and are resolved in the first two weeks of life. Thermoregulation in clone calves appears to normalize within two months of birth (Chavatte-Palmer et al. 2002; Cyagra 2003). Although there may be some physiological differences between clones and their comparators during the transition between the perinatal and juvenile developmental nodes, none of these differences indicate the presence of any subtle or frank food consumption hazards.

iv. Reproductive Development and Function in Bovine Clones (Developmental Node 4)

(a) Peer-reviewed Publications

The number of studies that explicitly address the reproductive function of bovine clones is smaller than studies of other endpoints. Puberty onset has been reported as either “within normal

limits” or somewhat (days) later in clones than controls. The Cyagra data received do not explicitly address the question of puberty onset or reproductive capability.

Reproductive Function of Female Clones

In a study of reproductive function in bovine clones, Enright et al. (2002) at the University of Connecticut evaluated the same clones and controls previously reported on by Xue et al. (2002) and Govoni et al. (2002). They reported that heifer clones reached puberty at a later age than controls (314.7 ± 9.6 days vs. 272 ± 4.4 days), and were reported as having higher body weights at first estrus (336.7 ± 13 vs. 302.8 ± 4.5 kg). No differences were noted between clones and controls in estrous cycle length, development of ovarian follicles, or profiles of hormonal changes. Three of the four clones and all four control heifers became pregnant following AI, although number of inseminations was not reported. Daily hormone profiles of luteinizing hormone (LH), follicle stimulating hormone (FSH), estradiol, and progesterone were similar between clones and controls. The cause of reproductive failure in one clone could not be determined; although this animal had reproductive hormone profiles similar to the other animals in the study, and no physical abnormalities could be found upon veterinary examination, poor signs of estrus were observed. This heifer did eventually conceive and produce a calf (Tian et al. 2005, further discussed below). The cause for the later age and higher weight of clones at the time of puberty is difficult to explain. The authors speculated that as the later onset of puberty can be genetically controlled in some cattle breeds, these clones may be expressing the genetics of the donor animal. Given that no records of age at puberty were kept for the donor cow, it is not possible to draw any conclusions regarding that hypothesis.

Heyman et al. (2002a) reported that from a group of clones derived from adult cells, five remaining animals were healthy and normal (one clone died of severe anemia (Renard et al. 1999), as previously discussed in the Perinatal section). They noted that some of the females were more than one year old at the time of publication and were cycling normally, but no data were provided. In a follow-up study (Heyman et al. 2004) the authors stated that female clones at the INRA facility generally began cycling at 10 months of age, and demonstrated estrous behavior by 12 months of age, within the normal range for their breed (Holstein). Ten female clones were bred by AI to the same non-clone bull. All 10 heifers conceived and produced live, apparently normal calves. Birth weight of progeny was 43.9 ± 4.1 kg, and gestation length was 281 ± 3.9 days, within the normal range for Holstein cattle.

The timing of puberty in clones at the INRA facility was compared to control heifers by Heyman et al. (2007). From eight to 15 months of age, 10 clones of three different genotypes and 11 AI-derived control heifers were observed twice daily for estrus, and plasma progesterone was measured at 10-day intervals. Onset of estrous cycles occurred 63 days later in clones compared

to control heifers (419.3 ± 42.5 days vs. 356.5 ± 50.5 days, respectively) and cloned heifers were 56 kg heavier than controls at the time of puberty (359.0 ± 38.9 kg vs. 303.0 ± 22.3 kg, respectively). These differences cannot be attributed to differences in growth rate since daily gain and feed intake up to 15 months of age were similar between groups. Mean estrous cycle length was similar between clones and controls. These results are consistent with those from the cohort of clones at the University of Connecticut (Enright et al. 2002) and provide further evidence, at least for Holstein heifers, that cloning may alter the timing of puberty.

Wells et al. (2004) reported conception rate to two AI was 83 percent (25/30) for Holstein heifer clones, compared to 90 percent (9/10) for as small group of heifers produced by AI. Gestation length was slightly longer for clones ($n=16$) than for nine comparators (287 ± 3 vs. 281 ± 3 days), but within the normal range for Holsteins. Wells (2005) notes that despite variations in gestation length, only conventional levels of animal management and husbandry are required for the calving of heifer clones, indicating that the signals for induction of parturition and actual birth are functioning appropriately. Although most of the clones were separated from their offspring soon after birth, as is conventional in dairy practice, those dams that were not separated from their progeny exhibited normal maternal behavior and successfully reared their young.

Forsberg et al. (2002) reported that Gene, the bull calf described previously, matured into a “healthy, fertile bull that has sired calves by artificial insemination and *in vitro* fertilization.” Specific data on measures of reproductive function were not provided.

Kato et al. (2000) report that one of the clones derived from a Holstein cumulus cell was artificially inseminated, conceived, and gave birth to a normal calf

The University of Connecticut (Tian et al. 2005) also reported first lactation milk yields and SCC for four clones and their non-clone comparators, indicating that lactation curves were similar for both groups. Total milk production for the first lactation was not different between clones and comparators ($8,646 \pm 743.8$ kg vs. $9,507.8 \pm 743.8$ kg). One clone gave birth prematurely to a stillborn calf, did not have complete udder development, and produced approximately 30 percent less milk during her first lactation compared to her clone mates. Overall, SCC was low for both clones and comparators (based on Figure 2b of the paper: $\sim 40 \times 10^3$ vs. 35×10^3 cells/mL), indicating a functional immune system, mammary gland, and low disease incidence. The role of good husbandry can also not be ruled out in this observation.

Heyman et al. (2007) reported that one of the 21 heifer clones heifers in their three-year prospective study at INRA died shortly after calving. The exact cause of death is not specified, but the authors point out that this heifer calved during the unusually hot summer months of 2003.

Yonai et al. 2005

Study overview: In the most comprehensive study of reproductive function in cattle clones, Yonai et al. (2005) (previously mentioned in other Developmental Nodes) performed an extensive analysis of reproductive performance in Holstein and Jersey clones with shortened telomeres, including puberty onset, estrus behavior, hormone cycling, the appearance of follicular waves, fertility and birthing for three estrus cycles. Once puberty onset had been determined, ovulation and formation of *corpora lutea* were monitored thrice weekly, with plasma samples to monitor progesterone levels collected every three days. After puberty, the estrous behavior of the clones was monitored twice daily until the animals became pregnant, with the length of the estrous cycles and occurrence of standing estrus recorded. Plasma samples and ultrasonography were used to identify follicular waves and monitor progesterone and 17- β estradiol concentrations between day 18 of estrus and the day of ovulation over 17 estrous cycles in the Jersey clones and 28 estrous cycles in the Holstein clones. All clones were bred by artificial insemination using semen from the same lot of one bull (breed unspecified). Pregnancies were diagnosed by ultrasonography at 40 days after AI. For the first and second postpartum cycles, all clones were artificially inseminated at first estrus, which usually occurred 90 days after parturition. The length of gestation and resulting calves' birth weights were recorded. Table VI-7 summarizes the data collected in this very detailed study.

Table VI-7: Reproductive Parameters Evaluated for Jersey and Holstein Clones (adapted from Yonai et al. 2005)	
Parameter	Mean \pm Standard Deviation
Jerseys (n = 4)	
Age at puberty	-
<i>Reproductive records from puberty to first parturition</i>	
Length of estrous cycle ¹ (days)	20.2 \pm 1.4
Follicle waves per cycle ¹ (number)	2.3 \pm 0.8
Plasma estradiol-17 β concentration on estrous day ²	
Detectable (17/17 cycles; pg/ml)	8.12 \pm 2.40
Not detectable (0/17 cycles; pg/ml)	-
Plasma progesterone under the curve ³ (ng/ml per cycle)	190.6 \pm 59.4
Number of AI for first conception	2.3 \pm 1.9
Age at first conception (days)	503 \pm 54.9
Gestation period (days)	279 \pm 2.5
Calf weight (first parturition) (kg)	22.0 \pm 2.1
<i>Reproductive records after first parturition</i>	
Interval from parturition to first ovulation (days)	51.3 \pm 42.8
Interval from parturition to first estrus (days)	85.0 \pm 52.7
Number of AI for second conception	1.3 \pm 0.5

Table VI-7: Reproductive Parameters Evaluated for Jersey and Holstein Clones (adapted from Yonai et al. 2005)	
Interval from parturition to second conception (days)	115 ± 16.8
Age of second conception (days)	897 ± 44.8
Calf weight (second parturition) (kg)	26.4 ± 1.1
Reproductive records after second parturition	
Interval from parturition to first ovulation (days)	32.5 ± 19.3
Interval from parturition to first estrus (days)	50.0 ± 27.8
Number of AI for third conception	1.5 ± 1.0
Interval from parturition to third conception (days)	129 ± 49.9
Age of third conception (days)	1,304 ± 46.6
Holsteins (n = 6)	
Age at puberty	323 ± 0.6
Reproductive records from puberty to first parturition	
Length of estrous cycle ⁴ (days)	20.3 ± 1.5
Follicle waves per cycle ⁴ (number)	2.3 ± 0.7
Plasma estradiol-17 β concentration on estrous day ⁵	
Detectable (19/28 cycles; pg/ml)	6.94 ± 2.64
Not detectable (9/28 cycles; pg/ml)	3.95 ± 1.74
Plasma progesterone under the curve ⁶ (ng/ml per cycle)	154.0 ± 58.0
Number of AI for first conception	2.0 ± 2.0
Age at first conception (days)	481 ± 35.0
Gestation period (days)	277 ± 5.8
Calf weight (first parturition) (kg)	37.8 ± 5.0
Reproductive records after first parturition	
Interval from parturition to first ovulation (days)	56.0 ± 41.5
Interval from parturition to first estrus (days)	86.0 ± 33.0
Number of AI for second conception	1.2 ± 0.4
Interval from parturition to second conception (days)	126 ± 41.7
Age of second conception (days)	881 ± 61.7
Calf weight (second parturition) (kg)	44.2 ± 1.9
Reproductive records after second parturition	
Interval from parturition to first ovulation (days)	79.3 ± 18.9
Interval from parturition to first estrus (days)	92.3 ± 19.2
Number of AI for third conception	1.3 ± 0.5
Interval from parturition to third conception (days)	138 ± 34.9
Age of third conception (days)	1,297 ± 75.0
¹ Twenty-six estrous cycles in four cloned heifers were included.	
² Plasma samples were collected from 17 estrous cycles in four cloned heifers.	
³ Plasma samples were collected every three days during the 26 estrous cycles.	
⁴ Thirty-three estrous samples in five cloned heifers were included.	
⁵ Plasma samples were collected from 28 estrous cycles in five cloned heifers.	
⁶ Plasma samples were collected every three days during the 33 estrous cycles.	

Reproductive function: First Estrus: Yonai et al. grouped their analysis of reproductive function into three stages: pubertal, post-pubertal conception and gestation, and post-parturition, including rebreeding. Although some of the clones entered puberty prior to the initiation of this stage of the study, Yonai et al. reported that changes in plasma progesterone were consistent with previous reports on puberty in conventional cows. They also reported that *corpus luteum* formation was consistent with that reported in conventionally bred cows, and that the clones exhibited appropriate estrous behavior at puberty. Overall, the observations at puberty indicated that these clones exhibited normal early reproductive development. With respect to post-pubertal maturation of the heifer clones, Yonai et al. noted that there was some difficulty detecting estrus by behavior in the Holstein heifer clones, and that there were differences in their estradiol levels, these were consistent with similar observations in conventionally bred Holstein heifers. There were no difficulties in observing estrus in the Jersey clones. Estrous cycle lengths in both clone lines were comparable to those observed in conventionally bred cattle. Additionally, the levels of progesterone secretion per cycle were reported as similar to those of conventionally bred heifers, which the authors interpreted as normal post-pubertal *corpus luteum* function. They conclude that the estrous cycles of the heifer clones were normal.

All of the heifers conceived upon artificial insemination, although one heifer clone and one comparator needed multiple cycles of insemination; the remaining clones and comparators all conceived after no more than two rounds of AI. All of the clones but one Holstein delivered healthy, live calves. The exception delivered a stillborn calf two weeks before expected parturition. No obvious abnormalities were observed in the stillborn. Two of the Holstein clones required limited assistance for delivery; the remaining Holsteins and all the Jersey clones did not require any assistance in delivery. The average gestational periods were normal for the clones and all of the resulting calves were within normal body weight ranges for their breeds. All the live-born calves were reported as being normal.

Second and Third Estrus. Yonai et al. noted a wide variation in the interval between parturition and first post-partum ovulation and estrus. The first postpartum ovulation in the Holstein clones occurred between 14 and 188 day (Table VI-7), and between 11 and 108 days in Jersey clones; the interval between parturition to first estrus was between 62 and 149 days for the Holstein clones, and 30 and 135 days for the Jersey clones. All clones had confirmed follicular waves, and pregnancy ensued in all of the clones following an average of 1.2 and 1.3 rounds AI for the Holstein and Jersey clones, respectively. The second parturition was largely uneventful for all of the clones, with one Holstein requiring minimal assistance calving. Gestation times for the all of the clones fell within normal ranges for the breeds; all of the calves had normal body weights,

appeared to be normal at birth, and survived. Similar responses were noted for the third conception.

Table VI-8: Results of Milk Yield in First and Second Lactations of Jersey and Holstein Clones (adapted from Yonai et al. 2005)	
Animal	Milk Yield
Jerseys (n = 4)	
<i>First Lactation</i>	
Clone 1	5,637.4
Clone 2	6,077.9
Clone 3	6,272.6
Clone 4	5,597.7
Mean ± Standard Deviation	5,896.4 ± 332.0
Donor Animal	5,064.0
<i>Second Lactation</i>	
Clone 1	7,006.8
Clone 2	7,539.2
Clone 3	7,309.6
Clone 4	7,195.6
Mean ± Standard Deviation	7,262.8 ± 222.6
Donor Animal	6,087.0
Holsteins (n = 6)	
<i>First Lactation</i>	
Clone 1	8,591.2
Clone 2	9,219.5
Clone 3	9,586.5
Clone 4	9,836.0
Clone 5	9,029.1
Clone 6	9,735.6
Mean ± Standard Deviation	9,333.0 ± 476.4
Donor Animal	10,968.0
<i>Second Lactation</i>	
Clone 1	10,678.6
Clone 2	12,402.6
Clone 3	11,341.4
Clone 4	10,376.0
Clone 5	10,110.2
Clone 6	12,719.4
Mean ± Standard Deviation	11,271.4 ± 1084.7
Donor Animal	11,442.0

Milk Production. Table VI-8 summarizes the yield of milk produced by the clones and their half-siblings and donor for the two lactation cycles following the first and second calvings. Data on the composition of this milk are addressed in the Food Composition portion of this chapter. Milk yield, although varying among the clones, was within the normal range for each breed for each lactation cycle. Interestingly, the Holstein clones produced less milk on average than their nuclear donor animal, while the Jersey clones produced more milk on average than their nuclear donor. The authors reported that mastitis was observed in the Holstein group of clones in two animals towards the end of the lactation cycle, and bloat was observed in two clones (not specified if the same animals) at approximately 130 days post-parturition. Neither was observed in the Jersey clones. Although not specified, the affected animals were most likely treated, and appear to have recovered as the number of animals did not change between cycles.

This study, which is the first to study multiple cycles of reproductive function in any species of clone provides detailed information on both the individual physiological parameters measuring growth and reproduction (including lactation), as well as integrated measures of those functions. The authors conclude that despite the observation that all of these clones had shortened telomeres, these Holstein and Jersey clones exhibited normal growth, reproductive and lactation characteristics.

Other Studies

Although Lanza et al. (2001) reported on transgenic clones, conception rates for female clones after AI were high, with 87.5 percent of the animals conceiving on the first insemination and the remainder conceiving on the second insemination attempt. The two transgenic clones that had given birth, as of the publication date, were reported to have delivered calves that appeared normal in all respects, although no specific data are provided.

Pace et al. (2002) reported that heifers began to display signs of reaching puberty at 10-11 months of age, within the normal age range of conventional Holstein heifers (9 to 12 months). They further report that all of the heifer clones that were inseminated (n=22) became pregnant, and calved at the age of 23-25 months, similar to non-clone cattle (approximately 75 percent of the cattle in Pace et al. (2002) were transgenic). No specific information on gestation length or health of the progeny was provided. Analysis of the milk from non-transgenic clones of this cohort (Walsh et al. 2003) is presented within Section 3 of this Chapter.

In an abstract, Aoki et al. (2003) present a preliminary report on the milk and milking behavior of two first-lactation Holstein clones derived from somatic cells isolated from the colostrum of mammary gland epithelial (MGE) cells described by Kishi et al. (2000), previously discussed in

the Perinatal section. These two clones were housed near the same automatic milking system as eight second-lactation control cows produced by AI. Comparisons were made between first lactation clones and second lactation controls. These cow clones were apparently followed for at least two calvings, and results were reported for the first through third post-partum ovulation and follicular development per estrous cycle. First postpartum ovulation was delayed in both of the clones, as well as the interval between the first to second postpartum ovulation. Clones were reported to have had two waves of follicular development per cycle. Both clones and comparator cattle were reported to calve normally, and did not appear to have different body weights and body condition scores, although no data were provided. The authors did not report differences between gestation length and duration of estrous cycle. They concluded that the clones were “normal in regard to delivery, lactation, and growth, and were similar in regard to the functions of their reproductive physiology.” Differences were observed, however, in the milking behavior, including the number of times that they voluntarily entered the automatic milking system relative to controls. In general, first lactation animals lack experience with milking equipment, and produce less milk than second and later parity cows, which likely contributed to differences in milking behavior between the two groups (Vasconcelos et al. 2004; Flis and Wattiaux 2005). Given that this is an abstract, the number of animals is very small, and the difference in the total number of lactation cycles the cows had experienced, the significance of the observation is unclear. Presentation of these data in a complete publication would aid this risk assessment and other analyses of clones.

Heyman et al. (2004) reported that first lactation milk yields ($9,341 \pm 304$ kg vs. $8,319 \pm 1,800$ kg for a 305 day lactation) and somatic cell counts (SCC), which are a measure of mammary gland health) for three female Holstein clones were similar to those of three age-matched non-clone comparators. Somatic cell counts for both clones and comparators ($116 \pm 103 \times 10^3$ vs. $113 \pm 50 \times 10^3$) were well below the level indicative of subclinical mastitis ($1,000 \times 10^3$), and the SCC limit cited by the Pasteurized Milk Ordinance for fluid milk entering commerce.

Reproductive Function of Male Clones

The reproductive function of male bovine clones has also been studied. Wells (2005) reported on the reproductive function of six bulls cloned from the same steer. The rates of *in vitro* embryo development following fertilization of abattoir-derived oocytes using sperm from these sires varied among the sires, but the development of blastocysts to quality grades suitable for embryo transfer were similar to that for four comparator bulls (10-25 percent for the clones and 13-30 percent for the comparators). Likewise, Heyman et al. (2004) reported that three clones of an eight year old bull were enrolled in an AI center, and semen was collected when the clones were between 13 and 15 months of age. Percentages of normal sperm, cleavage rate and blastocyst rate following IVF were not different between the clones and their nuclear donor. Results of AI

trials were only presented for one clone (no comparator). Forty-one cows became pregnant out of 63 animals inseminated, yielding a 65 percent pregnancy rate. Two pregnancies were lost by day 90 (5 percent loss). Only 26 pregnancies were allowed to go to term, yielding 25 live, healthy calves and one stillborn.

Shiga et al. (2005) reported on the semen quality of two clones of a 12 year old Japanese Black bull. Semen was collected over a four month period beginning when the clones were approximately 12 months old. Comparisons were made using frozen semen from the nuclear donor and using averages for the breed. Although ejaculate volumes of the two bulls were lower than the range for the breed (2.34 and 2.76 mL vs. 5-8 mL), sperm concentration, pH, and pre-freezing motility were within established ranges for Japanese Black bulls. Development of IVF embryos to the blastocyst stage was not different between clones and their nuclear donor (23.4 and 28.4 vs. 30.9 percent). Semen from one of the clones was used to inseminate 22 cows, compared to 102 cows inseminated by the nuclear donor. Pregnancy rates were similar between the clone semen and semen from the nuclear donor (54.5 vs. 62.7 percent). Two of the 12 (17 percent) resulting pregnancies from the clone aborted spontaneously in mid-pregnancy, compared to 5/64 (8 percent) abortions among the cows bred by the nuclear donor.

Semen and reproductive profiles of 3 cloned Holstein-Friesian bulls were reported by Tecirlioglu et al. (2006). Development of the reproductive organs and scrotal circumference were reported to be normal in the clones. Semen was collected from these clones at 16-18 months of age. Sperm morphology was similar between clones and their nuclear donors. One bull clone had lower motility of spermatozoa in fresh semen compared to its nuclear donor. Sperm velocity parameters in fresh semen were higher in clones compared to donors, but this difference was not observed in frozen-thawed semen. When used for *in vitro* fertilization, semen from clones resulted in higher cleavage rates (≥ 85 percent in clones vs. ≤ 66 percent for donors). The proportion of cleaved embryos that developed into blastocysts and the quality of blastocysts, however, were similar between clones and their donors. There were no differences between clones and donors in either pregnancy rate or offspring rate, but the authors point out that their animal numbers (a total of 26 calves sired by clones and donor bulls) were too small to conduct a valid statistical comparison for these parameters. No phenotypic abnormalities were observed in any of the calves sired by the clones.

Circulating testosterone concentrations in the three clone bulls (26.5 ± 2.6 nmol/L) were higher compared to age-matched controls (6.1 ± 1.4 nmol/L). Reasons for this difference are unclear, but Tecirlioglu et al. (2006) speculate that testosterone concentrations in control bulls may have been suppressed by elevated glucocorticoids due to handling stress whereas clones were more accustomed to blood sampling and interaction with humans. While the authors considered the results of this study to be preliminary due to the limited numbers of animals and traits

investigated, they concluded that the reproductive potential of cloned bulls is not different from their genetic donors.

(b) Unpublished data

Semen evaluations on four healthy post-pubertal clones derived from an Angus-Chianina nuclear donor cross were shared with CVM (Chapter 5, Table V-17). Semen was collected by a commercial reproduction service from May through June 2003, three times daily, the usual industry practice. The age of the bulls at the time of collection was not recorded. Semen evaluation showed that one clone had a low sperm concentration (average 169.5×10^6 cells/ml vs. the normal range $800\text{-}1,200 \times 10^6$ sperm/mL (Sorenson 1979; Beardon and Fuquay 1980; Hafez and Hafez 2000)) and low percentage of normal sperm (between 2 and 8 percent) during the observation period. This bull likely would have failed a breeding soundness exam, and if it had been a conventional animal, it would most likely have been sold to a feedlot for eventual slaughter. A second bull clone had marginal semen quality, and might have been retained depending on the perceived value of his genetics. The remaining two clones exhibited acceptable semen characteristics, and would likely have been retained for breeding.

Galli et al. (unpublished data) also presented data on breeding soundness and performance of three clones of a Holstein bull (Chapter V, Table V-10). Breeding soundness exams indicated that clones were acceptable for breeding. Artificial insemination trials using semen from one of the clones on four farms resulted in pregnancy rates ranging from 33 to 80 percent; however, few cows were actually bred ($n=63$ for all farms combined), there were no contemporary comparators used, and no details regarding farm management were provided, making these data difficult to interpret. Pregnancy rates to AI for this clone were within the range of the U.S. average for Holstein cattle.

(c) Summary Statement for Reproductive Development and Function in Bovine Clones (Developmental Node 4)

Although specific animals are rarely cited, all reports of reproductive function in bovine clones appear to indicate that the animals respond normally to developmental signals governing puberty onset and that they subsequently reproduce effectively. The results of the study by Yonai et al. (2005) provide further confidence by reporting on detailed physiological parameters required for successful reproduction, and demonstrate that the clones continued to cycle and function normally after the first pregnancy. The studies of lactation and milk yield indicate a consistent response demonstrating that these animals function normally post-partum and during subsequent reproductive cycles. Reproductive failure is a common phenomenon in conventional cattle, and among one of the most frequent causes for culling. Although cases of reproductive failure have

been reported among clones, they are not unusual among conventional cattle, and do not raise food safety concerns. Reproductive function is among the most tightly regulated functions that a mammal performs; the demonstration that clones can reproduce normally appears to indicate that those clones are functioning normally for this biological criterion.

v. Post-Pubertal Maturation in Bovine Clones (Developmental Node 5)

(a) Peer-reviewed Publications

Post-pubertal maturation includes the very long period of time between the development of reproductive capacity and the natural end of the animal's life. Most cattle in US agriculture never reach the end of their "natural" life-spans for economic reasons. In commercial dairy establishments, dairy cows are sent to slaughter some time between the end of their third to fifth lactations, or sooner, depending on their health and productivity. Beef cattle that are not being used for breeding are generally sent to slaughter when they reach about 1,000 to 1,400 lbs, or at approximately 18 to 24 months of age (depending on breed, season, environmental conditions, etc.). Most of the possible food consumption risks arising from edible products of clones (e.g., milk or meat) would occur during this Developmental Node.

We have not conducted a survey of clone producers or the investigators who have published on the health status of clones earlier in the clones' lives to determine their vital or health status. At this time, there are economic disadvantages to maintaining healthy clones without being able to realize financial investments, so many otherwise healthy clones have been euthanized. The following discussion therefore summarizes reports that have been obtained from the literature, and tends to focus on anomalies that have been noted.

Kato et al. (2000) reported that as of September 1, 1999, all of the surviving clones from their Holstein and Japanese Black cumulus cell and fibroblast donors were healthy and aged 117-350 days. No further publications were found regarding the fate of these animals.

Because of the relatively short time that cloning has been practiced, (Gene, the first bovine SCNT clone was born in 1997 (Cibelli et al. 1998)), little information is available on animals during this developmental phase, and much of that information comes in the form of single sentences or short mentions in journal articles that address some other issue. Abnormalities that have been noted in mature cattle clones appear to be sequellae of anomalies or defects noted earlier in life, and may be related to LOS or other earlier diseases. For example, Batchelder (2005) reported that one clone died suddenly at 25 months of age. Necropsy results indicated severe trace mineral deficiency (selenium and copper) as the cause of death. None of the non-clone cattle grazing the same pasture developed signs of mineral deficiencies. Nonetheless, this particular clone was reported to have exhibited frequent but mild signs of bloat as a juvenile, and it is possible that its subsequent death may have been the result of gastro-intestinal tract

problems resulting in reduced ability to absorb micro-nutrients. The two surviving clones were reported as healthy at 19 months of age.

Second Chance, the Brahman bull clone described by Hill et al. (2000a), has been outlined in detail in the preceding section. The researchers speculate that the early diabetes had resolved at eight months of age and the calf was clinically normal. At a conference in September of 2002, the bull was reported to be 3 years of age, with normal weight, growth, behavior, and normal semen production. The investigator presenting this information also reported that the bull's glucose level was elevated, although they could not rule out the role of stress resulting from medical procedures as a cause (Westhusin in PIFB 2003⁹⁰). In a subsequent conversation, Dr. Westhusin indicated that the blood glucose has remained within normal limits since the previous report.

Lanza et al. (2000) reported on 24 sexually mature transgenic bovine clones. Physical examinations were reported as normal including temperature, pulse, respiratory rate, general appearance, lymph nodes, and abdominal palpation. Blood and urinalysis indicated that in general, those variables were within normal ranges although six animals had total urine protein levels slightly below the comparator average. Studies with adaptive T-cell responses indicated that these transgenic clones had functional immune systems, and that the animals responded to periodic infection in the same manner as conventional cattle.

Pace et al. (2002) measured weight gain in their transgenic clones until the age of 540 days. Although comparison of the overall cohort with any comparator group is difficult because the clones were raised at different facilities, 52 of the clones raised at the same facility had similar weight gain over the first 120 days of life (approximately 1.15 kg/day). Weight gain of 17 clones from the same genetic line declined to 1.09 ± 0.14 and 0.92 ± 0.10 kg/day at 365 and 540 days, respectively, entirely consistent with weight gain profiles of conventional animals.

Yonai et al. (2005) reported on the growth characteristics of six Holstein and four Jersey clones with shortened telomeres from birth through two years of age. Those data have been summarized in Table VI-6. Evaluation for clones aged 12-24 months indicates that animals had normal weight gain for their breeds, indicating their overall health. With the exception of brief mentions of bloat and mastitis, no other illnesses were reported in this study. All of the animals that entered the study were alive at the time the manuscript was submitted for publication.

Wells et al. (2004) have reported that clones produced at AgResearch have an overall annual mortality of eight percent over four years. Most of the mortality observed appears to be due to the sequellae of LOS or accidents or mishaps; no contemporaneous comparator exists. They also

⁹⁰ <http://pewagbiotech.org/events/0924/presentations/Westhusin.pdf>

note that one clonal family and their half-siblings were all alive and healthy at 18 months of age, implying that there may be an association between the cell line used, susceptibility to LOS and its sequellae.

The immunologic competency of three Holstein bull clones was tested using skin allografts (Thoret et al. 2006). Skin grafts were chosen for this study because skin is the most immunogenic transplantable tissue and is easily obtained with little harm to the animal. The bulls were cloned using fibroblasts from 60-day fetuses as donor cells. At 18 months of age, the bull clones received a skin graft from an unrelated, 12-month old Holstein bull. Each clone also received a skin graft from the other two clones in the cohort. Grafts were placed along the back and left in place for 13 weeks. No immunosuppressive therapy was administered, and grafts were examined both macroscopically and microscopically via punch biopsies. In every case, skin grafts among clones were accepted, but third-party grafts from the unrelated bull were rejected. These results indicate that genetically identical adult bovine clones are immunologically compatible and sufficiently immunocompetent to be able to recognize and reject allografts from genetically unrelated cattle.

Immune function in cow clones was also investigated by Tanaka et al. (2006). Numbers of granulocytes, monocytes, B cells and several T cell subsets were measured in blood obtained from six Holstein clones and five age-matched, sexually-derived Holstein cows during early lactation (58-62 days post-calving) and mid to late lactation (179-300 days post calving). Cows were 2-4 years old at the time of sampling. Proportions of granulocytes, monocytes, B cells and most T cell subsets were similar in cloned and normal cows. At calving, there is normally a decrease in the proportion of $WC1^+\gamma\delta$ T cells. In this study, the percentages of $\gamma\delta$ and $WC1^+\gamma\delta$ T cells were significantly lower during early lactation in clones compared to controls. By mid to late lactation, both T cell subsets recovered and were no longer different from those in normal cows. As $WC1^+\gamma\delta$ T cells produce interferon- γ in response to several bovine pathogens, the authors suggest that decreased populations of these cells may cause cow clones to be more susceptible to infection during early lactation. It is important to note, however, that the milk yield of these clones ($11,731 \pm 1,397$ kg/300 days) was significantly higher than that of the comparators ($9,577 \pm 1,960$ kg/300 days). T cells migrate from the blood into the mammary gland during lactation, and there is evidence that this migration may be selective for certain subsets of T cells. Thus it is possible that the observed decrease in $WC1^+\gamma\delta$ T cells found in the milk of the clones in clones in this study was related to their higher milk production, resulting in a greater influx of these cells from the blood into the mammary gland.

Tecirlioglu et al. (2006) measured 13 biochemical parameters in blood collected from three post-pubertal, Holstein-Friesian bull clones. Blood samples were collected weekly for three weeks, and values were compared to age-matched controls housed under similar conditions at the same

farm. Although serum concentrations of calcium, protein, albumin, aspartate aminotransferase, creatine and testosterone were higher in clones compared to controls, and glutamate dehydrogenase was lower in clones, because all these values were within the normal reference ranges for cattle at the laboratory where the analyses were done, the authors considered these differences to be minor.

As part of a three year study of clones at INRA, Heyman et al. (2007) reported on the health and development of cloned Holstein heifers from four to 36 months of age. Data describing the post-pubertal period will be discussed here; other findings from this study are described in the Juvenile and Reproductive Development sections. The authors reported that repeated clinical evaluations revealed no differences in cardiovascular, respiratory, and locomotive functions, clinical biochemistry, or immune parameters through 36 months of age (data not shown). Concentrations of plasma cortisol and urinary epinephrine and norepinephrine, measured every two months, and were similar between clones (n=5) and controls (n=5) during the postpubertal period. Cortisol concentrations were more variable among the clones, however, than the comparators. These results do not support the hypothesis that clones are more sensitive to stress than conventional animals, and the authors conclude that the clones in their study were not suffering from chronic stress.

Extensive blood chemistry and hematological analyses were conducted in 11 cloned cattle and 11 sexually produced comparators produced at the University of Connecticut (Yang X et al. 2007b). Ages ranged from “>12 months” to 43 months. The six females and fives males in each group represented several breeds: Angus, Brangus, Holstein, and Red and White Holstein, and cross-breeds.

Blood samples were collected at the time of slaughter and analyzed at the Cornell University College of Veterinary Medicine Diagnostic Laboratory. No differences between clones and comparators were observed for the 24 blood chemistry parameters evaluated or the 16 hematological parameters. For “essentially all” parameters examined, standard deviations of the means were similar for clones and comparators, implying that the variability in blood chemistry and hematological measurements within the populations of clones and non-clones in this study were similar.

Cyagra Dataset: 6-18 Month Cohort

The oldest cohort of Cyagra animals spans 6-18 months of age, and actually overlaps the Juvenile and Post-pubertal Maturation developmental nodes. Clearly, the younger clones in this cohort have more in common with the older, but still juvenile, animals of the preceding cohort, while the older clones are more appropriately considered as nearing “adulthood.”

The 6-18 month Cyagra clones were virtually indistinguishable from the comparators. None of the animals had any visible anomalies on physical examination (See Appendix E for details). The laboratory values derived from blood samples drawn from the clones are virtually superimposable on those of the comparators. Only three of the 294 hematological values and six of the 592 clinical chemistry measurements were outside the clinically relevant range. In aggregate, 99 percent of the laboratory measurements were within the clinically relevant range established by the comparators.

Review of Chart E-301 indicates that only two analytes initially appeared to marginally exceed the range characterized by the comparators: estradiol-17 β (E2), and insulin-like growth factor-1 (IGF-I). Neither of these findings was judged to pose clinical significance for the animals or any food consumption risk. Although the E2 levels of five animals exceeded the comparator range, none exceeded the Cornell Reference range, which as previously discussed, is derived from adult cattle. By comparison, 14 of the 20 comparators had measurements that were lower than the Cornell Reference Range (Chart E-302). For a more complete discussion of the normal fluctuation of E2 levels in cattle, see Appendix E. IGF-I levels in the Cyagra cohort were slightly higher in males than in females, and in three of the bull calves (# 24, 33, and 35) were slightly increased (less than 10 percent) relative to the comparator Group. Review of the literature on IGF-I levels in cattle indicated that basal circulating levels of IGF-I vary with a range of factors and fluctuate dramatically among individual animals in herds (Vega et al. 1991). Plasma concentrations of IGF-I are strongly influenced by a number of factors including gender, age, and diet (Plouzek and Trenkle 1991 a,b). The primary nutritional determinants of basal IGF-I levels appear to be crude protein and the number of calories absorbed by the animal (Elsasser et al. 1989). Given that most non-transgenic clones are derived from animals of superior genetic merits for traits such as growth and development, 10 percent elevations in IGF-I levels are likely of no clinical significance for the animal, and pose no food consumption risk.

No remarkable dissimilarities were noted in the blood variables of clones and comparators. There were no indications of problems with respect to red or white blood cell measurements. One animal (Clone #98) exhibited higher basophil counts than the comparator range, but there appeared to be no clinical correlate to that value, and as a result it was judged insignificant to the health of the animal or food safety.

(b) Unpublished data

Hematology data for two Holstein heifer clones aged 14 months old were submitted to CVM by a private veterinary firm. They consisted of a Veterinary Certificate of Inspection, results of serological testing showing the animals were free of Bovine leucosis virus and Bovine viral diarrhea, and standard clinical chemistry and hematology panels. All hematology and clinical

chemistry results were within the range of the laboratory's reference values except red cell distribution width, which was slightly below the reference range used by the testing laboratory (see Chapter V). As discussed in Chapter V and Appendix E, RDW is a secondary indicator, and does not on its own suggest a health problem. Certificates of Veterinary Inspection accompanying the hematology data indicate that both heifers were healthy.

**(c) Summary Statement for Post-Pubertal Maturation in Bovine Clones
(Developmental Node 5)**

Clones in this age group exhibited no remarkable differences from non-clones with respect to their overall health. The Cyagra clones were indistinguishable from the comparator group on the basis of clinical and laboratory tests. The study of Yonai et al. 2005 indicates that clones continued to grow well for the duration of the study (two years). No residual health problems were noted in any of the clones in this Developmental Node that had not been identified in earlier developmental nodes. Some clones died prematurely for different reasons, including the sequellae of earlier disease. Individual animal reviews indicated no health problems, or changes in physiological parameters that would indicate a food consumption risk that would not be detected in existing food safety regulations (e.g., mastitis in milking cows).

vi. Progeny of Bovine Clones

From a food safety perspective, information on the progeny of clones is probably more important than data on the clones themselves because it will be primarily the progeny, not the clones, that enter the food supply.

Starbuck II Progeny

Ortegon et al. (2007) studied physiological parameters and telomere length in a group of seven Holstein heifers sired by a bull clone, Starbuck II. Dams were conventionally bred normally cycling Holsteins. Starbuck II progeny were monitored monthly for 12 months, from weaning to puberty, and data were compared to those from a group of breed- and age-matched comparators. At 14-15 months of age, five of female progeny heifers were inseminated with semen from a conventionally bred bull.

The Starbuck II progeny heifers appeared phenotypically normal and were healthy throughout the observation period. There were no differences between the clone progeny heifers and comparators in growth parameters (body weights, height, length) or several reproductive endpoints (range in age of puberty, serum progesterone concentrations, ovarian follicular dynamics, estrous behavior). Starbuck II heifers also responded normally to drugs commonly used in cattle for estrous synchronization (prostaglandin F_{2α}) and superovulation (follicle stimulating hormone). Clinical chemistry values in Starbuck II progeny were similar to values in

controls and within normal ranges. Relative to their comparators, the clone progeny were described as “less excitable” during handling in the chute. This difference in behavior was reflected in the physiological measurements showing that the Starbuck II progeny heifers exhibited lower heart rate (81.4 ± 17.2 beats per min (bpm) vs. 99.4 ± 17.8 bpm), lower respiratory rate (35.2 ± 10.2 respirations per min (rpm) vs. 45.8 ± 14.0 rpm), and lower body temperature ($38.9 \pm 0.4^\circ\text{C}$ vs. $39.2 \pm 0.3^\circ\text{C}$) compared to controls. However, all of these values fell within the range of normal values for dairy cattle. The authors speculated that the lower heart rates, respiratory rate and temperature in Starbuck II progeny were due to a faster adaptation to handling and manipulation. It is also possible that these results reflect more frequent human contact (observation and handling) of the Starbuck II progeny relative to their comparators. Pregnancy was confirmed in two of the five Starbuck II progeny that were inseminated, but these pregnancies were terminated at 45 days due to housing constraints.

Telomere lengths, measured in skin biopsies taken at 30 days of age, were similar in Starbuck II progeny ($n = 32$) and age-matched comparators ($n = 20$). Telomere length in Starbuck II was also similar to age-matched control bulls, but the lengths of Starbuck II’s telomeres and his progeny’s telomeres were not compared. Chromosomal analysis in peripheral blood leucocytes indicated similar frequencies of diploid karyotypes in progeny and comparators. Therefore, it appears that telomere length homeostasis and chromosome stability were normal in the Starbuck II clones.

None of the abnormalities previously reported in cattle clones, such as cardiac and respiratory abnormalities, hyperthermia during the first two months of life, or altered hematological and endocrine parameters (Chavatte-Palmer et al. 2002), were observed in the progeny of Starbuck II. Although no laboratory assays of immune function were performed, the authors concluded that the absence of illness and need for therapeutic treatments provides evidence of functional immune systems in Starbuck II progeny. The authors concluded that the clone progeny had normal phenotypic characteristics, normal behavior, and their growth, health, hematological and reproductive parameters were comparable to age-matched comparators.

Progeny of Clone x Clone Mating

In some cases, female clones may be mated to male clones to produce offspring for human consumption. This raises the question of whether progeny derived from mating pairs of clones will be different from those derived from conventional breeding, from matings in which only one of the parents is a clone. To begin to address this question, growth performance was described for a calf produced by a mating in which the dam and sire were both clones (Kasai et al. 2007). Donor cells were cultured cumulus and ear cells, respectively, obtained from Japanese Black cattle. Comparators in this study were “full siblings” ($n=7$; heifer calves) produced by artificially inseminating the cow used as the nuclear donor with semen from the bull used as the nuclear donor. Comparator embryos were flushed from the nuclear donor and transferred to recipients.

To produce the calf of the clones, the female cow clone was mated to the male bull clone by artificial insemination.

Following a 292-day gestation (11 days longer than the average for comparator pregnancies), the dam gave birth to a heifer calf. No assistance was needed at the time of parturition. Birth weight, clinical examination, hematology, serum biochemistry and telomere length of the calf of the clones were within the ranges measured for the comparator calves. Growth of the calf from birth to 12 months, as measured by body weight and shoulder height, was similar to that of the comparators. No serious health problems were observed in the calf and at the time of publication, the heifer was four months pregnant following artificial insemination at 18 months of age. To the authors' knowledge, this was the first report demonstrating normal growth performance in a calf derived from mating two clones. These results are consistent with those of Heyman et al. (2004), who did not observe LOS in offspring produced by mating either pairs of clones or clones to non-clones.

vii. Conclusions Regarding Food Consumption Risks from Bovine Clones and their Progeny

As the first prong of our strategy to address the food consumption risks associated with clones, we have used the Critical Biological Systems Approach (CBSA) as a framework to search for subtle differences between clones and comparators that may pose food consumption risks. In general, these differences cannot be detected macroscopically but may be evident as differences in physiological parameters during the five developmental nodes. For bovine clones, our health-based assessment of food consumption risks is facilitated by a significant body of evidence from the peer-reviewed literature together with a large data set from Cyagra. Many bovine clones do not survive the neonatal period, and several abnormalities (e.g., those related to LOS, prolonged recumbency, umbilical malformations) have been described during this developmental node. None of these abnormalities is unique to clones, and all have been observed in calves produced by other ARTS such as *in vitro* fertilization or following natural mating.

In clones that survive the neonatal period, some studies have identified differences in physiological measures between clones and comparators during the first few weeks of life. These findings support the notion that bovine clones are more physiologically unstable during the early juvenile period. There is evidence that the physiological transition from neonatal period to the juvenile period may take longer in calf clones (e.g., elevated body temperature during the first two months of life). Once their physiological status is stabilized, however, there is ample evidence to indicate that growth and development proceed normally in bovine clones. Similarly, several studies indicate that fertility in clones is normal, and there are no indications that the physiology or health of clones is compromised during the post-pubertal period.

In summary, we have searched for subtle differences between clones and their comparators to identify differences that may pose food consumption hazards. We have not found any such subtle differences, and based on this review of the health and physiology of bovine clones using the CBSA approach, we conclude that there is no reason to expect that food from bovine clones would pose additional food safety risks compared with the same products derived from conventionally-bred cattle.

Clone progeny are not expected to pose any increased food consumption risks compared with other sexually reproduced animals (NAS 2002b). Although the amount of data describing the health of progeny of clones is more limited than the amount describing the health of clones themselves, the results are consistent with the biological assumption. In the two studies that characterized the physiology of heifers produced by clones, growth, reproductive function, and telomere length were normal in clone progeny, and the incidence of general health problems was not increased in clone progeny compared with progeny of other sexually reproduced animals. Based on the CBSA approach, we therefore conclude that sexually reproduced progeny of clones are indistinguishable from other sexually reproduced animals, and pose no additional food consumption risks.

b. Swine Clones

There are approximately 45 papers, including some reviews, within the peer-reviewed literature that address cloning of swine; many of these report on the production of transgenic swine by SCNT. Unlike cattle, where improvement of breeding stock has been a major driving force for advances in reproductive technologies, many of the earlier studies of SCNT in swine have focused on transgenic animals for use as xenotransplant organ sources (reviewed by Prather et al. 1999; Westhusin and Piedrahita 2000; Wheeler and Walters 2001; Carter et al. 2002; Machaty et al. 2002; and Prather et al. 2003). Nonetheless, cloning swine for agricultural purposes has become the focus of at least one large commercial venture (ViaGen, Inc.), and others (Archer et al. 2003a,b) have also reported extensively on the health and physiological status of non-transgenic swine clones.

The cloning of swine was first described in 2000 by Polejaeva and her colleagues at what was then PPL Therapeutics in Blacksburg, Virginia and Roslin, UK. Several laboratories followed that publication with their own reports of swine cloning using different approaches to cell fusion, oocyte maturation, or other technical issues (Betthausen et al. 2000; Onishi et al. 2000; and Bondioli et al. 2001). In the subsequent years, additional studies have reported on the difficulties of overcoming the early stage failures (Boquest et al. 2002, and Yin et al. 2002a; Lee GS et al. 2005a; Zhu et al. 2004).

Another issue contributing to the difficulty of cloning swine is that unlike cattle, sheep, and goats, swine require a minimum number of viable embryos, thought to be approximately four, to initiate and sustain pregnancy (Polge et al. 1966; Dzuik 1985). This has posed a technical limitation for the development of cloning in this species because the high loss of embryo clones throughout the pregnancy necessitates the transfer of a very large number of clone embryos into the surrogate dam (between 150 and 500) to ensure that the minimum number of embryos is maintained. A recent paper by King et al. (2002) explored hormonal treatments to sustain limited numbers of viable embryos to term, and demonstrated that pregnancies can be established with a mixture of fertilized and parthenote embryos and that small numbers of fertilized embryos can develop to term successfully with hormonal support.

Because of these difficulties, most of the available reports describe only the implantation and early perinatal phase. Two publications by Archer et al. (2003a,b) describe the behavior and clinical chemistry of juvenile swine clones.

i. Cell Fusion, Nuclear Reprogramming, Embryonic and Fetal Development Through the Perinatal Developmental Period in Swine Clones (Developmental Nodes 1 and 2)

(a) Peer-reviewed Publications

In the first published report of swine clones by Polejaeva et al. (2000), two rounds of nuclear transfer were employed, with *in vivo* matured oocytes as recipients and cultured granulosa cells as donors, to produce five live female piglet clones. Piglets were delivered by C-section on day 116 of the pregnancy. The only data on the health of these piglets indicated that the average birth weight of the clones of 2.72 pounds (range 2.28-3.08 pounds) was approximately 25 percent lower than in piglets produced using natural mating in the same population as the donor cells (average birth weight of 3.6 pounds, range 3.3-3.9 pounds in an average litter size of 10.9 piglets).

In the second report of swine cloning, after several unsuccessful attempts, Onishi et al. (2000) produced a single female piglet named “Xena” from cultured embryo fibroblast cells. The clone’s birth and placental weights were 1.2 kg and 0.3 kg, respectively, which the authors state were in the normal range for conventional offspring of that breed (Meishan). Xena was described as a “healthy female” but, with the exception of a photograph, no data were provided to confirm that observation.

Bethausen et al. (2000) also describe multiple attempts at establishing successful pregnancies in surrogate dams receiving swine embryos resulting from SCNT. Of the seven pregnancies that

were established, three were with non-transgenic embryo clones. Four live births resulted from two pregnancies, out of 427 embryos implanted into surrogate dams. The first litter yielded two male piglets born alive by vaginal delivery, weighing 2.0 and 3.0 pounds each. The second litter also produced two live vaginally delivered male clone piglets and one mummified fetus. The live piglets in this litter weighed 2.2 and 3.5 pounds. The third pregnancy was aborted at 40 days of gestation. No further information was provided on the health status of the clones at birth. Subsequently, the senior author on this report wrote a Letter to the Editor of the publication (Bishop 2000) to inform that the piglets from the second litter had died one week after their birth due to the aggressive behavior of the first-time surrogate mother. This behavior limited the amount of time the piglets were able to nurse, and the consequent lack of adequate nutrition proved to be fatal to the piglets (Bishop 2000). CVM is unaware of any publications providing additional information on the health status of the first litter.

An Australian group (Boquest et al. 2002) described the birth of live piglets from cultured fetal fibroblast cells that were frozen for two years, employing a novel cell fusion method in which donor nuclei were exposed to inactivated oöplasm for a period of time prior to chemical activation (to begin the process of cell replication). They believe that the lag time between fusion and activation allows for the more efficient reprogramming of the donor cell nuclei. The investigators transferred between 40 and 107 embryos to 10 surrogate dams, resulting in five pregnancies. Three of those pregnancies were aborted, and each of the two remaining pregnancies yielded one live piglet. No information is provided about the health status of the clones.

Yin et al. (2002a) also developed a novel method for the production of pig clones by treating oöcytes to be used as recipients with demecolcine such that the condensed chromosomes produce a protrusion at the cell membrane that can easily be removed by micro-aspiration. Donor cells were obtained from an adult female four year old Landrace pig, and included cultured heart and kidney cells. Six surrogate dams were implanted with between 137 and 341 embryos. Three of the recipients never became pregnant, and one aborted the pregnancy on day 62. The remaining two pregnancies, both with embryos of heart tissue origin, resulted in live births. The first litter included four live female clones, and one dead fetus. The second resulted in another four live female clones, and two dead fetuses. None of the clones, live or dead, exhibited any morphological anomalies. The authors reported that the eight surviving clones were eight months old at the time of publication, and “appear quite healthy.” No further information is provided.

Lee GS et al. (2005a) found that supplementing culture media with epidermal growth factor (EGF) improved cleavage rate of NT embryos, but not the rate of blastocyst formation compared to unsupplemented media, although total cell numbers in surviving blastocysts were higher in EGF supplemented media. Adding EGF after morula formation did not affect blastocyst

formation rate or cell numbers. Zhu et al. (2004) found embryos produced with stem cells isolated from fetal porcine skin cultures had higher preimplantation development rates than embryos produced using fetal fibroblast cells. Karyotypic analysis of the two donor cell cultures indicated that porcine stem cells accumulated fewer abnormalities and were more stable through multiple passages compared to fibroblast cells. Porcine stem cells also yielded more blastocysts than fibroblast cells. Because neither of these groups attempted to transfer embryos to recipients, there is no way to know whether these improvements in early embryo developmental efficiency would have resulted in a higher proportion of live clones.

Bondioli et al. (2001) reported on the generation of transgenic pig clones from cultured skin fibroblasts derived from an α -1,2-fucosyltransferase (H-transferase) transgenic boar. (H-transferase is involved in producing the sugars on the surface of a pig cell that are partially responsible for the acute phase of rejection observed when non-human tissues are transplanted into humans.) Of the 217 embryos transferred into five surrogate dams, two pregnancies resulted. One of the surrogate dams was euthanized at 90 days of gestation for health reasons that the authors state were unrelated to embryo transfer. One mummified fetus and one apparently viable fetus were recovered. The other pregnancy yielded two live piglets that were delivered by C-section at 116 days of gestation. The piglets were reported as “healthy,” and a photograph of two apparently normal piglets at two months of age is provided in the paper.

Walker et al. (2002) have reported on the largest litters of piglets produced by SCNT. Donor cells were derived from Duroc fetal fibroblasts, and fused with *in vitro* matured oocytes. A total of 511 embryos were transferred into five surrogate dams, with between 59 and 128 embryos per recipient. All five recipients were confirmed pregnant by ultrasound between days 28 and 40 post-implantation. Four of the five pregnancies went to term, and litters containing between 5 and 9 piglet clones (total of 28) were delivered. Three of the four surrogate dams were induced and delivered on gestational day 115. The fourth was allowed to deliver naturally, and produced her litter on gestational day 117. One of the 28 clones was stillborn, but no abnormalities were noted on necropsy. One of the live born clones presented with anal atresia (no anus or tail), and was the smallest of all of the clones (birth weight of 0.72 kg, and crown rump length of 23.5 cm). The authors noted that anal atresia is a developmental abnormality seen at a natural low frequency in conventional piglets. The question of whether this is a random event due to genetic or inappropriate reprogramming cannot be answered from this dataset.

Table VI-9: Summary of Birth Characteristics of Piglet Clones

(source: Walker et al. 2002)

Litter size	Mean Birth Weight (kg) ¹	Mean Placental Weight (kg) ¹	Crown-Rump Length (cm) ¹
9	1.15 ± 0.17	0.29 ± 0.09	68.8 ± 2.1

5	1.06 ± 0.23	0.23 ± 0.02	71.6 ± 7.6
7	1.35 ± 0.13	0.29 ± 0.07	74.9 ± 1.8
7	1.29 ± 0.26	NR	NR
Control ²	1.37 ± 0.12	NR	NR

¹ All values presented as means ± SD.
² The control birth weight was derived from the average weight ± SD from 10 litters of piglets from naturally bred Duroc pigs.
NR = Not reported.

The remaining piglets had birth weights that appear to be a little lower than conventional piglets of the same breed. The authors noted with explicit surprise that there was little correlation between litter size, placental weights, and fetal weights (Table VI-9). They predicted a correlation of 0.639 between placental and fetal weight, but noted that the lowest mean birth weights occurred in the litters with the smallest number of piglets. The authors asserted that without the appropriate controls for litter size, *in vitro* oöcyte maturation and other manipulations, it is inappropriate to assign the SCNT process as the cause of the difference in birth weights. Two of these litters subsequently served as the source of the clinical and behavioral studies of Archer et al. (2003a,b).

Longer gestational length was reported for dams carrying two groups of clone pregnancies produced by Carroll et al. (2005). These groups of piglets were derived using two different fibroblast cell lines developed from two porcine fetuses (day 35). Gestational length in clone pregnancies (n = 5 litters total) was 118.8 ± 0.97 days compared to 114 ± 0.41 days in natural pregnancies in the same herd from which the cells for the clones were obtained. The first group of clones (C1) consisted of just 2 piglets from a single litter (no information is provided about the littermates of these piglets), while the second group of clones (C2) consisted of 7 piglets from 4 litters. Birth weights are not reported in this study, and the authors did not report any abnormalities in the clone piglets. The acute phase immune response of these clones was subsequently evaluated at 27-30 days of age (for a discussion see the Juvenile Developmental Node).

Jiang et al. (2007) described body weights, organ weights, and gene expression in nine piglet clones that died at birth or shortly thereafter. Five of these were transgenic with one disrupted allele of the porcine α 1,3-galactosyltransferase gene, and the other four were not transgenic. Controls (n=5) were age-matched piglets generated via sexual reproduction. The number of litters from which piglets were derived is not specified. The results for controls were compared with results from all nine piglet clones (transgenic and non-transgenic). Three of the dead piglets “had minor phenotypic abnormalities, such as curled toes”. The authors note that this anomaly has been observed occasionally in piglet clones, and that piglets with similar minor abnormalities

can survive and develop normally. Consistent with other studies (Polejaeva et al. 2000; Lai et al. 2002; Yin et al. 2002a), the clones weighed less than controls at birth (1.2 kg. vs. 2.1 kg, respectively). Weights of heart, liver, kidney and spleen were similar between newborn clones and control piglets. Mean lung weight, expressed as a percentage of body weight, however, was significantly lower in the deceased clones (1.6 percent vs. 2.1 percent in controls).

To investigate whether lower body weight in piglet clones is correlated with expression of genes that regulate growth, Jiang et al. (2006) measured the expression of four imprinted genes in their cloned piglets: IGF-2, PEG3, IGF-2R, and GRB10. These genes either promote (IGF-2, PEG3) or inhibit (IGF-2R, GRB10) growth. Using quantitative real-time reverse transcription polymerase chain reaction (RT-PCR), the authors observed lower levels of expression of the IGF-2R gene in lung, brain and spleen tissue from the deceased newborn clones compared to controls, but these levels of gene expression were not related to body weight.

Shibata et al. (2006) used SCNT to produce eight female pigs of the Jin Hua breed. Compared with non-clone Jin Hua control piglets, the clones had similar mean birth weights (0.87 kg in controls vs. 0.91 kg in clones; no estimate of variance was provided) and were phenotypically normal. Reproductive and growth performance was subsequently characterized in these pigs, as described in the Juvenile and Reproductive Development nodes.

To determine whether the cellular age of donor cells is altered by nuclear reprogramming during SCNT, Jeon et al. (2005) analyzed telomere length and telomerase activity in 12 newborn piglet clones. Donor cells for SCNT were fibroblasts isolated from day 30 fetuses. Clones had longer telomeres compared to donor fetal fibroblasts. Although telomerase activity in these clones was similar to that in the donor cells, telomerase activity was higher in SCNT blastocysts. The authors suggest that in clones derived from porcine fetal fibroblasts, telomeres can be rebuilt/elongated at the blastocyst stage of development.

(b) Unpublished data

ViaGen Inc. provided birth weights of seven male swine clones as part of the data package presented to CVM. Clones were smaller at birth than AI comparators of similar genetic background (See Appendix F: ViaGen Dataset). No detailed health data were available on these clones for this developmental node. All clones survived the neonatal period.

Additional data submitted to CVM included birth weight, average daily weight gain (ADG), body temperature, and pulse rates on another cohort of neonatal swine clones (see Chapter V). Birth weights for three clones ranged from 1.1 to 1.4 kg, and ADG ranged from 0.46 to 0.55 kg; however, because the breed of swine was not identified, it is not possible to determine whether these data are within normal ranges. The report indicated that two of the five piglets, both from the same litter and weighing 1.0 kg at birth, died within the first 48 hours. The cause of death was not reported, and no other details were provided. Body temperatures of the piglets were low (range 98.8 to 101.8°F) during the first 48 hours compared to reference body temperature for adult swine (102-103°F). This finding is not unusual, however, as neonatal swine generally have difficulty regulating body temperature, and require supplemental heat after birth (see Chapter V).

(c) Summary Statement on the Embryo/Fetal to Perinatal Developmental in Swine Clones (Developmental Nodes 1 and 2)

The production of swine clones differs from the other livestock species discussed in this risk assessment because of the requirement for a minimum number of viable fetuses to maintain the pregnancy. The gestational losses observed are a function of the combined low “success rate” for embryonic and fetal development for the individual clone and the requirement for a minimum number of growing fetuses to implant. Clone piglets do not appear to exhibit the overgrowth phenomena observed in cattle, and if anything, newborn swine clones may be smaller than their non-clone counterparts. Most swine clones at this developmental stage appear to be healthy; there is only one report of a fairly commonly observed congenital malformation (anal atresia), and one that is less frequently observed (curled toes).

ii. Juvenile Development and Function in Swine Clones (Developmental Node 3)**(a) Peer-reviewed Publications**

Archer et al. (2003a,b) have investigated the degree of behavioral and physiological variability exhibited among litters of swine clones and their closely related conventional siblings. The derivation of these clones has been described in Walker et al. (2002). The clone cohorts consisted of two litters of 5 and 4 female swine derived from the same cell line born 6 weeks apart. The control groups consisted of a litter of four female full siblings (both parents in common) and a litter of four female half-siblings taken from three sows mated to the same boar. All animals were farrowed (born) in conventional farrowing crates, and weaned at 5-6 weeks of

age when they were placed in adjacent identical pens and given continuous access to identical standard rations and water. Results in these studies were presented as means and ranges; individual animal data were not provided.

One study (Archer et al. 2003b) evaluated behavioral characteristics including food preference (for apples, bananas, saltine crackers, and carrots), temperament (as judged by time to remove a towel placed on the pig's head, attempts to escape mild restraints, being placed on their backs, and being lifted off the ground), and time budgets (the amount of time spent engaged in a particular activity in their pens). The results of this study indicated that the behaviors of swine clones were no more homogenous than the behaviors of siblings and may be more variable than the comparator animals, although the statistical power to draw such a conclusion was limited. The authors conclude that “...using nuclear transfer to replicate animals to reproduce certain behavioral characteristics is an unrealistic expectation.” The relevance of the study to an evaluation of the health of swine clones, however, is that the animals behaved in much the same manner as conventional animals, and displayed no behavioral anomalies at the times tested (15-16 weeks of age for the food trials, 8-9 weeks and 14-15 weeks for the towel test, 7 weeks for the restraint tests, and 13-15 weeks for the time budget tests).

Another study performed by this group (Archer et al. 2003a) evaluated whether the SCNT process introduced epigenetic changes into animal clones that could be manifested at the genomic (*e.g.*, methylation status) (See Chapter IV), physiological (*e.g.*, blood chemistry), and anatomical (*e.g.*, weight, size, coat) levels. Body weights of all the animals overlapped and were within the normal range for the age and breed, with the exception of a single clone that was small at birth, and never attained the size of its littermates. This is likely a case of “runting,” which is observed in conventional animals as well. Teat number was the same for all animals (6, 6 distribution) except for one clone (6, 7 distribution), within the normal variability in conventional pigs.

One of the clones also exhibited an unusual hair growth pattern (*e.g.*, longer and sparser), which the authors state prompted an examination of the histology of the skin. Results of that investigation indicated that with one exception, skin morphology showed no unusual variations among the pigs. The exception was a clone that exhibited morphology indicative of hyperkeratosis.⁹¹ Hyperkeratosis, also referred to as parakeratosis, also occurs in naturally bred and AI pigs between the ages of 6 and 16 weeks, and is generally associated with zinc and essential fatty acid deficiency or excess dietary calcium or phytates. Gastrointestinal disorders may also affect zinc absorption, and contribute to the development of this condition (Cameron

⁹¹ Hyperkeratosis, generally referred to as parakeratosis in swine, is characterized by lesions of the superficial layers of the epidermis. These lesions rapidly become covered with scales, and then develop hard, dry crusts with deep fissures.

1999). Other possible causes of hyperkeratosis include heredity, and other non-specific causes of skin inflammation (Blood and Radostits 1989). Dermatitis vegetans is the inherited form of this disease in swine, and is a semi-lethal recessive gene (Blood and Radostits 1989). Although the phenotypic variation is interesting, it is of limited concern for food safety, as pork skin that exhibits severe hyperkeratosis would be condemned at slaughter.

Measurement	Merck ¹	Clones		Controls	
		Week 15	Week 27	Week 15	Week 27
Creatinine (mg/dl)	0.8-2.3	1.02 ± 0.22 ³ (0.7-1.4)	1.11 ± 0.14 (0.9-1.3)	1.58±0.95 (0.8-3.6)	1.25 ± 0.32 (0.9-1.8)
Alkaline Phosphatase (U/l)	41.0-176.1	208.67 ±11.60 (192-226)	100.78 ±17.89 (80-128)	235.25 ±33.12 (201-294)	117.88 ± 49.54 (56-196)
BUN ² (mg/dl)	8.2-24.6	9.69 ± 1.45 (7.7-11.9)	10.09 1.29 (8.9-11.7)	9.58 ± 2.84 (6.3-11.9)	7.85 ±2.04 (5.8-11.7)
ALT (SGPT) (U/l)	21.7-46.5	46.78 ± 4.24 (46-56)	38.44 ± 2.55 (34-42)	53.25 ± 9.16 (41-70)	38.88 ± 8.32 (22-48)
Albumin (g/dl)	2.3-4.0	4.21 ± 0.13 (4.0-4.3)	4.12 ± 0.26 (3.6-4.3)	4.40 ± 0.21 (4.1-4.7)	4.15 ± 0.55 (3.0 -4.7)
Phosphorus (mg/dl)	5.5-9.3	10.29 ± 0.42 (9.6-10.6)	7.87 ± 0.60 (7.0-8.8)	10.75 ± 0.82 (9.5-11.8)	7.75 ± 0.85 (6.1-8.9)
Calcium (mg/dl)	9.3-11.5	11.30 ± 0.24 (11.0-11.7)	11.50 ± 0.84 (10.7-12.5)	11.49 ± 0.57 (10.4 -12.2)	11.35 ± 1.12 (9.5-12.7)
Serum Protein (mg/dl)	58.3-83.2	6.34 ± 0.35 (5.7-6.8)	6.96 ± 0.44 (6.2 - 7.7)	6.09 ± 0.32 (5.9-6.5)	7.00 ± 0.60 (6.4-8.2)
Glucose (mg/dl)	66.4-116.1	100.56 ± 10.03 (101-113)	86.89 ± 7.03 (70-94)	115.88 ± 14.89 (105-151)	99.13 ± 7.40 (87-107)
Globulins (g/dl)	3.9-6.0	2.13 ± 0.33 (1.6 - 2.8)	2.83 ± 0.57 (2.2 -3.6)	1.69 ± 0.22 (1.3 - 2.0)	2.85 ± 0.89 (1.9-4.0)
A/G ratio	na	2.01 ± 0.30 (1.43-2.56)	1.52 ± 0.39 (1.03-2.10)	2.65 ± 0.36 (2.15-3.23)	1.62 ± 0.62 (0.75-2.47)
Total T3 (ng/dl)	na	70.95 ± 10.05 (60.09 -92.99)	48.60 ± 9.37 (36.71-54.63)	95.48 ± 17.85 (74.12- 120.07)	43.99 ± 19.41 (15.00-66.87)
Cortisol (g/dl)	na	5.56 ± 2.52 (1.2-8.9)	4.58 ± 1.76 (3.2-8.9)	6.56 ± 2.39 (3.1-10.9)	4.66 ± 3.55 (0.9-10.0)

¹ Merck Veterinary Manual, <http://www.merckvetmanual.com/mvm/index.jsp>, References, Table 07.
² For abbreviations, see Appendix F: The Comprehensive Veterinary Examination and Its Interpretation
³ Values presented are means ± SD, range in parenthesis.

Blood samples were taken from the animals for analysis at 15 and 27 weeks of age (Table VI-10) (Archer et al. 2003a). Although the hypothesis being tested in this study addressed the degree of variability among clones relative to the degree of variability among controls, these data are very instructive in that they provide the most extensive analysis of the physiological status of swine clones at two different times in development. (See previous discussion of Cyagra dataset). Unlike the Cyagra dataset, however, very few animals were evaluated (nine clones and eight controls). Nonetheless, the data are compelling in that they demonstrate that the physiological parameters investigated do not indicate any material differences between the clones and controls. In addition, they provide confidence that these clones are responding appropriately to age-specific signals. Just as the Cyagra cattle clones, the piglet clones initially exhibit relatively high alkaline phosphatase levels: at week 15 both clones and controls have mean levels of 209 and 235 U/L, respectively, while 8 weeks later (at 27 weeks of age), the mean alkaline phosphatase levels have decreased to 101 and 118 U/L, respectively. (Alkaline phosphatase provides a measure of bone growth in young animals.) Phosphorus levels, also an indicator of bone growth, show similar age-related changes, as does T3.

Genomic methylation levels (also discussed in Chapter IV) were evaluated in two repeated sequences, one found at the centromere⁹² and the other in the euchromatin⁹³ regions of the chromosomes (Archer et al. 2003a). The investigators discovered that one euchromatin region of clones had a different degree of methylation from controls. They further observed that another region had an increase in the variability of the degree of methylation in clones relative to controls. The investigators stated that it was not possible “to prove cause and effect” between alteration in methylation patterns and any of the measurements that they had taken on these animals. Additionally, because all of the animals in this study (clones and controls) appear to be healthy, with the exception of the pig with hyperkeratosis/parakeratosis, the developmental relevance of these methylation changes are not clear. It may be that animals that do not survive have higher degrees of variability or derangement of methylation patterns, and that what is being observed in this study is a set of animals that has adapted to or compensated for differences in methylation, or the inherent tolerance of biological systems for changes in methylation status of genes.

The authors concluded that “*while cloning creates animals within the normal phenotypic range, it does affect some traits by increasing variability associated with that phenotype.*” Their final conclusion with respect to phenotypic variability among clones was that they were not necessarily less variable than their closely related, sexually reproduced half-siblings. For pigs, at least, this implies that although genetics may have a strong influence, various environmental

⁹² The constricted region of a mitotic chromosome that holds sister chromatids together—the crossing point in the “X” often used to depict chromosomes.

⁹³ The region of an interphase chromosome that likely to be transcriptionally active.

influences, including intra-uterine environments, may play a significant role in eventual phenotype of the animal.

Carroll et al. (2005) described the acute phase immune response at 27-30 days of age in seven piglet clones derived from four different litters). This response occurs in the early phases of infection, and serves as a defense mechanism that enables the body to control infection while the adaptive immune response is being developed (Janeway et al. 1999).⁹⁴ Control piglets were obtained from natural pregnancies in the same herd. To trigger the acute phase immune response, piglets were challenged i.v. with 25 µg/kg of bacterial lipopolysaccharide (LPS), and blood samples were collected every 30 min for analysis of cortisol and two cytokines, tumor necrosis factor-alpha (TNF-α) and interleukin-6 (IL-6).

Parameter	Control	Group C1	Group C2
Pre-treatment cortisol (ng/ml)	27.6 ± 3.0	4.6 ± 0.6*	18.8 ± 2.3*
Peak cortisol (ng/ml)	268.9 ± 1.7	263.1 ± 24.5	148.1 ± 12.4
Time to peak cortisol (h)	2.5	2.5	1.5
Pre-treatment TNF-α (ng/ml)	0.16 ± 0.02	0.04 ± 0.01*	0.06 ± 0.01*
Peak TNF-α (ng/ml)	202.8 ± 28.0	76.9 ± 34.3*	4.5 ± 1.0*
Time to peak TNF-α (h)	1	1	1
Peak IL-6 (ng/ml) ²	10.7 ± 2.2	2.7 ± 4.5	0.04 ± 0.01
Time to peak IL-6 (h)	2.5	1	1

¹ Values are presented as mean ± SE
² Pre-treatment concentrations of IL-6 were non-detectable.
* Indicates values that are different from control values (p < 0.05)

Body weights were similar among groups at the time of LPS treatment. Compared to controls, pre-treatment serum concentrations of cortisol were significantly lower in clones than in comparators, consistent with the observation of Chavatte-Palmer et al. 2002 in very young cattle clones. Basal serum concentrations of TNF-α, another component of the response, were also lower in clones. Basal secretion of IL-6 was not detectable in this study.

Following LPS treatment, cortisol secretion increased in all three groups. The increase in serum cortisol in clones was lower in magnitude and shorter in duration compared to controls. Peak cortisol concentrations in clones were observed at 1.5 h whereas peak cortisol concentrations in controls were detected at 2.5 h. Peak serum concentrations of TNF-α were observed in all groups

⁹⁴ Immediately upon exposure to bacteria, immune cells in the blood release the proinflammatory cytokines tumor necrosis factor α (TNFα) and interleukins-1 and 6 (IL-1 and IL-6). These cytokines stimulate the liver to produce acute phase proteins which act like antibodies by binding to bacteria, leading to their destruction.

1 h following treatment with LPS. However, the peak concentrations of TNF- α in both groups of piglet clones were higher than peak concentrations in controls. Serum concentrations of IL-6 were increased in controls by 30 min after LPS treatment and peaked at 2.5 h. In C1 and C2 clones, increased concentrations of IL-6 were not detected until 1 h after LPS treatment. Compared to controls, maximal concentrations of IL-6 in C1 and C2 clones were observed earlier (2 h and 1.5 h) and were much lower in magnitude.

Based on these patterns of LPS-induced cortisol and cytokine secretion, the authors conclude that the acute phase immune response was altered in these clones, although the mechanisms, beyond a suggestion of altered epigenetic status, were not specified. The authors hypothesize that donor cells may play an important role in this response, as the patterns of response were clearly different between the two groups of clones (which were derived from different cell lines). This observation could also be influenced by the very small number of genetically identical clones in the C1 group (n=2).

Jiang et al. (2006) described body weights, organ weights, and gene expression in seven genetically identical piglet clones sacrificed at one month of age. This study also included piglet clones that died at or shortly after birth; observations from these piglets are described in the Perinatal section. In their description of the one-month old clones, the authors cite the study on the acute phase immune response in 27- to 30-day old swine clones by Carroll et al. (2005). Thus, it appears that these two studies used the same cohort of piglets. Controls were five age-matched, non-clone piglets.

Body weights were lower in clones compared to controls (6.1 kg. vs. 8.0 kg). Relative to body weight, weights of liver (3.1 percent), kidney (0.7 percent) and spleen (0.6 percent) in piglet clones at one month were significantly greater than controls (2.7 percent, 0.6 percent, and 0.4 percent, respectively). Although the authors suggest that altered organ weights may be indicative of developmental abnormalities in these organs, no histological, biochemical, or physiological measurements are provided to support this speculation. Further, as all of these animals were sacrificed at the one month time point, it is not known whether these organ weight differences would have resolved or continued to remain different as the animals matured. To investigate whether lower body weight in the piglet clones was correlated with expression of genes that regulate growth, the authors measured levels of expression of genes that either promote (IGF-2, PEG3) or inhibit (IGF-2R, GRB10) growth. Relative to controls, clones had higher levels of expression of IGFR in lung, liver and kidney and higher levels of expression of GRB10 in the liver. However, the biological significance of these differences is uncertain as there were no significant correlations between gene expression and either body weight or organ weights. Interestingly, variances in levels of gene expression in the one month-old, genetically identical

clones were larger than those in controls, providing further evidence of the variability in epigenetic reprogramming in clones.

Growth performance was characterized in the cohort of eight female Jin Hua pigs produced by Shibata et al. (2006). Compared to control pigs of the same breed, body weight was greater in clones from three to six weeks of age. This difference was transient, as there were no differences in growth parameters between clones and control pigs either at birth or after seven weeks of age. Thus, the authors concluded that growth of Jin Hua pig clones was “essentially identical” to growth of conventional Jin Hua pigs.

(b) ViaGen Dataset

The data on which the following discussion is based are found in Appendix F, along with a more detailed description of the results of the study.

Two groups of swine clones were used for ViaGen Study 1. In the first group, seven clones (one Duroc and six Hamline) were evaluated for survival, health, growth, meat and carcass characteristics. Fifteen conventional barrows (young males) (all Hampshire) were selected as comparators. Because the study was initiated after the birth of the clones (delivered by C-section), the observation period did not begin until shortly after they were weaned. Clones were followed from 50 days after birth through slaughter at approximately 6 months of age. comparators⁹⁵ were selected as age-matched pigs selected from litters sired by the Hampshire nuclear donor boar in a conventional breeding (AI) program.

Clones raised to slaughter weight (approximately 270 lbs) took on average 27 days longer to reach that approximate weight, and when finally slaughtered tended to weigh less than their comparators. These observations are likely due to the husbandry of the clones. Further, because these animals were delivered via C-section, they faced additional stress during the earliest stages of life. In addition, because of the late initiation of the study, these clones were raised under pathogen-free conditions until 50 days of age before being transferred to more conventional (pathogen containing) rearing conditions, while all of the comparators were raised under conventional conditions. Clones also did not receive colostrum, and were deprived of passively transferred maternal immunity. Combined with the change from pathogen-free rearing, the significant immune challenge that the clones experienced would have slowed growth as the animals adapted to their new environment, regardless of whether the animals were produced by sexual reproduction or nuclear transfer.

⁹⁵ Although this study is a more controlled experiment than the retrospective review of the Cyagra clones, the word “comparator” is used for consistency with the discussion of Cyagra data, rather than the more common term “control.”

Four of the clones exhibited appropriate responses to the immune challenge and were able to adapt and grow, albeit at a lower rate than animals which had been raised under more conventional conditions from birth. Three of the clones were considered “poor-doers:” animals that exhibit slow growth rates and other health problems, such as chronic scouring. At slaughter, organ weights as a percentage of body weight were smaller for clones than for their comparators. The clones also had lower blood IGF-I and estradiol levels in their blood than comparators. It is unknown how the change from a pathogen free environment to a more conventional one may have impacted organ weights or hormone status, as none of the comparators were subjected to similar immune challenges because they were all raised under conventional conditions from time of birth (Appendix F). However, given the physiological and immunological stress that the animals experienced, in the opinion of CVM’s veterinarians, these animals performed as well as could possibly be expected.

The second group of swine clones was used to study reproductive function; that discussion is found in Section iii.

(c) Summary Statement for Juvenile Development and Function in Swine Clones (Developmental Node 3)

The dataset reported by Archer et al. (2003a,b) in which both behavior and physiological variables were measured on an individual animal basis is the larger and more tightly controlled study of the two studying juvenile swine clones. Those studies indicate that swine clones overlap their conventional counterparts in behavior and health, and that there are no significant differences between the two groups. Measures of age-appropriate physiological responses (*e.g.*, alkaline phosphatase, phosphorus, and serum protein indicative of increased globulins) indicate that clones are responding normally to growth signals. One case of parakeratosis was observed in this clone cohort. It is not possible to determine whether its incidence was due to SCNT, as it is a condition that is also present in conventional pigs. The ViaGen dataset is less-well controlled, and its outcome confounded by the unconventional shift from a pathogen free environment to more conventional husbandry. Based on the lack of colostrum immediately after birth, and the transfer from pathogen free to conventional housing, it is not possible to ascribe any of the differences in growth to cloning. Further, most of the animals were able to respond appropriately to immune challenges. None of these outcomes were observed in the studies of Archer or Martin, again implying that the changes in husbandry were likely responsible for the outcomes. Finally, none of the swine clones exhibited any adverse outcomes that have not been observed in conventionally bred and reared swine.

iii. Reproductive Development and Function in Swine Clones (Developmental Node 4)**(d) Peer-reviewed Publications**

Martin et al. (2004) described birth outcomes of clone females which were mated via artificial insemination to clone males as normal in duration and uneventful. The 62 live offspring of the clone X clone mating were reported to be normal at birth with the exception of one pig that had contracture of the flexor tendons of both hind limbs. The authors reported that the rate for this abnormality (1.6 percent) was similar to estimates of the frequency within the Australian swine industry (1.2 percent). The stillborn rate for the clone offspring litters was 4.5 percent while a comparator group had a stillborn rate of 8 percent. Evaluation of the semen from the boars, showed similar ejaculate volume, sperm concentration and motility between the clones and comparators. These investigators further reported that 100 percent of gilt clones (5) became pregnant following insemination at second estrus. Consequently, the limited data indicate that gilts and boars from cloning mature similar to non-clones.

Reproductive development and performance were examined in cloned female Jin Hua pigs by Shibata et al. (2006). The age of puberty (defined as receptivity to a boar) in seven of eight gilt clones was within the published range for the Jin Hua breed (109 ± 15 days; this age is young relative to other swine breeds). These gilts exhibited normal estrous cyclicity (cycle length 18-22 days) and duration of estrus (2-3 days). Following two to four rounds of artificial insemination with semen from a naturally produced (non-clone) Jin Hua boar, all seven clones conceived and farrowed spontaneously. Compared to breed- and age-matched controls, mean litter size, number of live-born piglets, and number of piglets surviving to weaning were similar in litters produced by clones and their comparators. Mean birth weight, however, was smaller in piglets born to clone dams (0.72 kg vs. 0.91 kg in control Jin Hua piglets). From these results, the authors concluded that the reproductive performance of their Jin Hua female clones was similar to naturally produced females of the same breed.

Reproduction was measured in four boar clones in the ViaGen dataset (Appendix F). The four clones (three Hampshire and one Duroc) were compared to three genetically related boars derived by AI. No differences were observed between clones and comparators in semen quality. Farrowing rates were higher for swine clones than comparators (73.5 vs. 62.5 percent), although this difference was attributed to the fact that the Hampshire comparator was five years old, and may have been nearing the end of his reproductive life. Litter size was more variable for boar clones, and mean litter size was slightly smaller for clones vs. comparators (10.94 vs. 11.76 pigs/litter), but were similar to US commercial swine production (10.66 pigs/litter).

iv. Post-Pubertal Maturation in Swine Clones (Developmental Node 5)

CVM was not able to identify any peer-reviewed studies on non-reproductive post-pubertal studies in swine clones. The ViaGen dataset (Appendix F) indicated that no remarkable differences were observed between clones and comparators for any of the characteristics evaluated. The small differences in backfat thickness and marbling are likely due to the lighter weight of clones vs. comparators at slaughter.

v. Progeny of Swine Clones

Martin et al. (2004) reported on the pre-weaning performance and health of five litters of piglets derived from one clone boar and five clone gilts. Compared to piglets in comparator litters (five litters of piglets born to conventionally bred parents), litter size, proportion of live births, survival rate to weaning, and weights at birth and weaning in the progeny of the clones were similar. Of the 65 piglets produced by clones in this study, only one was born with a phenotypic abnormality, contracture of the flexor tendon in both hind limbs. The frequency of this congenital anomaly in this study (1.7 percent) was consistent with reported estimates for the Australian swine industry (1.2 percent). A second piglet developed a hematoma in the left ear one week prior to weaning, possibly due to trauma (e.g., from fighting with other pigs). This health event did not affect the growth or weaning rate of this piglet. Daily health observations revealed no other abnormal health conditions in either the progeny of the clones or in comparator piglets.

To investigate whether phenotypic differences in swine clones are transmitted to the next generation, Mir et al. (2005) published a follow-up to the study of swine clones reported by Archer et al. (2003a). To produce progeny, nine clone and five comparator gilts were bred to the same non-clone boar, and all gilts were allowed to farrow naturally. There was no difference in the size of litters produced by clones and comparator gilts, although mean litter sizes were small compared to industry standards (7.8 ± 2.6 and 7.4 ± 3.0 pigs/litter for clones and comparators, respectively). No phenotypic abnormalities were reported. Blood samples were obtained from piglets at 15 and 27 weeks of age for serum chemistry and endocrine measurements, and body weights were recorded at 27 weeks (an age when pigs may be slaughtered for meat). In addition to comparisons between progeny of clones and progeny of non-clones, data from the progeny of the clones were also compared to similar data collected at the same age in their clone parents.

At 27 weeks of age, body weights in the progeny of clones were similar to contemporary controls and to body weights of their clone parents at the same age. In the study of Archer et al. (2003a), mean blood concentrations of creatinine, ALP, and BUN at 15 and 27 weeks of age were less variable in clones relative to comparators. Mean concentrations of phosphorus and calcium in clones were less variable at 15 weeks, and mean concentrations of SGPT, albumin,

triiodothyronine (T3) and cortisol in clones were also less variable at 27 weeks. In the progeny of these clones (Mir et al. 2005), the variability of all of these parameters was similar to the variability in non-clone control pigs, with the exception of BUN at 15 weeks and ALP at 27 weeks. Therefore, the authors conclude that all but two of the blood parameters that were found to be statistically different between clones and comparators by Archer et al. (2003a) were “corrected” in their offspring. Reasons for the difference between the progeny of clones and controls in the variability of BUN and ALP are unknown, but because each of these differences was only observed at a single time point, the authors speculate that these differences may be due to outlier animals rather than a permanent effect on phenotype. These results provide support for the hypothesis that the progeny of clones are physiologically very similar to offspring produced by conventional, non-clone parents.

A large study of progeny of swine clones was submitted by ViaGen, Inc. (Appendix F). The study included data from 402 progeny of swine clones and 300 age-matched, genetically-related comparator pigs and was conducted at the USDA’s Meat Animal Research Center, Clay Center Nebraska, and blood samples were masked and sent to the Cornell University Animal Health Diagnostic Laboratory for clinical chemistry and hematology evaluations.

All of the swine progeny in this study were allowed to farrow and raised to slaughter under similar conditions. The percentage of animals reaching slaughter age was lower for progeny of clones than for comparators (295/402, 73.4 percent vs. 243/300, 81 percent); however, much of this difference can be attributed to the loss of a single litter of clone progeny. When data from this litter is excluded, the percentage of neonatal deaths was similar for progeny of clones and comparators, and was similar to the averages for commercially raised U.S. swine. Abnormalities noted among pigs in this study (e.g., anal atresia and spraddle legs) have been documented in the commercial U.S. swine population at similar rates. There were no consistent differences between progeny of clones and comparators for growth rate, blood clinical chemistry, or hematology, and the few minor differences noted did not indicate any health concerns.

Shibata et al. (2006) described the growth of offspring of their Jin Hua female clones. A total of 44 offspring (23 male and 21 female) derived from six litters were studied. Sires were sexually produced Jin Hua boars. Although birth weight was lower in piglets born to dams that were clones compared to breed-matched controls (0.72 ± 0.02 kg vs. 0.91 ± 0.05 kg, respectively), average daily gain of these offspring was similar to controls until body weight reached 30 kg. Between 30 and 70 kg, daily gain was significantly greater in the offspring of clones than comparators (549 ± 9 g/day vs. 444 ± 20 g/day). It appears that this period of increased daily gain allowed these piglets to “catch up” to their non-clone peers, as there was no difference in body weight between offspring of clone and sexually derived dams at weaning. The offspring in

this study were slaughtered at 70 kg, and carcass characteristics and meat quality are described below in the section on Compositional Analysis.

vi. Conclusions Regarding Food Consumption Risks from Swine Clones and their Progeny

These studies and data evaluated indicate that there are no apparent anomalies present that would have a direct impact on the safety of food products derived from swine clones or their progeny. The measurements taken at 27 weeks of age are appropriate for the evaluation of food consumption risks because this is the approximate age at which pigs are sent to slaughter in the US. The identified abnormalities in the Archer et al. (2003a) (parakeratosis) and the ViaGen dataset (lung adhesion) are not unique to swine clones and do not pose a food consumption risk, as the affected tissues from the carcass would be condemned at the slaughterhouse and would not enter the food supply. The apparently normal status of the clinical measurements indicates that the clones in this study possess the same physiological functions and behaviors as their conventional counterparts, and thus do not contain subtle hazards that would pose food consumption risks compared with food from conventionally-bred swine.

Health information on the progeny of swine indicates that although occasional phenotypic abnormalities were observed in these progeny during the neonatal and perinatal periods (contracture of the flexor tendon, anal atresia, spraddle legs), none of these anomalies are unique to cloning and all occurred at frequencies similar to those observed in conventionally produced swine. Progeny of swine clones are healthy, grow at the normal rate, and do not appear any more susceptible to infection or disease than conventional pigs. Importantly, the studies reviewed indicate that physiological and phenotypic differences that might be observed in swine clones are not passed down to their progeny. These results provide further support for the hypothesis that epigenetic errors in clones are reset during gametogenesis, resulting in progeny that are healthy and physiologically normal. Therefore, based on the CBSA portion of this assessment, we conclude that progeny of swine clones, produced by normal sexual reproduction, do not contain any subtle hazards that would pose any increased food consumption risk compared with the offspring of any other sexually reproduced swine.

c. Sheep Clones

i. Peer-reviewed Publications

As sheep were the first mammal to be cloned by SCNT, the relative paucity of papers on the developmental success of sheep clones is somewhat surprising. The seminal paper in the history of animal cloning is that prepared by Wilmut et al. (1997) in which they describe the generation of “Dolly,” the first mammal to be born (July 5, 1996) as the result of SCNT. (Gene, a bull clone

was being gestated at the same time, but due to differences in the length of pregnancy between cattle and sheep, Dolly was born first). Dolly was derived from the mammary epithelium of a 6-year-old Finn Dorset ewe. The trial from which Dolly was derived included cells from two other sources besides the mammary epithelium, and included fetal fibroblast cells from a 26-day-old Black Welsh fetus, and cells derived from a nine-day-old Poll Dorset embryo.

Cell type	Number of embryos transferred	Number of pregnancies/ Number recipients (%)	Pregnancy Duration (days)	Number of Live Lambs	Birth Weight (kg)
Finn Dorset Mammary epithelium	29	1/13 (7.7)	148	1	6.6
Black Welsh Fetal (gd29) fibroblast	34	4/10 (40.0)	152	2	5.6
	6	1/6 (16.6)	149		2.8
Poll Dorset 9 day embryo	72	14/27 (51.8)	156	1	3.1
			149	4	6.5
			152		6.2
			148		4.2
152	5.3				

Table VI-12 summarizes the outcomes Wilmut et al. 1997 paper. Pregnancy rate, as measured by detectable pregnancy at days 50-60 post transfer, ranged between ~ 8 percent to as high as ~50 percent. A total of 62 percent of the implanted fetuses were lost. Wilmut et al. (1997) reported that at approximately day 110 of the pregnancies, four dead fetuses derived from the embryo cell lines were detected. Their surrogate dams were euthanized, and post-mortem examination of the fetuses revealed two cases of abnormal liver development, but no other abnormalities or evidence of infection. A total of eight live lambs were born. One lamb, derived from fetal fibroblasts, died within a few minutes of birth. No abnormalities were noted at the post-mortem. Wilmut et al. cite the mortality rate of 12.5 percent (1 of 8) as similar to that observed in a large study of commercial sheep breeding, where 8 percent of the lambs died within 24 hours of birth. The birth weights of all of these sheep were within the range of single lambs born to the surrogate Blackface dams used at the Roslin farm (up to 6.6 kg), and were reported to be appropriate to the birth weights of the donor breeds.

The following year, Shiels et al. (1999) compared the telomere lengths of Dolly and one of each of the sheep derived from the different cell sources described in Table VI-12, with age-matched control sheep, donor mammary gland tissue, and donor cells. As expected, the mean size of

telomere fragments in control animals decreased with increasing age. Mean telomere sizes were smaller in all three sheep clones than in age-matched controls. Dolly's mean telomere size in particular, was smaller than other one-year-old age-matched sheep, and more consistent with the telomere fragment sizes derived from a 6-year-old sheep (the age of the animal from which the donor cells were derived). These observations led to speculation that clones would reflect the age of the donor cell, rather than effectively "resetting the biological clock" to their chronological age.

Dolly's health was scrupulously observed over the course of her life. She developed arthritis at an early age, and was reported to have been overweight. Dolly was euthanized in early 2003 at approximately six and one half years of age having contracted a virulent form of lung disease that was endemic at the facility where she had been housed. It is not clear whether any of the abnormalities that were observed with Dolly were the result of SCNT, the conditions under which she was housed, or some combination of the two.

In 2002, Edwards et al. described postnatal growth and hypothalamic-pituitary-adrenal (HPA) function during the first 4 weeks of life in female Merino lambs produced by SCNT using granulosa cells as donor cells. Similar to the studies of Wilmut et al., there was a high rate of attrition of lambs clones in this study; of 209 recipients, 93 ewes (44 percent) became pregnant but only 43 (21 percent) of these pregnancies proceeded to term. Thirty-three lambs died at or shortly after birth, and of the 15 lambs that survived the neonatal period, only seven lived for more than 10 days; four of these lambs were used for the study. Neonatal deaths were attributed to either respiratory distress or bacterial infection. Comparators in this study were female lambs produced by natural mating (controls) and lambs produced by *in vitro* maturation and fertilization (IVC). No gross physical abnormalities were reported in the clones. At birth and during the first four weeks of life, there were no significant differences among groups in gestation length, birth weights, or several growth parameters (crown rump length, abdominal circumference, ponderal index, weight gain, crown-rump length growth rate, abdominal circumference growth rate). Although birth weights were similar among groups, the authors noted a high degree of variability in birth weights in the lamb clones (1.9 – 11.0 kg). The authors attribute this wide range of birth weight in genetically identical fetuses to the influence of intrauterine environment on prenatal growth.

From Day 1 to Day 28 of life, concentrations of plasma glucose and insulin were similar among controls, clones, and IVC lambs. Plasma cortisol concentrations, however, were approximately two-fold higher in clones and IVC lambs compared to controls. Plasma concentrations of adrenocorticotropin (ACTH) were also significantly elevated in clones compared to controls, while plasma ACTH concentrations in IVC lambs were intermediate (*i.e.*, not significantly different from either controls or clones). Reasons for the elevated cortisol and ACTH were

unclear, but it appears most likely that disruption of the hypothalamic-pituitary axis in this study was more related to *in vitro* manipulation rather than cloning *per se* because hypersecretion was observed in both clones and IVC lambs. In sheep, parturition is believed to be triggered by an increase in fetal ACTH and cortisol secretion. Therefore, it is interesting to speculate that the postnatal elevations in ACTH and cortisol observed in clones and IVC lambs in this study may reflect a continuation of the prenatal state and thus indicate a delayed/prolonged adaptation to life outside the uterus.

Most of the other papers in the literature refer to sheep clones generated from transgenic somatic cells to propagate animals with pharmaceutical potential, and data in those papers deal with expression of transgenes, molecular mechanisms that may be involved with fetal overgrowth syndromes (Young et al. 2001), or techniques to increase survival of nuclear transfer (Papadopoulos et al. 2002; Ptak et al. 2002) or other *in vitro* produced embryos. McCreath et al. (2000) reported on the post-mortem examination of transgenic lambs that died *in utero* or in the perinatal phase of development. These animals revealed a range of abnormalities including a high incidence of kidney defects, liver and brain pathology. This research group did not discuss the health of the transgenic lambs that survived.

Recently, Rhind et al. (2003) published a commentary on pathology findings from both transgenic (n=5) and non-transgenic (n=3) sheep clones that were not viable after birth. (The transgenes were intended to be targeted deletions of the α -1,3 galactosyl transferase or prion protein genes). Of the eight animals evaluated, seven were euthanized at birth or shortly thereafter, the eighth survived but was euthanized after 14 days. The authors concluded that many of the defects (*e.g.*, hepatobiliary changes, kidney structure changes, and pulmonary hypertension) may not be contained within the “large offspring syndrome” (LOS) classification that may be common to other animal clones. Pulmonary hypertension has been observed in transgenic cattle clones (Hill et al. 1999), and in swine clones derived from transgenic “knock-out” piglets (Lai et al. 2002), suggesting that this syndrome may be common to a many species of animal clones, and that a common defect may be responsible. The authors call for additional research into the developmental mechanisms that may be responsible for the common defects in clones, although it is important to note that most of the animals with defects were derived from transgenic donor cells. The relevance of these observations to food consumption risks are limited, as clones that have died would not be used for food consumption purposes.

ii. Conclusions Regarding Food Consumption Risks from Sheep Clones

Very few conclusions can be drawn about the health of sheep clones, due to the small database available for evaluation. Only one study provided detailed physiological data from sheep clones, and these data were limited to only a few metabolic and endocrine endpoints. Despite Dolly's

high public visibility, there are very few other reports of non-transgenic sheep clones. Until additional specific information regarding the health of sheep clones becomes available, the only inferences that can be made would be drawn from interspecies extrapolation from other ruminant clones, *i.e.*, cattle and goats.

d. Goat Clones

Relative to cattle, the database on goat clones is relatively small, but quite rich for its size. Much of the work that has been reported on non-transgenic goat cloning arises from data collected in an attempt to perfect systems by which SCNT can be harnessed to develop transgenic goats for commercial applications, and are effectively limited to publications from one group.

i. Perinatal Development and Function in Goat Clones (Developmental Node 2)

In 2002, Keefer et al. (2002) published a report on the birth of nine goat clones derived from two lines of adult granulosa cells and one line of fetal fibroblasts. Ninety-one female granulosa cell-derived embryo clones were transferred into eight surrogate dams. Four of those dams became pregnant, as confirmed by ultrasound on gestational day 30 and 60. All of these pregnancies went to term, and seven clones were born. Table VI-13 summarizes the outcomes of Keefer et al. 2002. One of the recipients delivered a single kid; the remaining three surrogates gestating granulosa cell-derived clones delivered twins. One of the female twins died at birth, but appeared to be normal.

Surrogate	Donor Cell Type	Gestation (days)	Birth weight (kg)	Gender	Status	Suckling Response
1	Granulosa Line 1	144	1.8	Female	Live	Good
2	Granulosa Line 2	150	2	Female	Live Dead	None NA ¹
	Granulosa Line 2		1.2	Female		
3	Granulosa Line 2	145	1.6	Female	Live Live	Poor/None Poor/None
	Granulosa Line 2		1.4	Female		
4	Granulosa Line 2	145	1.5	Female	Live Live	Good Good
	Granulosa Line 2		2.2	Female		
5	Fibroblast Line 1	148	1.5	Male	Dead Live	NA None
	Fibroblast Line 1		1.2	Male		

¹ NA = not applicable

In addition, 54 male fetal fibroblast-derived embryo clones were implanted into six surrogate dams. Only one of those dams had a confirmed pregnancy and delivered two male kids, one of which died during delivery. The authors state that this kid also appeared normal.

The birth weights of all of the kids were cited as being within the normal range for this breed (Nigerian Dwarf) at an average of 1.7 ± 0.13 kg versus 1.3 ± 0.06 kg for females resulting from natural breeding. The authors reported that the placentae of these kids appeared normal, and had cotyledon numbers that were comparable to those from the placentae of naturally bred Nigerian Dwarf goats. Suckling response was delayed in half of the granulosa cell-derived kids, and in the fibroblast-derived kid. These animals were fed colostrum by intubation, and good suckling was reported to occur by Day 2. The clones were otherwise reported as healthy, and having no apparent abnormalities.

ii. Juvenile Development in Goat Clones (Developmental Node 3)

In the Keefer et al. (2001a) study, the authors reported that blood profiles of the clones were monitored for one year, and showed no anomalous results. No data addressing this statement were presented in the Keefer et al. (2002) publication. There is, however, an abstract that was published in 2001 (Keefer et al. 2001b) in which some blood parameters are provided (Table VI-14: Selected Laboratory Parameters for Goat Clones). In this brief account, only a few measurements were reported; the duration of monitoring was six months. Alkaline phosphatase levels show the appropriate age-related changes to be expected for rapidly growing infants and very young animals, dropping to lower levels as the animals aged. Given the adult range for alkaline phosphatase in Nigerian Dwarf goats is reported as 16-33 U/L, these animals likely were still growing at 6 months of age.

Behboodi et al. (2005) compared hematology and blood clinical chemistry of four transgenic goat clones with four age-matched comparators and a published range for goat blood values (Pugh 2002). Hematology values were similar between clones and comparators, and all hematology values fell within the published range (Pugh 2002). For clinical chemistry, 18/24 values were not significantly different between clones and their age-matched comparators. Of the 19 clinical chemistry values for which published ranges were available, 18 of the values for clones and comparators fell within the published range. The one value out of the published range, creatine kinase (244.6 vs. 204.4 IU/L for clones and comparators), was not different between clones and comparators.

Table VI-14: Selected Laboratory Parameters for Goat Clones (from Keefer et al. 2001b)			
Measurement	1 week	3 months	6 months
Lymphocyte counts (cells x 10 ⁹)			
Clones	2.34	4.94	9.84
Control ¹	2.64	5.94	7.14
Glucose (mmol/L)			
Clone	5.64 ± 0.3	4.14 ± 0.2	3.54 ± 0.1
Control	4.84 ± 0.3	4.54 ± 0.1	3.54 ± 0.1
Alkaline phosphatase (U/L)			
Clone	7,434 ± 84	5,554 ± 73	374 ± 33
Control	Not provided	Not provided	Not provided

¹ Controls were reported as taken from Mbassa et al. 1991 (*Zentralbl Veterinarmed [A]* 38: 510-522). It is likely that these values are from adult animals, as the control values for the alkaline phosphatase levels were not explicitly provided. Instead, the adult range of 16-33 U/L was cited.

iii. Reproductive Development and Function in Goat Clones (Developmental Node 4)

One paper compares the sexual maturation and fertility of male Nigerian Dwarf clones to conventional bucks (Gauthier et al. 2001). Three clones (Stewart, Clint, and Danny) and four conventional animals that served as controls (Blue, Star, Banzai, and Ed) were trained to serve an artificial vagina beginning at the age of one month. Average age at first semen collection for both clones and controls was approximately 20 weeks, although volumes were small at the initial collection (<0.1 ml). Subsequent collections showed increased volume and increased sperm count (see Table VI-15 for a summary of the reproductive function in goat clones, comparators, and clone progeny).

Semen collected from two goat clones, Clint and Danny, at seven months was used to impregnate six Nigerian Dwarf does (three does for each buck). Although not explicitly stated, the implication is that the does were not clones. Five of the six does became pregnant. Two does impregnated by Clint gave birth vaginally to two sets of twins. Two does impregnated by Danny gave birth to singletons, and one doe gave birth to triplets. Nine kids were produced, and all appeared to be normal and healthy. Average birth weights for the male and female clone progeny were 1.7 ± 0.2 kg and 1.66 ± 0.1 kg, respectively, which do not differ significantly from average birth weights for conventional animals of this breed (1.7 ± 0.07 kg (n = 41) for males and 1.3 ± 0.31 kg (n = 79) for females). Semen was first collected from one of the progeny males at 28.4 weeks (Table VI-14).

Table VI-15: Reproductive Function in Goat Clones, Comparators, and Clone Progeny
(from Gauthier et al. 2001)

Animal Derivation	Number of bucks	Mean age at collection \pm SEM ¹ (weeks)	Mean sperm concentration \pm SEM (sperm $\times 10^9$ /ml)	Mean Ejaculate Volume \pm SEM (ml)	Range of Motility %
Control bucks	4	20.2 \pm 3.1	ND ²	ND	ND
	3	36.5 \pm 0.3	2.5 \pm 0.6	0.37 \pm 0.14	~70-90
	3	59 \pm 1	2.1 \pm 0.6	0.4 \pm 0.06	~45-90
Clones	3	20.2 \pm 1.2	0.6 \pm 0.07 ³	0.25 \pm 0.1 ³	75-85
	3	23 \pm 0.6	1.2 \pm 0.71	0.28 \pm 0.11	30-98
	2	79.5 \pm 0.5	4.4 \pm 0.3	0.37 \pm 0.02	75-90
Progeny buck	1	28.4	4.6	0.4	65

¹ SEM = standard error of the mean
² ND = not done
³ The sample from the first collection from one buck was too small to measure

The authors concluded that male Nigerian Dwarf goat clones developed sexual maturity similarly to their conventional counterparts. Further, these goat clones are fertile, and their progeny appear to be fertile as well.

In their study of goat clones generated from transgenic fibroblasts, Reggio et al. (2001) were able to produce a total of five healthy kids. Twenty-three surrogate dams were each impregnated with an average of eight embryo clones. Five of the dams that were detected as pregnant at day 30 completed their pregnancies, and gave birth naturally, providing a 100 percent success rate based on detectable pregnancy. All of the kids appeared healthy and vigorous. Birth weights averaged 3.8 kg (normal for the Toggenberg breed that served as the donor cell), and weaning weights were also within normal range (19.1-24.5 kg) for the breed. Each of the kids exhibited estrus, and has been bred to a buck. No reports of progeny were provided. Although this study is based on transgenic clones, it reiterates the high success rate that is experienced by researchers producing goat clones.

iv. Post-Pubertal Maturation in Goat Clones (Developmental Node 5)

CVM was not able to identify any published reports of measures of post-pubertal non-reproductive maturation in goat clones. Further, in the course of several presentations at scientific meetings, the Center learned that that the cohort of clones studied in Keefer et al. 2001a and 2002 has been terminated for business reasons.

v. Summary Statement on Health Status of Goat Clones

Based on these data, goat clones appear to have the least difficulty of any of the livestock species with respect to the SCNT process. Successful pregnancy outcome (when confirmed by ultrasound detection) is very high, and clones appear to be born at birth weights within the appropriate breed- and species- range. Suckling response was weak in some of the goat clones immediately after birth, but they appear to have recovered within one day. Available information on physiological parameters indicates that these animals appear to be normal. Data on reproductive function in these animals indicates that they enter puberty at the normal age range, produce viable semen, and normal, live offspring. The minimal reporting on one progeny animal also indicates that progeny are fertile.

vi. Conclusions Regarding Food Consumption Risks from Goat Clones

Based on the data reviewed, there do not appear to be any anomalies present in the goat clones that would have an effect on the safety of food products derived these animals, and no subtle hazards were identified in these clones that could pose food consumption risks. Goats appear to be relatively “cloning friendly” with a high degree of successful live births following confirmation of pregnancy. All reports of health of the goat clones seem to indicate that they are normal and healthy. The available data on the physiological parameters of goat clones indicate that these animals respond as their conventional counterparts to internal signals for growth. The apparently normal status of the clinical measurements indicates that the clones in this study possess the same physiological functions and behaviors as their conventional counterparts. Further, unlike the other livestock clones, data on the reproductive behavior of male goat clones indicate that reproductive function is normal. Finally, although cursory in mention, it appears that male progeny of clone bucks also reach puberty at the appropriate time. Thus, although the number of animals that has been evaluated is not as large as in the case of bovine clones, goat clones appear to be healthy, and do not appear to be materially different from conventional goats.

3. Compositional Analysis Method**a. Overview**

The operating hypothesis of the second prong complement to the Critical Biological Systems Approach is that if food products from healthy animal clones and their progeny meet the local, state, and federal regulatory requirements set forth for those products (*e.g.*, Pasteurized Milk

Ordinance,⁹⁶ USDA inspection criteria, absence of drug residues), and are not materially different from products from conventionally bred animals, then they would pose no more food consumption risk(s) than corresponding products derived from conventional animals.

Information on the composition of meat or milk from animal clones has been limited for several reasons. Few of the cattle clones are old enough to have been bred, given birth, and begun lactating. In addition, there is uncertainty regarding the kinds of analyses that could or should be performed in order to determine whether milk from animal clones is materially different from milk from non-clone animals. The issues associated with the compositional analysis of meat are similar, but have additional practical and economic components. During the course of preparing this Risk Assessment, CVM has contacted several food testing laboratories to inquire about the minimum sample size that would be required in order to perform a compositional analysis of meat. The Center's hope was that systems were sufficiently miniaturized to allow analysis of "punch biopsies" of a shoulder or rump, but were informed that the minimum sample size would require sacrificing an animal. Nonetheless, there are now several studies that have evaluated the composition of the milk and meat of both cattle and swine clones, and one large study that has evaluated the composition of the meat of the progeny of swine clones.

b. Nutritional Risk

The primary concern for milk and meat from animal clones is that inappropriate reprogramming of the nucleus of donor cells does not result in epigenetic changes creating subtle hazards that may pose food consumption risks (Chapter III). Because, as previously discussed, there is no *a priori* reason to expect that SCNT will introduce any new, potentially toxic substances into the milk or meat of otherwise healthy animals, the remaining food safety concerns addressed whether subtle changes have occurred that would alter the presence of important nutrients. The most likely dietary risk would then be the absence or significant decrease in levels of vitamins and minerals whose daily requirements are in large part met by milk or meat.

The overall strategy we used to determine which milk or meat components could characterize their respective nutritional "footprints" involved selecting certain key nutrients and compositional parameters, while at the same time allowing sufficient flexibility in the non-essential components that vary with the genetic make-up and husbandry of the production animal. Finally, evaluation of the levels of the results of complex biochemical pathways in clones (*e.g.*, saturated fats, vitamins) can further ensure that the clone is functioning appropriately, and thus indirectly support the hypothesis that the clones are appropriately

⁹⁶ The Grade "A" Pasteurized Milk Ordinance recommends for statutory adoption regulations for the production, collection, processing, sale, and distribution of milk and certain milk products.

reprogrammed and not materially different from their conventionally bred counterparts.

In order to identify the nutrients in milk or meat whose alterations would most likely affect the overall diet, even if all of the dairy and meat products from conventional animals in the daily diet were replaced by counterparts produced by clones, we first determined which nutrients made a “major” or “moderate” contribution to the total daily diet of milk or meat consumers. For the purposes of this Risk Assessment, a nutrient in meat or milk was considered a major dietary source if it provides 50 percent or more of its recommended dietary allowance (RDA) in that food.⁹⁷ Likewise, a nutrient in a food providing 10 to 50 percent of its RDA in that food is considered a moderate dietary source. For example, a single eight ounce serving of whole milk provides milk drinkers with between 10 to 50 percent of the RDA of vitamin B₁₂, riboflavin (B₂), pantothenic acid (B₅), calcium, phosphorous and selenium. Another example, a single serving of three ounces of roasted eye of round beef provides a meat consumer with a moderate source of zinc, niacin, vitamin B₆, phosphorus, iron, and riboflavin, and a major source of vitamin B₁₂ and selenium.

In order to determine typical meat and milk consumption in the adult US population, we consulted the one-day food survey conducted by the National Health and Nutrition Examination Survey (NHANES) of 2000-2001. According to NHANES, the mean daily consumption of milk among adult milk drinkers was 11.5 ounces. At this level of consumption, milk becomes a major source of vitamin B₁₂, and a moderate source of thiamin (B₁), zinc, and potassium, in addition to the nutrients previously listed as provided in moderate amounts. The same survey showed that the 90th percentile consumption of milk by users was 24.1 ounces per day, making milk a major source of calcium, phosphorus, riboflavin, and pantothenic acid, and adding magnesium and vitamin B₆ to the list of nutrients provided in moderate amounts. Among subjects who consumed meat, the mean intake of meat was 4.2 ounces or 120.2 grams. Among the 90th percentile of meat eaters, consumption was 8.4 ounces or 239.4 grams. In order to determine whether evaluating the mean or 90th percentile consumption of milk and meat, changed the actual number of nutrients designated as moderate or major, we found that there were no nutrients were added or deleted, although with increased consumption rates, some of the nutrients changed from moderate to major contributors to the diet.

Proteins are of dietary importance because once they are digested, they provide the body with amino acids. In particular, some amino acids are of dietary concern because of the inability of mammals to synthesize them *de novo* in sufficient quantity to meet the body’s needs. For this reason they are designated as “essential.” Therefore, for purposes of assessing nutritional risk from food products from animal clones, the nature of the protein in its initial food matrix (e.g.,

⁹⁷ <http://www.iom.edu/?id=4576&redirect=0>

casein or actin) is less important than whether it contains the same level of essential amino acids as its counterpart in foods derived from conventional animals. Finally, certain fatty acids such as linolenic (18:3) and linoleic (18:2) acid are essential components of the diets of mammals (including humans) and have also been selected as “key nutrients.”

Table VI-16 summarizes the analytes that we believe could be used to assess the composition of milk and meat from clones and comparators to demonstrate that there are no material difference between the two groups of animals with respect to key nutrients and overall nutritional characteristics. Included are key essential vitamins, minerals, and fatty acids. Other less essential constituents (e.g., vitamin A in milk is often supplemented, iron is not a key nutrient in milk) are also included to illustrate that checking on the levels of non-essential nutrients can also provide a useful tool to demonstrate the similarity of milk and meat from clones and their contemporary comparators.

Table VI-16: Compositional Analyses of Milk and Meat That May be Used for Showing No Material Differences Between Clone and Comparator Food Products¹	
Milk Composition	Meat Composition
Proximates ²	Proximates
Vitamins and minerals for which milk is a moderate to major source Vit A, C, B ₁ , B ₂ , B ₁₂ , niacin, pantothenic acid, Ca, Fe, P	Vitamins and minerals for which meat is a moderate to major source Vit A, C, B ₆ , B ₁₂ , niacin Ca, Fe, P, Zn
Fatty Acid Profiles Saturated: 4:0, 6:0, 8:0, 10:0, 12:0, 14:0, 16:0, 18:0 Unsaturated: 18:1, 18:2, 18:3	Fatty Acid Profiles Saturated: 10:0, 12:0, 14:0, 16:0, 18:0 Unsaturated: 18:1, 18:2, 18:3, 20:4 Cholesterol
Protein characterization Essential amino acid profile	Protein characterization Essential amino acid profile
Carbohydrate	
¹ The information in this table was compiled from FDA’s Nutritional Labeling Requirements (21 CFR 101.9, 9 CFR 317.300) and USDA’s Nutrient Database (http://www.ars.usda.gov/ba/bhnrc/ndl). ² Most foods are comprised of water, protein, fat, ash, and carbohydrates; the sum of these values approximates a complete analysis, hence the term “proximates.”	

i. Milk

For the purposes of this Risk Assessment, CVM uses the term “milk” to mean the “lacteal secretion, practically free from colostrum, obtained by the complete milking of one or more healthy cows,” and that “milk that is in final package form for beverage use shall have been pasteurized or ultra-pasteurized, and shall contain not less than 8 1/4 percent milk solids not fat and not less than 3 1/4 percent milkfat” (21 CFR 131.110(a)).

The Grade “A” Pasteurized Milk Ordinance (a model ordinance for adoption by states, counties and municipalities to regulate the production, collection, processing, sale and distribution of milk and certain milk products) echoes this definition, replacing “cows” with the term “hooved animals.” Therefore, in this Risk Assessment, unless otherwise specified, the term “milk” will refer to the lacteal secretions of cows, goats, or sheep. Although most of the discussion in this Risk Assessment refers to cow’s milk, similar arguments may be applied to the milk of goats or sheep. Other hooved animals whose milk is covered by the PMO include water buffalos, although they are not covered by this Risk Assessment.

The biological role of the milk of any mammal is to provide nutrition to its own newborn and young. In addition to mother’s milk, humans consume the milk of a few other species, principally from cows. Milk and milk products provide a considerable portion of the nutrition of other age groups, including growing children and adolescents, pregnant and lactating women, and the elderly. In 2001, *per capita* American consumption of milk among all age groups was approximately 23 gallons of fluid milk, 30 pounds of cheese, and 27 pounds of frozen dairy products (USDA ERS 2003).⁹⁸ In particular, bovine milk and milk products (excluding butter) provided approximately nine percent of the energy, 19 percent of the proteins, 12 percent of the fats, and 4.5 percent of the carbohydrates consumed by milk drinkers in the US in 2001⁹⁹. Ensuring that these dietary levels do not alter significantly is a key component of evaluating the potential nutritional risk from the milk of animal clones.

The degree to which individuals may experience risk from the consumption of milk appears to be a function of individual susceptibility, rather than the intrinsic toxicity of milk. For example, certain individuals suffer from Cow’s Milk Protein Allergy, which has an incidence of 2-6 percent among young infants (Exl and Fritsché 2001). Cow’s Milk Protein Allergy usually presents during the first year of life, and generally resolves by school age (Bernstein et al. 2003). Lactose intolerance is another milk-related condition found in adults and children (to a lesser degree) that is also a function of individual physiology (*i.e.*, decreased expression of the enzyme lactase), particularly among certain ethnic groups. Excess consumption of saturated fats, including those from dairy products, can lead to atherosclerosis and its consequent morbidities; again, these harms are a function of individual behavior and susceptibility and not an intrinsic hazard of milk itself.

State regulatory agencies have managed the risk(s) posed by milk by adopting the PMO. It was first developed (1924) by what was then known as the Public Health Service, a precursor to today’s U.S. Food and Drug Administration. Now known as the Grade “A” Pasteurized Milk

⁹⁸ <http://www.ers.usda.gov/Amberwaves/June03/DataFeature/>

⁹⁹ <http://www.ers.usda.gov/Data/FoodConsumption/NutrientAvailIndex.htm>

Ordinance, the PMO is revised biennially (most recently in 2003) by the Center for Food Safety and Applied Nutrition and other centers of the FDA, with input from industry and state regulatory agencies.

Table VI-17: Pasteurized Milk Ordinance (PMO) Requirements for Grade A Milk Compliance		
Standard	Raw Milk	Pasteurized Milk and Bulk-Shipped Heat-Treated Milk
Temperature	Cooled to 10°C (50°F) or less within four (4) hours after the commencement of the first milking, and to 7°C (45°F) or less within two (2) hours after the completion of milking. Provided that the blend temperature after the first and subsequent milkings does not exceed 10°C (50°F)	Cooled immediately to 7°C (45°F) or less and maintained thereat
Bacterial limits: Standard Plate Count	Individual producer milk not to exceed 100,000 mL prior to commingling with other producer milk. Not to exceed 300,000 mL as commingled milk prior to pasteurization	20,000 mL limit
Coliforms	...	Not to exceed 10 mL. Provided, that in case of bulk milk transport tank shipments, shall not exceed 100 mL
Somatic cell counts	Individual producer milk not to exceed 750,000 per mL ¹	...
Drugs	No positive results on drug residue detection methods as referenced in Section 6 of the PMO Milk must not test positive for any drug residues as described in section 6 of the PMO	No positive results on drug residue detection methods as referenced in Section 6 of the PMO Milk must not test positive for any drug residues as described in section 6 of the PMO
Phosphatase	...	Less than 350 milliunits per liter for fluid products and other milk products by the Fluorometer or Charm ALP or equivalent.
¹ Goat milk somatic cell count NTE 1,000,000 cells/mL. Source: 2003 Grade A Pasteurized Milk Ordinance.		

Table VI-17 lists the PMO requirements for Grade A milk (as adopted by state and local governments). As milk from dairy clones would be subject to the same requirements as that from

conventional dairy cows, potential risks associated with subtle changes in immune function that might result in increased rates of mastitis, for example, would be controlled by the somatic cell and bacterial load requirements of the PMO. Likewise, even if clones suffered more bacterial infections, and required additional treatment with antibiotics, existing requirements restrict the presence of antibiotic residues in Grade A milk, thereby ensuring that milk from clones would pose no more bacteriological or drug residue risk than milk from non-clone cows.

Is it possible to be reasonably certain that milk from animal clones and their progeny is indistinguishable from that now available in commerce? The complexity of milk itself is one of the primary difficulties in determining whether residual non-PMO managed hazards exist in the milk of animal clones. Milk from cows, sheep, and goats are mixtures that are estimated to be composed of more than 100,000 molecules (Jeness 1988), whose presence and proportion is a function of both the genetics of the animal and its environment. Not every component in milk has been identified and characterized; thus determining whether animal clones are producing a hazardous substance in their milk although theoretically possible, is highly impractical.

As for new components or changes in currently present but unknown and uncharacterized components of milk, it is unlikely that the cloning process would trigger expression of a novel substance that would not have independently arisen through random mutations in cow populations. In addition, it seems unlikely that a reprogramming error would lead to expression of an excess of a metabolically active protein with no adverse effects on the producing animal itself. This is especially true if the many nutrients that are monitored by the comparison scheme in Table VI-16 are within the ranges of contemporary comparators, and the physiological and biochemical parameters monitored in assessments of animal health are also within the ranges exhibited by contemporary comparators.

All milk is subject to Nutrition Labeling Requirements promulgated by FDA's Center for Food Safety and Applied Nutrition under 21 CFR 101.9. These requirements provide a good starting point for milk characteristics that could be used as a basis of comparison. Additionally, the USDA Nutrient Database for Standard Reference (<http://www.ars.usda.gov/ba/bhnrc/ndl>) compiles data from a range of scientific, technical, food industry, and government agency sources to arrive at "composite" values of key nutrients in milk from cows, sheep, and goats. Table VI-16 provides a compilation of the key constituents and nutrients of milk from FDA's Nutritional Labeling Requirements and USDA's Nutrient Database.

If milk from clones and conventional animals does not materially differ in these constituents, it is unlikely that individuals consuming milk from animal clones will face increased risk(s) relative to individuals consuming milk from conventionally bred animals.

ii. Meat

For purposes of this Risk Assessment, CVM uses the term “meat” to mean “(1) *The part of the muscle of any cattle, sheep, swine, or goats, which is skeletal or...tongue,...diaphragm,...heart, or...esophagus, with or without the accompanying and overlying fat, and the portions of bone, skin, sinew, nerve, and blood vessels which normally accompany the muscle tissue... It does not include the muscle found in the lips, snout, or ears....*” and “(2) *The product derived from the mechanical separation of the skeletal muscle tissue from the bones of livestock using the advances in mechanical meat/bone separation machinery and meat recovery systems that do not crush, grind, or pulverize bones, and from which the bones emerge comparable to those resulting from hand-deboning....*” (9 CFR 301.2)

Meat comprises a large proportion of the average American’s diet, for both cultural and economic reasons (meat is relatively inexpensive in the US). In 2001, total annual per capita consumption of beef, veal, pork, lamb and mutton on a retail weight basis was approximately 122 pounds, and is estimated to have been about the same for 2002. The species-specific breakdown is approximately 69 pounds from beef and veal, 52 pounds from pork, and a little over a pound for lamb and mutton (USDA-NASS Statistical Highlights of US Agriculture 2001 and 2002¹⁰⁰). Goat consumption tends to be centered in various ethnic groups, but when averaged over the US population is about a half a pound per capita per year (USDA).

Meats provide a substantial portion of the nutrition in a non-vegetarian American diet. For example, beef provides approximately 50 percent of the total protein in a 2,000 calorie American diet, as well as approximately a third of the daily requirement of zinc and vitamin B₁₂. It provides about 20 percent of the daily requirement for selenium, phosphorus, and niacin, and lesser although substantial amounts (*i.e.*, 10-15 percent) of daily requirements for vitamins B₆, riboflavin, thiamin, and iron (USDA Nutrient Database for Standard Reference Release 15, 2002¹⁰¹).

Similar to milk, consumption of meat for millennia has taught that there are no significant intrinsic toxicants in meat from cattle, swine, sheep, or goats. Examples of meat allergies are rather rare, although they do exist. Cases of human immune-mediated allergies to the cattle

¹⁰⁰ <http://www.nass.usda.gov/index.asp>

¹⁰¹ <http://www.nal.usda.gov/fnic/foodcomp/search/>

proteins bovine serum albumin and bovine gamma globulin have been reported (Wuthrich et al. 1995; Han et al. 2000; Fiocchi et al. 2000; Tanabe et al. 2002). Humans allergic to cat serum albumin may also exhibit cross-reactivity to swine serum albumin (Hilger et al. 1997), in a phenomenon referred to as the pork-cat syndrome (Drouet et al. 1994). Children exhibiting positive skin prick test to bovine serum albumin may also cross react with sheep serum albumin (Fiocchi et al. 2000). As is the case for all immune-mediated allergic response, the individual's susceptibility is in large part the driver for the response, as allergies are examples of the dysfunction of the immune response.

Just as for milk, there are no chemical composition schemes that “define” beef, pork, mutton, or goat meat. Due to the physiological function of muscles, and their need for rapid perfusion and oxygenation, meat also reflects the materials circulating the blood of the animal prior to slaughter. Myoglobin, the major storage protein for oxygen, is found in high concentration in muscle tissues. Unknown numbers of other large and small molecules are also found in meat, whose origins can be environmental, dietary, or endogenous. Each of these contributes to the complex profile that is responsible for the distinctive tastes and smells of meats.

The muscle tissue that makes up meat is composed of two major protein types: myofibrillar proteins, actin and myosin, which make up the fibers in muscle bundles, and connective tissue, which primarily consists of collagen and elastin. Collagen is the major component of gelatin, which results from the melting of collagen in the presence of hot water. Elastin is not greatly affected by cooking.

Tenderness, one of the primary considerations in carcass merit, is affected by the interplay of the myofibrillar and connective tissue proteins, and changes over the age of the animal and the amount of time since slaughter. The more connective tissue there is in a piece of meat, the tougher it tends to be; cooking, by solubilizing the collagen, decreases meat toughness. Collagen levels and structure tend to change in animals as they age, with the amount in young animals considerably lower than in older animals. With age, collagen undergoes more cross-linking, rendering it more insoluble and less likely to dissolve during the cooking process. The amount and distribution of fat in a muscle also influences tenderness. Marbling, or the presence of fatty deposits within muscles, also affects tenderness by functioning as a “lubricant” on the teeth or in the mouth, and by leaving “pockets” between muscle bundles as it melts during cooking. Changes in the amount of collagen or fat in the animal may affect meat quality with respect to tenderness or other qualities, but these would not pose nutritional or other food consumption risks. Further, it is likely that beef cattle clones will have changes in the amount or nature of marbling relative to average conventional beef cattle, as breeders will select animals as nuclear donors that have carcass qualities producing more uniformly tender and tasty meat. Similar selection procedures are being applied to animals used in conventional animal breeding

programs, so the effect of cloning would be to speed the rate at which these desirable traits are introduced into breeding and production herds.

When an animal is slaughtered, *rigor mortis* (muscle stiffening observed after death) causes stable cross-links to form in muscle fibers due to the free flow of calcium across the cell membranes. The carcass stiffens and lactic acid levels accumulate resulting in a decrease in pH. The net result is that muscle fibers contract, and the meat appears “tough.” As the meat ages, however, a set of enzymes called calpains (calcium activated proteases) break down some of the structural components of the muscle, relieving the contraction, and degrading the connective tissue proteins, also releasing the degree to which the muscles are held together. Calpains are thought to function in concert with their antagonistic regulators, calpastatins, such that if calpastatin levels are high, calpain activity will be inhibited and less post-mortem degradation will occur, resulting in tougher meat. Most processors age beef for a minimum of 14 days to allow sufficient time for the calpains to work. Changes in levels of either calpains or calpastatins may thus affect meat tenderness, but likely would not pose food consumption risks.

As is the case for milk, the question of the appropriate comparator for meats may be approached from two perspectives. In order to determine whether cloning results in potential food consumption hazards relative to close genetic relatives, comparisons could be made to animals that are matched as closely as possible by age, husbandry (including diet), and environment. The second approach compares meat samples from animal clones more broadly to the national herds by using composite data sources.

Unlike milk, however, meat consists of various cuts that although made up mostly of muscle, contain different minor tissues, and whose function may affect composition. For example, the muscle in loin cuts may differ in composition from the muscle used to make bacon (*i.e.*, belly muscles). In order to provide the most useful data for purposes of determining similarity to conventionally bred animals, it would be useful to compare cuts from each species that have the following characteristics:

- High US consumption levels (*e.g.*, loin, rib, shoulder roasts, pork bellies, lamb or mutton shoulder or leg), and
- Cuts of different muscularity that may have different overall compositions (*e.g.*, if one tissue is lean, another may be fatty).

USDA’s Nutrient Database (<http://www.ars.usda.gov/ba/bhnrc/ndl>) contains composite tables that provide chemical compositions of several cuts of beef, pork, and lamb meats. Goat meat composition is only available as a single source.

There are no full chemical characterizations for meats. Moreover, as the definition of meat actually contains several tissue types, and each varies according to the genetics, breed, species, and environment of the food animal, it is unlikely that “complete” characterizations will ever be developed. The USDA requires nutritional labeling on “mixed” pork and beef products, and allows the voluntary labeling of raw products (9 CFR 317.300). Included in the labeling are calories, calories from fat, total fat, saturated fat, cholesterol, sodium protein and iron. Because meats are declared not to be a significant source of total carbohydrate, dietary fiber, sugars, vitamins A and C, and calcium, USDA does not require labeling information on them.

c. Characterization of Milk from Cow Clones

i. Peer-reviewed Reports

Walsh et al. (2003) evaluated the composition of milk produced by 15 dairy cow clones from five different donor cell lines and three different breeds. These animals were produced by Infigen, Inc., and they have been described by Forsberg et al. (2002), reviewed in the Critical Biological Systems Section earlier in this Chapter. Clones were bred by AI between 14 to 16 months of age; the paper does not specify whether all of the heifers were inseminated with semen from the same bull. Five different cell lines were used as donors to generate the clones, and the breeds represented by the cell lines included two Holsteins, and two cell lines derived from cows resulting from crossbreeding Jersey and Holstein cattle.

Comparator cows were housed at different farms from the clones, but were approximately age and lactation-stage matched. They consisted of five Holsteins living on one farm, and one Brown Swiss cow raised at a farm different from the clones or the comparator Holsteins. Because of the different rearing sites, clones and their comparators were fed different rations, and for the clones, the ration was changed during the course of the lactation. Each cow was lactating for at least 30 days prior to sample collection, and samples were collected at approximately two month intervals over the entire lactation cycle. In addition to comparators, Walsh et al. (2003) also compared the gross composition of milk (solids, fat, protein and lactose) from cow clones to previously published values (see Table VI-19).

Cows were milked into individual buckets, the contents of the bucket mixed and distributed into various vessels appropriate to each analysis. Samples were coded at the collection site, although the coding was broken approximately half-way through the study for unspecified reasons. The milk components that were analyzed are found in Table VI-18.

Table VI-18: Milk Components Analyzed (by Walsh et al. 2003)	
<ul style="list-style-type: none"> • Total fat • Lactose • pH • Nitrogen • Solids • Somatic Cell Count¹⁰² 	<ul style="list-style-type: none"> • Minerals including: sodium, calcium, sulfur, potassium, zinc, iron, strontium, and phosphorus • Fatty acids including: C 4:0, 6:0, 8:0, 10:0, 12:0, 14:0, 14:1, 16:0, 18:0, 18:1, 18:2, 18:3, and 20:0¹⁰³ • Milk proteins including: total protein, caseins (α, β, and κ subtypes), β-lactoglobulin, α-lactalbumin, and immunoglobulin fraction, and a category entitled “other proteins.”
<ul style="list-style-type: none"> • Acid Degree Value¹⁰⁴ 	

Mastitis is an infection of the udder, and a common problem in dairy cattle, that characteristically causes an increase in the number of somatic cells (cells from the circulation) in the milk of affected cows. The somatic cell count of the milk from both clones and non-clones indicated that none of the milk being sampled came from cows with mastitis. This implies that the immune function of the clones was sufficient to ward off infection under the husbandry conditions that the cows experienced. (A similar lack of impact on somatic cell count was reported by Heyman et al (2004) for milk from 50 clone cows compared to milk from 68 contemporary non-clone controls). The pH of the milk from the clones was within the range of healthy cows (~6.5-6.8). Acid degree values, which indicate rancidity or off-flavors, were also within the normal range for fresh milk.

No significant differences ($p > 0.05$) were observed when the gross composition of milk from Holstein clones and Holstein non-clones was compared over the course of the entire lactation cycle (Table VI-19). The authors state that all values for gross composition in Holstein and Brown Swiss cows fell within the range of values reported for each breed (see Table VI-19). Although a statistical comparison is not possible, it is notable that the values for gross

¹⁰² Somatic Cell Count is a measure of milk quality, and is derived from counting the number of epithelial cells (normally shed cells) and leukocytes (white blood cells that fight infection). Both cell types are normally present in milk at low levels. High levels are likely caused by mastitis, which is an inflammation of the udder, usually caused by a bacterial infection. The Pasteurized Milk Ordinance sets a ceiling for somatic cell counts in milk of dairy animals.

¹⁰³ The International Commission on Biochemical Nomenclature has accepted the following method for fatty acid nomenclature. The number before the colon represents the number of carbon atoms, and the number following the colon represents the number of double bonds in the carbon chain. For example, linoleic acid (or cis-9, cis-12-octadecadienoic acid) is named 18:2; it has 18 carbon atoms, 2 double bonds.

¹⁰⁴ The Acid Degree Value helps to predict off-flavors in milk that arise from the breakdown of fat by an enzyme called lipase. High values of certain free fatty acids can make milk taste rancid. Pasteurization inactivates many lipases, but the acid degree value may still rise slowly during long storage.

composition of milk from Holstein clones¹⁰⁵ reported by Walsh et al. (2003) are within 10 percent of respective values in the current USDA Nutrient Database (<http://www.ars.usda.gov/ba/bhnrc/ndl>).

No significant differences were reported between milk from clones and comparators with respect to the individual milk proteins that were assayed, although difference in the concentrations of α_s -casein, κ -casein, and α -lactalbumin were noted over the course of the lactation.

Table VI-19: Comparison of Gross Characteristics of Milk from Clones and Non-Clones (from Walsh et al. 2003)									
	Clone BrSw ¹	Non- Clone BrSw	BrSw Lit Value ²	Clone Hlstn 1 ^{3,4}	Clone Hlstn 2	Non- Clone Hlstn	Hlstn Lit Value	Clone H X Jersey 5 1	Clone H X Jersey 5 2
Animals/ Samples	1/5	1/5	NP ⁶	1/5	11/63	5/26	NP	1/5	1/5
Solids (%)	13.4 ± 0.7 ⁷	13.5 ± 0.7	13.3	12.6 ± 1.0	12.9 ± 1.1	12.9 ± 1.4	12.3	12.9 ± 0.9	13.5 ± 0.5
Fat (%)	4.3 ± 0.9	4.5 ± 1.0	4.1	3.8 ± 0.9	3.9 ± 1.3	4.3 ± 1.2	3.6	4.1 ± 1.3	4.7 ± 0.5
Protein (%)	3.6 ± 0.2	3.2 ± 0.12	3.6	3.0 ± 0.1	3.0 ± 0.1	3.1 ± 0.2	3.3	3.2 ± 0.2	2.9 ± 0.1
Lactose (%)	5.3 ± 0.5	5.3 ± 0.4	5.0	5.0 ± 0.1	5.0 ± 0.1	4.9 ± 0.15	4.9	4.9 ± 0.02	5.0 ± 0.1

¹ BrSw = Brown Swiss
² Lit Value = Reference values from Kaufmann W and Hagemester (1987) and Walstra and Jenness (1984)
³ Hlstn = Holstein
⁴ 1 and 2 indicate different genetic lines within breed
⁵ H X Jersey = a cross between a Holstein and Jersey, referring to the source of the animal that provided the donor cell for SCNT.
⁶ NP = not provided
⁷ Values are presented as means ± standard deviation.

For 12 of the 14 fatty acids analyzed, no significant differences were noted between clone and non-clone milk. Significant differences ($p < 0.05$) were noted in the amount of palmitic acid (C16:0) and linolenic acid (C18:3) between clones and non-clones. The authors noted that the palmitic and linolenic acid levels for milk from clones and non-clones fall within published references for that substance, and speculated that difference between the levels in clones and non-clones could be attributed to diet. Differences were observed in the fatty acid profiles of the milks over the course of the lactation cycle. These were noted as being consistent with published

¹⁰⁵ Most of the milk in the US is produced by Holstein cows. The values for the individual components that comprise milk the USDA Nutrient Database are therefore expected to be more similar to levels of individual components derived from milk from Holsteins compared to other dairy breeds.

accounts of lactation cycle differences, diet, and seasonality. The greatest variability was observed in the mineral content of milk from clones and non-clones, with significant differences noted for potassium, zinc, strontium, and phosphorus levels. The authors attribute these differences to the different diets that clones and non-clones were fed. (Clones and comparators were housed at different farms, and fed different rations.) The authors' overall conclusion was that there were "no obvious differences between milk from clones and non-clones."

In an abstract, Aoki et al. (2003) described the generation of two clones from cells derived from the colostrum of a Holstein cow, as well as providing summary comments regarding milk characteristics and milking behavior. According to the abstract, milk yield (measured in kilograms per week) was measured every four weeks over a 16 week period. They noted that significant differences were observed between milk yield at weeks 1, 9, 11, and 13, but in the other weeks, "they shared similar lactation curves." Milk composition was apparently measured as milk fat, protein, lactose, solids-non-fat, and total solid percentages. The authors reported that there were "considerable resemblance[s]" between the milk of clones and non-clones. It should be noted, however, that the measurements in this study were made between clones in their first lactation, and comparators in their second lactation. There are often differences in milk yield and composition between successive lactations (Vasconcelos et al. 2004; Flis and Wattiaux 2005).

Wells et al (2004) reported on the composition of milk from six 2-year old Friesian cow clones in their first lactation (Table VI-20). The milk composition was compared to that of the single donor cow in her third lactation. All animals were managed together as part of a single dairy herd. The comparison was made based on a single milk sample take at mid lactation. Although one of the protein levels (bovine serum albumin (BSA (162 ± 6 vs. 105 mg/L)) and two of the fatty acids in the clones (C18:2 (3.76 ± 0.06 vs. 3.00), C18:3 (1.19 ± 0.07 vs. 0.90)) were found to be statistically different ($p > 0.05$) from the donor cow's milk, they were reported to be within normal limits for this breed of cow, and not considered by the authors to be biologically significant. The authors conclude that overall milk composition of the clones was what might be expected for healthy cows.

Component of Milk (g/kg milk unless otherwise stated)	Clones (n=6)	Donor Cow
Fat	35.1 ± 1.00	36.2
Protein	31.2 ± 0.36	31.5
Lactose	50.1 ± 0.36	51.7
α_s -casein	10.8 ± 0.17	10.6
β -casein	8.98 ± 0.15	9.13

κ-casein		2.94 ± 0.09	2.76
α-lactalbumin		0.99 ± 0.12	1.37
β-lactalbumin		4.52 ± 0.10	4.48
BSA (mg/L)		162 ± 6	105
IgG (mg/L)		563 ± 13	536
Fatty acids (% of total)	C4:0	4.59 ± 0.02	4.61
	C6:0	2.45 ± 0.07	2.48
	C8:0	1.32 ± 0.03	1.37
	C10:0	2.14 ± 0.22	2.36
	C12:0	2.32 ± 0.15	2.30
	C14:0	8.79 ± 0.30	8.95
	C16:0	25.9 ± 0.55	25.5
	C16:1	1.01 ± 0.07	1.38
	C17:0	1.77 ± 0.06	1.62
	C18:0	12.4 ± 0.26	11.3
	C18:1	26.5 ± 1.06	28.6
	C18:2	3.76 ± 0.06	3.00
	C18:2 CLA	1.18 ± 0.07	1.05
C18:3	1.19 ± 0.03	0.90	
SFC at 10°C (g/kg fat)		520 ± 15.8	477
Magnesium (mg/100g)		9.3 ± 0.23	10.2
Calcium (mg/100g)		124 ± 1.8	128
Sodium (mg/100g)		33 ± 0.6	33
Potassium (mg/100g)		149 ± 1.5	152

Subsequently, Wells (2005) compared vitamin and mineral composition in milk from 3 clone cows each from 3 clonal families (n=9) and non-clone comparators (n=5, Tables VI-21, 22, and 23). No details were provided for the comparator animals. No differences were reported in selected vitamins in milk. Wells concluded that the composition from these clones was within the normal range for milk.

Table VI-21: Vitamin Composition of Bovine Whole Milk Harvested In Spring (from Wells 2005)			
Vitamin	Units	Clone milk (n = 9)	Comparator milk (n = 5)
A	IU/100 ml	128 ± 22	140 ± 29
B2	mg/100 ml	0.27 ± 0.03	0.24 ± 0.04
B12	µg/100 g	0.40 ± 0.09	0.20 ± 0.07

Table VI-22: Mineral Composition of Bovine Whole Milks Harvested In Spring (from Wells 2005)		
Mineral (mg/100g)	Milk from Clones (n = 9) Mean ± Standard Deviation	Milk from Comparators (n = 5) Mean ± Standard Deviation
Calcium	133.0 ± 15.7	134.4 ± 10.1
Iodine	0.0010 ± 0.0005	0.0022 ± 0.0009
Magnesium	10.1 ± 1.5	10.0 ± 0.0
Phosphorus	115.2 ± 12.5	103.6 ± 5.3
Potassium	129.9 ± 13.9	125.8 ± 15.1
Selenium	0.0005 ± 0.0	0.0008 ± 0.0004
Sodium	27.0 ± 5.1	26.8 ± 5.0
Zinc	0.495 ± 0.768	0.515 ± 0.077

Table VI-23: Amino Acid Composition of Bovine Skim Milk Harvested In Spring (from Wells 2005)		
Amino acid (mg/g)	Milk from Clones (n = 9) Mean ± Standard Deviation	Milk from Comparators (n = 5) Mean ± Standard Deviation
Alanine	1.31 ± 0.17	1.31 ± 0.14
Arginine	1.32 ± 0.20	1.33 ± 0.12
Aspartic acid	3.07 ± 0.40	3.02 ± 0.26
Cystine	0.36 ± 0.05	0.38 ± 0.04
Glutamic acid	8.78 ± 1.16	8.65 ± 0.70
Glycine	0.74 ± 0.11	0.75 ± 0.07
Histidine	1.02 ± 0.14	1.01 ± 0.07
Isoleucine	1.82 ± 0.28	1.76 ± 0.16
Leucine	3.83 ± 0.51	3.75 ± 0.30
Lysine	3.22 ± 0.45	3.16 ± 0.26
Methioine	0.89 ± 0.12	0.88 ± 0.09
Phenylalanine	1.85 ± 0.26	1.83 ± 0.15
Proline	3.87 ± 0.53	3.80 ± 0.33
Serine	2.19 ± 0.30	2.17 ± 0.19
Threonine	1.78 ± 0.25	1.76 ± 0.18
Tryptophan	0.48 ± 0.08	0.48 ± 0.06
Tyrosine	1.81 ± 0.27	1.80 ± 0.17
Valine	2.15 ± 0.32	2.08 ± 0.17
Totals	40 ± 5.57	39.94 ± 3.40

The laboratories at the University of Connecticut continued their surveillance of a set of Holstein clones (see CBSA section) by analyzing the composition of milk from clones (the composition of meat from Japanese Black clones is discussed in the Meat Composition section) (Tian et al. 2005). Ten dairy clones were produced through SCNT using skin fibroblast (n=4) or cumulus cells (n=6) of a 13 year old Holstein cow. Four of the surviving cumulus cell derived clones were compared with four age- and parity-matched comparator heifers. All animals were raised at the same facility from 2 months of age, with the same management and feeding. Both groups were bred by artificial insemination using semen from the same bull at 14-15 months of age.

Milk production was monitored starting immediately after calving through 305 days of lactation; milk samples were collected three times daily. Two milk samples were collected from each of three milkings on a given day of each week throughout the entire first lactation. One of the collected samples was used for the analysis of total protein, total fat, lactose, total solids, milk urea nitrogen, and somatic cell counts as routinely monitored by the Dairy Herd Improvement Association (DHIA) at a DHIA-designated laboratory. Individual fatty acids that were measured included C4:0, 6:0, 8:0, 10:0, 12:0, 14:0, 14:1, 16:0, 16:1, 18:0, 18:1, 18:2, 18:3, and 20:0. The second collected sample was analyzed for protein profiles using denaturing SDS/PAGE (sodium dodecyl (lauryl) sulfate-polyacrylamide gel electrophoresis) stained with Coomassie blue. Relative quantities of each band were determined. Antibody concentrations (IgM, IgA, and IgG) were determined in colostrums from the first milking with a commercial assay.

The investigators report that clones and comparators showed comparable lactation curves, with milk production increasing during the first month, and decreasing thereafter during the course of the lactation. The exception was one clone that birthed prematurely, and produced 30 percent less milk, as would be expected. There were no significant differences between the composition of milk from clone and age-matched, closely related comparators, or breed comparators. The authors also state that the composition of milk from clones was within normal industry standards (www.dhia.org). Analysis of key milk proteins indicated that there were no differences among major or minor bands as analyzed by SDS/PAGE. The four major bands representing α -caseins, β -caseins, κ -caseins, and β -lactoglobulins were consistent in all milk samples whether from clones or their comparators. Similarly, there was no difference between groups for minor protein bands. Antibody concentrations in colostrums were also similar between clone and non-clone cows, and reported to be in the typical range for colostrum antibody composition.

Yonai et al. (2005) (see previous discussions of these animals in the Developmental Nodes) presented milk composition data for six Holstein and four Jersey clone cows (Table VI-24). Overall milk yield, fat, protein, and other solids not fat (SNF) were considered to be normal by the authors, with the observed inter-clone differences and differences from the donor animals attributed to diet and environmental conditions. The authors note that the heritability of milk

production is approximately 30 percent and, considering the impact of environmental conditions on milk production, suggest that standardizing individual feeding conditions may be helpful for future comparisons.

Table VI-24: Milk Composition (mean \pm standard deviation) in First and Second Lactations (adapted from Yonai et al. 2005)							
	Milk Yield	Fat (%)	Fat (kg)	Protein (%)	Protein (kg)	SNF* (%)	SNF (kg)
Jersey (n=4)							
First lactation	5,896.4 \pm 332.0	5.0 \pm 0.2	300.3 \pm 23.9	3.8 \pm 0.2	225.5 \pm 20.4	9.4 \pm 0.2	560.0 \pm 47.8
Donor animal	5,064.0	4.9	242.3	4.0	197.1	9.6	477.2
Second lactation	7,262.8 \pm 222.6	5.13 \pm 0.13	375.3 \pm 26.2	3.78 \pm 0.10	274.8 \pm 13.4	9.35 \pm 0.10	681.3
Donor animal	6,087.0	4.6	280	3.67	224	9.30	566
Holstein (n=6)							
First lactation	9,333.0 \pm 476.4	4.7 \pm 0.1	440.3 \pm 36.7	3.3 \pm 0.1	304.2 \pm 27.6	9.0 \pm 0.1	835.5
Donor animal	10,968.0	4.1	452.0	3.3	359.0	—	—
Second lactation	11,271.4 \pm 1084.7	4.5 \pm 0.2	510.5 \pm 53.4	3.1 \pm 0.1	353.5 \pm 31.4	8.7 \pm 0.1	978.7
Donor animal	11,442.0	3.9	446.2	2.8	320.4	—	—
* SNF is milk solids not fat							

Heyman et al. (2007) studied milk fat composition in five cattle clones and five of their comparators. Milk samples were collected at days 80 and 180 during the first lactation. Comparisons were limited in this study to milk from comparators vs. milk from clones; no comparisons to historical reference values were reported. At both time points, the proportion of stearic acid was lower in the milk from the clones. In addition, two indices of delta-9 desaturase activity, C18:0 and C18:2-*c9-t11*, were higher in the milk from the clones compared to milk from control cows. The proportion of C18:t11 was also lower in clones, but only at the 180 day time point. As all five clones in this study were of the same genotype, the authors speculate that differences in delta 9 desaturase activity may reflect a within breed genotypic difference. Milk composition data from clones of more genotypes are needed to confirm these results.

In the most comprehensive analysis of milk composition from clone cows published to date, Laible et al. (2007) collected milk samples from nine clones (six Friesian and three Jersey) and five sexually-derived cows (3 Friesian and 2 Jersey) during early, mid, and late lactation. Because, in general, milk from Jerseys has higher concentrations of protein, casein and fat compared to milk from Holstein or Friesian cows, the authors compared results within each breed, rather than across all breeds. That is, for each constituent assayed, data were presented for Friesian controls (n=3), Jersey controls (n=2), two lines of Friesian clones (n=3 per line) and Jersey clones (n=2). In addition to comparing milk from clones and non-clones, the authors made comparisons with published reference ranges. The compositional analysis in this study included all of the proximates identified in Table VI-16 as useful for showing no material differences between milk from clone and comparator cows (vitamins, minerals, fatty acid profiles, amino acid profiles, and carbohydrate (lactose)).

Proximate analyses indicated that the ranges of concentrations of fat, protein, casein, lactose, and pH in milk from clones were all within normal ranges for milk from conventional, pasture-fed dairy cattle in New Zealand (Mackle et al. 1997; Mackle et al. 1999; Auldist et al. 2004). The authors state that there were no differences between milk from control and clones in the percentages of 24 fatty acids, or in the total percentages of saturated, monounsaturated or polyunsaturated fatty acids. For each group of clones, mean values for three to six individual fatty acids in milk were outside the respective reference ranges. These observations were not unique to the clones, as values for three individual fatty acids in controls were also outside the reference ranges. Concentrations of minerals (calcium, magnesium, phosphorus, potassium, selenium, sodium and zinc) in milk from clones cows were similar to concentrations in comparator samples and within reference values, with the exception of potassium in Jersey controls (116 ± 10 mg/100 g vs. reference range of 126-173 mg/100g) and selenium in one line of Friesian clones (0.4 ± 0.2 mg/100 g vs. reference range of 0.6-2.0 mg/100 g). For 18 amino acids and vitamins (A, B₂, B₁₂), most of the values (57/90 and 13/15, respectively) were outside the reference ranges, but there were no dramatic differences between values for clones and their breed-matched controls.

In their Discussion, the authors point out that the discrepancies between the observed values and reference ranges may be explained by the small numbers of animals in their study, and that comparison of milk from clones to milk from a wider selection of control animals that represent more genotypes, nutrition and farming systems would put the values from clones within reference ranges for normal bovine milk. Thus from this pilot study, the authors conclude that the composition of milk from the nine clones was “broadly similar” to milk from the five comparator cows.

ii. The Report of the Japanese Research Institute for Animal Science in Biochemistry and Toxicology

The Japanese Research Institute for Animal Science in Biochemistry and Toxicology provided a report entitled “*Investigation on the Attributes of Cloned Bovine Products*” published by the Japan Livestock Technology Association (Japan 2002: Appendix I). CVM was able to obtain a seven page English-language summary translation of the original 489 page Japanese report. Only the English-language summary is reviewed in this risk assessment.

The study investigated blood, milk, and meat constituents in blastomere nuclear transfer clones (BNT) and SCNT cattle clones. No information was provided on the production of the BNT or SCNT clone cattle in the English translation, and the comparator group was identified as “ordinary cattle.” The results for milk composition are discussed in this section; results for meat composition are discussed in the section entitled Compositional Data on Meat from Clones. In addition, the Japanese report includes results of rodent feeding studies conducted with edible products derived from the cattle clones. These and other studies are discussed below in the section entitled Allergenicity and Feeding Studies in Rodents.

Milk constituents were compared between ordinary cattle, BNT clones, and SCNT clones. The results are reported as the mean of samples obtained three and six weeks after parturition and provided in Table VI-25. No biologically significant differences were observed between any of the groups for the parameters tested.

Classification	Cattle No.	Protein	Fats	Sugars	Ash content	Water content	Calcium	Cholesterol
		g/100 g					mg/100 g	
Ordinary cattle	Min. value	3.0	2.2	4.6	0.7	88.1	100	8
	Max value	3.4	3.3	4.6	0.7	89.7	110	10
	Mean value	3.3	2.7	4.6	0.7	88.9	105	9
BNT clones	No.1	2.9	2.3	3.0	0.8	91.1	95	9
	No.2	2.9	3.6	3.5	0.7	89.3	105	9
SCNT clones	No.1	3.1	4.3	4.6	0.7	87.4	120	9
	No.2	3.3	2.6	4.4	0.7	89.1	115	11
	No.3	3.3	3.1	4.5	0.7	88.5	115	10

iii. Summary Statement on Composition of Milk from Clones

Several peer-reviewed studies describe the composition of milk from bovine clones. In addition to gross composition (percent solids, fat, protein, and lactose), some reports include a detailed analysis of fatty acids, vitamins, minerals, and amino acids, and in some cases, comparisons are made with previously published reference values for milk composition. These studies indicate that milk from cow clones is not significantly different in composition from milk from non-clones. Some minor differences have been identified in the composition of milk from clones compared to non-clones or reference values, but in each of these reports, the authors attribute the minor differences to diet, environmental conditions, small numbers of animals, and limited numbers of genotypes, rather than to cloning *per se*. None of these differences, however, indicate the presence of hazards that could pose food consumption risks, as they all fall within published historical values for milk. We therefore find that milk derived from bovine clones does not materially differ from milk from conventionally bred cattle.

d. Characterization of Meat from Clones and Their Progeny

i. Cattle

Peer-reviewed Reports

Two early linked reports on carcass merit¹⁰⁶ (e.g., dressing percentage, fat depth, rib-eye area, yield and quality grade) of cattle produced via BNT have been published (Diles et al. 1996a,b). Neither paper addresses food safety issues. Both papers evaluate the degree to which body measurements are heritable (Diles et al. 1996a), and the degree to which there is phenotypic variability among clones and closely related siblings. The studies conclude that animals derived from BNT provide good models for determining which traits have strong genetic correlations.

The study by Tian and her colleagues at the University of Connecticut discussed previously for milk composition also reports the results of studies on the composition of meat from bovine SCNT clones (Tian et al. 2005). Cultured skin fibroblasts or cumulus cells were used to clone an adult Japanese Black beef bull, selected as a superior breeding stud with superior marbling traits at 17 years of age. Six bull clones were produced; four survived and were apparently normal. The clone bulls were maintained on the same diet and raised in the same facility with eight

¹⁰⁶ Carcass merit programs have been initiated by a group of academics and beef producers to correlate bovine genetics and phenotypic markers for consumer-desired traits such as marbling, tenderness, and composition. Because consumers desire consistency in meat products, producers demonstrating that their herds have good performance and carcass data can leverage higher market prices for their beef. Identification of genetically determined traits can also lead to selective breeding programs that improve herd meat quality in a directed manner.

comparator non-clone animals which were produced by artificial insemination using semen from the son of the original donor bull. The “genetically” matched comparators therefore shared 25 percent of their genetic makeup with the clones. In addition, 20 age-matched sexually reproduced Japanese Black beef cattle were used as breed comparators to establish the normal range for each measured parameter. All bulls were castrated at 3 months of age and raised on standard growing ration from 8 to 26 months of age. The comparators and two of the clones were slaughtered and subjected to standard meat analyses. Variables measured included the following:

- Organ or body part weights
- Total proportion of meat and fat in the dressed carcass
- Cross section of the left dressed carcass between the 6th and 7th rib
- Moisture in six muscles (infraspinatus, longissimus thoracis, latissimus dorsi, adductor, biceps femoris, and semitendinosus)
- Crude protein in six muscles (infraspinatus, longissimus thoracis, latissimus dorsi, adductor, biceps femoris, and semitendinosus)
- Crude fat content in six muscles (infraspinatus, longissimus thoracis, latissimus dorsi, adductor, biceps femoris, and semitendinosus)
- Fatty acid composition (lauric acid, myristic acid, palmitic acid, palmitoleic acid, stearic acid, oleic acid, linoleic acid and linolenic acid) of five major fat tissues (subcutaneous fat, inter-muscular fats, celom fat, and kidney leaf fat)
- Amino acid composition of the longissimus thoracic muscle
- Histopathology of all organs

The 90 percent confidence intervals for each parameter were compared in a paired analysis between the clone and non-clone genetic comparators. There were 12 instances where the clones and genetic comparators showed statistical differences:

- Amount of mesentery fat
- Proportion of longissimus thoracic muscle over body weight
- Muscle moisture
- Amount of crude protein in the semitendinosus muscle
- Amount of linolenic acid in the kidney leaf fat
- Amount of linolenic acid in the longissimus thoracis
- Amount of linolenic acid in the semitendinosus muscles
- Amount of oleic acid in the semitendinosus muscle
- Amount of palmitic acid in the semitendinosus muscle
- Amount of linoleic acid in the semitendinosus muscle

All of the measurements were higher in the clones than in the genetic or breed comparators, except for crude protein or muscle moisture in semitendinosus muscle. The differences in mesentery fat and fatty acid content were attributed to the characteristics of the donor bull (superior marbling). It is noted that the clones had a marbling score of 8 out of 12, compared to an industry standard of 5.2, and genetic comparator score of 6.5. All of the other variables fell within normal industry standards. The researchers conclude that the meat from somatic animal clones falls within normal industry standards and does not significantly differ from those of the genetic or breed comparators. The differences observed were considered due to the superior genetics of the donor bull from which the line of clones was derived. No abnormalities were reported in the pathology or histopathology for tissues collected from the clones.

This group has also reported meat composition in 11 cattle clones and 11 sexually-derived comparators (Yang X et al. 2007b). (Blood chemistry and hematological parameters in these animals have been discussed previously in the Post-pubertal Maturation Developmental Node.) Ages ranged from “>12 months” to 43 months. There were six females and five males in each group representing several breeds (Angus, Brangus, Holstein, Red and White Holstein, and cross breeds). No anatomical or other obvious defects were observed in carcasses after slaughter. Mean values for meat composition, amino acids, fatty acids, vitamins, minerals, cholesterol, and proximate analyses of water, fat, protein and carbohydrate are presented. No differences were detected between clones and comparators for any of the constituents analyzed. In addition to differences between mean values, the authors also analyzed the standard deviations of the means and found that for essentially all traits examined, standard deviations were similar between clones and non-clones. Moreover, statistical outliers were no more likely to occur in the clone group. This finding indicates that variability in meat composition in beef cattle clones is not different from that in sexually-derived comparators.

Citing the fact that meat quality is influenced by both contractile and metabolic characteristics of muscle, Heyman et al. (2007) evaluated muscle characteristics in nine heifer clones and eight comparator heifers. Samples of semitendinosus muscle were obtained by biopsy at 8, 12, 18 and 24 months of age¹⁰⁷. The findings of this study are summarized textually; no actual data are provided. At eight and 12 months, the proportions of myosin heavy chain (MyHC) I (a slow oxidative isoform of a key muscle protein), and MyHC IIa (a fast oxido-glycolytic isoform of the same key muscle protein) were significantly higher in clones compared to comparators, while the proportion of MyHC IIb (another fast glycolytic isoform) was lower in clones. At the same time points, oxidative metabolism, measured by isocitrate dehydrogenase activity (ICDH), was significantly higher in clones compared to comparators. Glycolytic metabolism (the mechanism

¹⁰⁷ Only a summary of results is presented in this report; no actual data are shown. Many of these results were published in INRA reports in 2004 and 2006. Because these reports are not readily available via PubMed, the results are also discussed here.

by which muscles break down stored carbohydrates), measured by lactate dehydrogenase (LDH) activity, was similar between clones and controls. No differences in the proportions of myosin heavy chains, ICDH, or LDH were found between clones and controls at 18 or 24 months. The authors interpreted these findings as evidence that clones had slower muscles associated with more oxidative metabolism compared to the comparators, and that the contractile and metabolic profile of muscle from clones corresponded to a delay in muscle differentiation at 8 and 12 months of age. This conclusion is based solely on biochemical data; no corresponding histological or physiological studies are reported.

In the same muscle samples used to evaluate contractile and metabolic parameters, Heyman et al. (2007) also determined the fatty acid composition. At eight months of age, clones had a higher proportion of polyunsaturated fatty acid (37.2 percent vs. 28.4 percent in controls) and a lower proportion of monounsaturated fatty acid (18.7 percent vs. 24.7 percent in controls). These differences may be related to the lower marbling in meat from clones compared to controls. At 24 months of age, clones had lower proportions of stearic acid, C18:1-*t*11 and C18:1-*c*11 than controls. At 18 and 24 months of age, clones had higher indices of delta-9 desaturase activity, but only for the C:16 index.

It is interesting that the alterations in fatty acid composition and delta-9 desaturase activity in meat from clones were also observed in milk from clones (Heyman et al., 2007; see section on Characterization of Milk from Cow Clones). Taken together, these results support the authors' conclusion that lipid metabolism may be altered in their population of cloned cattle. It is important to note, however, that comparisons in these studies were limited to clones vs. comparators ($n = 5$ to 9 per group), and no comparisons were made with historical reference values for either milk or meat. Clearly, similar studies using larger numbers of animals representing multiple genotypes are needed to verify these results.

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Table VI-26: Meat Constituents in BNT and SCNT Clones and Ordinary Cattle (from Japan 2002)							
Classification	Cattle No.	Protein (g/100 g)	Fats (g/100 g)	Sugars (g/100 g)	Ash content (g/100 g)	Water content (g/100 g)	Cholesterol (mg/100 g)
Ordinary cattle	Min. value	17.8	13.8	0.4	0.9	58.0	50
	Max value	19.6	22.9	0.8	1.0	64.8	68
	Mean value	18.4	19.3	0.6	0.9	60.8	59
BNT clones		17.4	21.2	0.4	0.9	60.2	56
SCNT clones		16.8	23.8	0.5	0.9	57.9	68

As discussed in the section on milk composition, the Japanese Research Institute for Animal Science in Biochemistry and Toxicology provided an unpublished bound report “Investigation on the Attributes of Cloned Bovine Products” by the Japan Livestock Technology Association (Japan 2002).¹⁰⁸ The results for meat are discussed in this section. Takahashi and Ito (2004) have published a summary of these data, including some information characterizing the clones and their comparators. SCNT and BNT clones were derived from Japanese Black cattle at the Nara Prefectural Animal Research Center. Comparator animals were selected as conventionally bred Japanese Black cattle. All animals used for compositional analysis were sacrificed between 27 and 28 months of age, after fattening.

Meat constituents were compared between ordinary cattle, BNT clone cattle, and SCNT clone cattle. The results are reported as the mean of analytical samples obtained from 9 sites; shoulder, chuck loin, rib loin, loin end, brisket, round, silver side, rump, and tender loin, and are provided in Table VI-26.

No biologically significant differences were observed between any of the groups of cattle (ordinary cattle, BNT clone cattle, and SCNT clone cattle) for the parameters tested.

Cyagra dataset

Cyagra, the cloning company that provided the extensive physiological data discussed earlier in the risk assessment, also provided meat composition data. Eleven clones (6 female, 15 to 43 months; 5 male, 12 to 17 months) and an equal number of comparator cattle (over 12 months) were selected for the study. All animals were fed a standard ration for 30 days prior to slaughter. Samples (500 g each) were obtained of chuck arm roast, bottom sirloin tip roast, and short loin for analysis by an independent laboratory.

No biologically significant differences are observed in the composition of meat from clones and comparators. The results of the compositional analysis summarized across gender and cuts of meat are summarized in Table VI-27. A detailed presentation of the results is provided in Appendix E, the Cyagra Dataset.

¹⁰⁸ Some of these data are presented in Takahashi and Ito 2004; however, we have cited the original report as the data reporting is more complete.

Table VI-27: Meat Composition from Cyagra Clones and Comparators					
Meat Analysis		Overall Comparison			
Sample Number					
Marked ID		Clone		Comparator	
Analyte	Units	Mean	Std dev	Mean	Std dev
Crude Fat	%	11.62	10.08	8.62	8.10
Moisture	%	66.18	7.68	68.57	5.51
Protein – Combustion	%	20.69	2.96	21.72	2.58
Protein – Kjeldahl	%	20.74	2.90	21.58	2.51
Ash	%	1.03	0.17	1.05	0.13
Balance (protein+moist+ash+fat)	%	99.56	1.72	99.82	0.89
Amino Acid Profile (results below)					
Tryptophan	%	0.25	0.03	0.26	0.03
Aspartic Acid	%	1.96	0.31	2.08	0.23
Threonine	%	0.93	0.15	1.01	0.12
Serine	%	0.79	0.14	0.86	0.12
Glutamic Acid	%	3.22	0.54	3.33	0.71
Proline	%	0.97	0.21	0.91	0.16
Glycine	%	1.08	0.27	1.08	0.21
Alanine	%	1.28	0.21	1.36	0.18
Cystine	%	0.23	0.04	0.24	0.04
Valine	%	0.89	0.21	1.07	0.14
Methionine	%	0.54	0.09	0.56	0.08
Isoleucine	%	0.81	0.20	0.98	0.12
Leucine	%	1.61	0.27	1.78	0.20
Tyrosine	%	0.69	0.11	0.74	0.08
Phenylalanine	%	0.84	0.14	0.91	0.10
Histidine	%	0.70	0.12	0.77	0.11
Lysine, Total	%	1.77	0.31	1.98	0.23
Arginine	%	1.33	0.23	1.41	0.17
Hydroxyproline	%	0.17	0.07	0.16	0.07
Fatty Acid (results below)					
C14:0 Tetradecanoic (Myristic)	%	0.28	0.24	0.23	0.24
C14:1 Tetradecenoic (Myristoleic)	%	0.15	0.17	0.09	0.10
C15:0 Pentadecanoic	%	0.04	0.04	0.04	0.04

C15:1 Pentadecenoic	%	0.00	0.00	0.00	0.00
C16:0 Hexadecanoic (Palmitic)	%	2.65	2.29	2.04	2.00
C16:1 Hexadecenoic (Palmitoleic)	%	0.69	0.68	0.45	0.44
C16:2 Hexadecadienoic	%	0.08	0.08	0.06	0.06
C17:0 Heptadecanoic (Margaric)	%	0.10	0.08	0.09	0.09
C17:1 Heptadecenoic Margaroleic	%	0.11	0.11	0.08	0.08
C18:0 Octadecanoic (Stearic)	%	1.18	0.93	1.05	1.10
C18:1 Octadecenoic (Oleic)	%	4.94	4.49	3.43	3.34
C18:2 Octadecadienoic (Linoleic)	%	0.31	0.24	0.22	0.18
C18:3 Octadecatrienoic (Linolenic)	%	0.05	0.05	0.05	0.05
C18:4 Octadecatetraenoic	%	0.07	0.08	0.04	0.04
C20:1 Eicosenoic (Gadoleic)	%	0.03	0.04	0.02	0.03
C20:4 Eicosatetraenoic (Arachidonic)	%	0.01	0.01	0.01	0.01
Total Monounsat. Fatty Acids Calc.	%	5.92	5.42	4.08	3.93
Total Polyunsat. Fatty Acids Calc.	%	0.54	0.44	0.40	0.33
Saturated Fatty Acids	%	4.25	3.51	3.47	3.42
Total Fat (as triglycerides)	%	11.24	9.76	8.34	7.99
Calcium	mg/100g	12.01	13.78	14.30	13.67
Iron	mg/100 g	2.29	0.74	2.32	0.71
Phosphorus	mg/100 g	179.09	27.31	191.21	28.12
Zinc	mg/100 g	3.86	0.67	4.14	0.64
Cholesterol	mg/100g	64.92	7.79	68.43	8.64
Niacin	mg/100 g	4.96	1.18	5.00	1.07
Vitamin B1 - Thiamine Hydrochloride	mg/100 g	0.10	0.04	0.09	0.02
Vitamin B2 – Riboflavin	mg/100 g	0.24	0.05	0.29	0.04
Vitamin B6	mg/100 g	0.33	0.08	0.37	0.11
Vitamin E	IU/100g	0.50	0.15	0.44	0.15
Hydroxyproline	%	0.17	0.07	0.16	0.07

ii. Swine
(a) Clones

ViaGen, Inc., worked in consultation with CVM to design two studies comparing the composition of meat from swine clones vs. age-matched, genetically related, AI-derived comparator animals. Experimental design, raw data, and CVM's analysis of the data are provided

in Appendix F, The ViaGen Dataset. Meat composition data were available for five clones (four Hamline and one Duroc) and 15 comparator animals (all Hamline). There were no differences between the Duroc and Hampshire clones, so data for clones were pooled.

Carcass characteristics were provided on four Hamline clones and 15 comparator barrows and are summarized in Table VI-28. The Duroc clone barrow carcass was condemned at slaughter due to a lung adhesion, and thus data relating to growth and carcass characteristics were not included for these parameters. In some cases of lung adhesions due to bacterial infection, animals fail to thrive, thereby affecting their growth rate and carcass characteristics – this was considered to be the case for the Duroc clone. Two other clones were approximately 100 pounds lighter than any of the other animals in the experiment at the time of slaughter, and for this reason were excluded from carcass evaluation. Hot carcass weights averaged 189.0 and 199.5 pounds for clone and comparator barrows, respectively. Carcass lengths were 82.4 and 84.5 cm for clones and comparators, respectively. Dressing percentages were 70.1 and 70.2 percent for clones and comparators, and were similar across groups. Backfat thickness over the first rib, tenth rib, last rib, and lumbar vertebra were slightly greater for comparator barrows than for clone barrows which may, in part, be due to the heavier body weight of comparator barrows at the time of slaughter.

Qualitative characteristics including USDA carcass muscle score, color, firmness, and marbling were similar across breeding regimens and are illustrated in Table VI-29. All animals received score 2 for carcass muscle. All of the clone and comparator barrows had marbling scores of either 1 or 2.

Table VI-28: Comparison of the Carcass Characteristics of Barrows Derived by Somatic Cell Nuclear Transfer (Clones) or Conventional Breeding (Mean ± standard deviation) <i>(from ViaGen, Inc.)</i>		
	Clones (n=4)	Conventionally Bred (n=15)
Hot Carcass Weight (lbs)	189.0 ± 13.8	199.5 ± 13.7
Carcass Length (cm)	82.4 ± 1.5	84.5 ± 2.7
Dressing Percentage (%)	70.1 ± 0.8	70.2 ± 1.4
Back fat Thickness (mm)		
First Rib	35.3 ± 2.1 ^b	38.7 ± 3.1 ^a
Tenth Rib	18.5 ± 3.1	22.2 ± 4.9
Last Rib	20.5 ± 4.7	23.3 ± 3.4
Last Lumbar	17.3 ± 3.2	21.0 ± 3.1
Loin Eye Area (cm ²)	44.0 ± 4.4	45.8 ± 4.0

Measurements of pH at 24 hours post-slaughter on the longissimus muscle were similar. Loin eye area for meat cuts for clone and comparator barrows were only slightly different at 45.8±4.0

and 44.0 ± 4.4 inches, respectively. The Hunter L*, a* and b* values were only slightly different between the groups of animals with the meat from clones being slightly darker and more red than meat from comparator barrows.

	Clones (n=4)	Conventionally Bred (n=15)
Longissimus pH at 24 hours	5.6 \pm 0.1	5.7 \pm 0.1
Carcass Muscle Score	2.0 \pm 0.0	2.2 \pm 0.40
NPPC Quality Scores		
Color	3.0 \pm 0.0	2.7 \pm 0.6
Marbling	1.5 \pm 0.6	1.9 \pm 0.5
Firmness	3.5 \pm 0.6	2.9 \pm 0.9
Hunter Color		
L*	52.2 \pm 2.0	56.3 \pm 4.4
a*	9.5 \pm 1.4	7.6 \pm 1.2
b*	17.6 \pm 0.7	16.9 \pm 1.2

Meat composition data were available for five clones (four Hamline and one Duroc) and 15 comparator animals (all Hamline). There were no differences between the Duroc and Hamline clones, so data for clones were pooled. Means \pm standard deviations for fatty acids, amino acids, cholesterol, minerals and vitamins measured are presented in Table VI-30. Differences in individual analytes for clones and comparators were very small and not biologically relevant. Values for niacin and vitamin B₁₂ in both clones and control swine were above USDA values for a similar type of swine muscle (shoulder blade and loin). Values for cholesterol and vitamin B₆ were similar to the USDA values.

Carcass qualitative characteristics were similar for clones and comparators. Differences in backfat thickness and marbling may be due to the lighter weight of clones at slaughter vs. comparators. Differences in meat nutrient composition were very small and likely not biologically relevant. No biologically relevant differences were observed in the food composition values between muscle of swine clones and comparators.

Table VI-30: Compositional Analysis of Meat from Swine Clones¹
(from ViaGen, Inc.)

Component	Clones	Comparators
<i>Amino acids (g)</i>		
Alanine	1.26 ± 0.04	1.30 ± 0.04
Arginine	1.41 ± 0.03	1.47 ± 0.04
Aspartate	2.55 ± 0.28	2.43 ± 0.19
Cystine	0.25 ± 0.03	0.26 ± 0.02
Glutamate	3.41 ± 0.11	3.46 ± 0.09
Glycine	0.98 ± 0.04	1.02 ± 0.10
Histidine	0.99 ± 0.05	1.03 ± 0.05
Isoleucine	1.04 ± 0.05	1.05 ± 0.03
Leucine	1.74 ± 0.05	1.79 ± 0.04
Lysine	1.91 ± 0.06	1.96 ± 0.04
Methionine	0.54 ± 0.06	0.58 ± 0.03
Phenylalanine	0.86 ± 0.02	0.89 ± 0.02
Proline	0.85 ± 0.03	0.90 ± 0.06
Serine	0.90 ± 0.03	0.92 ± 0.02
Threonine	1.11 ± 0.04	1.14 ± 0.03
Tyrosine	0.77 ± 0.02	0.79 ± 0.02
Valine	1.10 ± 0.05	1.12 ± 0.04
<i>Fatty Acids² (g)</i>		
14:0	0.09 ± 0.06	0.05 ± 0.03
16:0	1.31 ± 0.82	0.95 ± 0.49
16:1	0.09 ± 0.04	0.14 ± 0.05
17:0	0.01 ± 0.01	0.00 ± 0.01
17:1	0.01 ± 0.01	0.00 ± 0.01
18:0	0.66 ± 0.41	0.55 ± 0.27
18:1	1.84 ± 0.84	1.49 ± 0.50
18:2	0.26 ± 0.08	0.19 ± 0.06
18:3	0.01 ± 0.01	0.00 ± 0.01
20:0	0.01 ± 0.01	0.00 ± 0.01
20:1	0.05 ± 0.03	0.04 ± 0.02
20:2	0.01 ± 0.01	0.01 ± 0.01
22:6	0.02 ± 0.03	0.01 ± 0.01
Cholesterol (mg)	55.5 ± 6.95	52.81 ± 2.69
<i>Minerals (g)</i>		
Calcium	0.004 ± 0.000	0.005 ± 0.003
Phosphorus	0.20 ± 0.01	0.21 ± 0.01
Iron	0.001 ± 0.000	0.001 ± 0.001
Zinc	0.002 ± 0.000	0.001 ± 0.000
<i>Vitamins</i>		
Niacin (mg)	10.90 ± 0.83	11.16 ± 1.58
B ₆ (mg)	0.41 ± 0.09	0.48 ± 0.12
B ₁₂ (mcg)	0.21 ± 0.28	0.00 ± 0.00

¹Data expressed as quantities per 100 g of homogenized meat.

²Data presented reflect those fatty acids with detectable levels in pork.

(b) Swine Clone Progeny

ViaGen also provided CVM with data from a study comparing the quality and composition of meat derived from the progeny of swine clones and sexually-derived swine. The data from this study, which was performed at USDA's Meat Animal Research Center, Clay Center, Nebraska, have subsequently published in the peer-reviewed literature (Walker et al. 2007). Sexually derived offspring of clones (n=242) were generated by artificially inseminating conventionally bred gilts with semen from four sires that were clones. Control offspring (n=162) were produced by artificially inseminating conventionally bred gilts with semen from three conventionally bred boars. Litters (n=61) of offspring of clones and control offspring were reared using the same management procedure. Pigs were slaughtered when their body weight reached approximately 123 kg. In addition to carcass characteristics, loin samples were collected and assayed for 58 different nutrients including numerous amino acids and fatty acids, vitamins (B₁₂ and B₆, niacin) and minerals (iron, phosphorus and zinc).

Carcass characteristics for the progeny of clones and their comparators are provided in Table VI-31, and discussed in more detail in Appendix F.

Table VI-31: Carcass Characteristics for Progeny Derived from Clone Boars or Conventionally Bred Boars (Means ± standard deviation) (from ViaGen dataset, Appendix F)				
	Hampshire Comparator	Hampshire Clone	Duroc Comparator	Duroc Clone
Hot Carcass Weight (lbs)	176.2 ± 8.6	175.0 ± 8.7	173.9 ± 9.5	179.0 ± 9.1
Carcass Length (cm)	82.7 ± 2.2	81.6 ± 2.1	82.3 ± 2.2	81.5 ± 2.3
Loin Eye Area (cm ²)	6.7 ± 0.8	6.8 ± 0.8	6.6 ± 0.8	7.2 ± 0.9
Back fat Thickness (mm)				
First rib	22.2 ± 4.2	23.4 ± 4.4	23.8 ± 4.1	25.9 ± 4.2
Last rib	16.0 ± 2.9	16.9 ± 3.2	17.4 ± 2.4	19.0 ± 2.8
Last Lumbar	16.6 ± 3.4	17.0 ± 3.2	18.1 ± 2.6	19.3 ± 2.7
Longissimus pH at 24 hours	5.8 ± 0.2	5.7 ± 0.1	5.7 ± 0.1	5.7 ± 0.1
Carcass muscle score	3 ± 0	3 ± 0	3 ± 0	3 ± 0
NPPC Quality Scores				
Color	3 ± 0.3	3 ± 0.2	3 ± 0.1	3 ± 0
Marbling	3 ± 0.7	3 ± 0.8	3 ± 0.8	3 ± 0.9
Firmness	2 ± 0	2 ± 0	2 ± 0	2 ± 0
Hunter Color				
L*	55.54 ± 2.1	55.88 ± 2.4	56.40 ± 2.4	57.24 ± 2.4
a*	7.47 ± 0.9	7.58 ± 1.0	7.21 ± 1.0	7.17 ± 1.0
b*	13.88 ± 0.9	14.12 ± 0.9	13.88 ± 0.8	14.35 ± 0.6

Although some minor differences were observed in backfat thickness and meat color in clone progeny vs. comparators, none of these differences pose hazards that could affect food consumption risks. In addition, none of these differences would have any impact on the quality of the meat.

Table VI-32 provides the comparison of key nutrients between the progeny of clones and their comparators. Data were reported for 412 swine, of which 242 were the progeny of clones and 162 were the progeny of comparator boars. In total, 404 loin samples were analyzed, generating 23,432 data points. Of the 14,036 data points for offspring of clones, 29 values (0.2 percent) fell outside the respective range for controls. Twenty-eight of the samples corresponding to these values were thus re-assayed. (One sample was not re-assayed due to an oversight.) Following the second assay, only two data points exceeded the comparator range by more than 10 percent. One of these data points was for C18:1 octadecanoic (oleic) acid, a monounsaturated omega-9 fatty acid found in various animal and vegetable sources. The measured values for this parameter (5.44 percent and 5.66 percent) were well within the range for pork loin listed in the USDA National Nutrient Database (0.13 percent to 23.32 percent). In the sample that inadvertently omitted from re-assay, the value for threonine was 0.75 percent compared to 0.83-1.60 percent in the comparators, which also fell within the range listed in the USDA National Nutrient Database for pork (0.27 percent to 4.58 percent threonine). The only outlier in the entire data set was for C20:2 eicosadienoic acid. Concentrations of this analyte in a single sample were 0.04 percent and 0.06 percent, compared to 0.01 percent-0.03 percent in the comparators. No reference range was found for C20:2 eicosadienoic acid in pork. These results clearly demonstrate, that based on a large number of samples and composition-specific analytes that pork from the offspring of swine clones does not differ materially from pork from the offspring of conventionally bred swine (Walker et al., 2007).

Table VI-32: Comparison of Nutrient Concentrations in Meat from Progeny of Swine Clones and Comparators (from ViaGen dataset, Appendix F, and Walker et al. 2007)		
Nutrients¹	Progeny from Clone Boars mean \pm std. dev.	Progeny from Comparators Boars Mean \pm std. dev.
Amino Acids		
Aspartic acid	2.31 \pm 0.19	2.29 \pm 0.16
Cystine	0.25 \pm 0.02	0.25 \pm 0.01
Glutamic acid	3.76 \pm 0.34	3.71 \pm 0.27
Glycine	1.14 \pm 0.15	1.12 \pm 0.13
Histidine	0.98 \pm 0.09	0.98 \pm 0.07
Isoleucine	1.03 \pm 0.12	1.03 \pm 0.10
Leucine	1.90 \pm 0.14	1.89 \pm 0.12
Lysine	2.06 \pm 0.17	2.07 \pm 0.16
Methionine	0.61 \pm 0.05	0.62 \pm 0.04
Phenylalanine	0.96 \pm 0.09	0.94 \pm 0.08
Praline	1.09 \pm 0.13	1.11 \pm 0.13
Serine	0.96 \pm 0.08	0.95 \pm 0.07
Threonine	1.09 \pm 0.09	1.08 \pm 0.07
Tyrosine	0.81 \pm 0.06	0.81 \pm 0.05
Valine	1.09 \pm 0.12	1.10 \pm 0.10
Fatty Acids and Cholesterol		
8:0 (Caprylic acid)	<0.01 ²	0.01
10:0 (Capric acid)	0.01 \pm 0.002	0.01 \pm 0.002
11:0	<0.01	<0.01
12:0 (Lauric acid)	0.01 \pm 0	0.01 \pm 0
14:0 (Myristic acid)	0.08 \pm 0.027	0.08 \pm 0.029
14:1 (Myristoleic acid)	<0.01	<0.01
15:0	<0.01	<0.01
15:1	<0.01	<0.01
16:0 (Palmitic acid)	1.39 \pm 0.38	1.40 \pm 0.49
16:1 (Palmitoleic acid)	0.17 \pm 0.06	0.16 \pm 0.05
17:0 (Margaric acid)	0.01 \pm 0.003	0.01 \pm 0.002
17:1 (Margaroleic acid)	0.01 \pm 0.003	0.01 \pm 0.002
18:0 (Stearic acid)	0.66 \pm 0.24	0.68 \pm 0.25
18:1 (Oleic acid)	2.26 \pm 0.76	2.20 \pm 0.72
18:2 (Linoleic acid)	0.3 \pm 0.11	0.29 \pm 0.11
18:3 (Linolenic acid)	0.02 \pm 0.001	0.01 \pm 0.005
18:4	0.01 \pm 0.0001	0.01 \pm 0.004

Table VI-32: Comparison of Nutrient Concentrations in Meat from Progeny of Swine Clones and Comparators (from ViaGen dataset, Appendix F, and Walker et al. 2007)		
20:0 (Arachidic acid)	0.01±0.005	0.01±0.005
20:1 (Gadoleic acid)	0.08±0.04	0.07±0.04
20:2 (Eicosadienoic acid)	0.02±0.01	0.02±0.005
20:3 (Eicosatrienoic acid)	0.01±0.01	<0.01
20:4 (Arachidonic acid)	0.01±0.003	0.01±0
20:5 (Eicosapentaenoic acid)	0.01 ± 0	0.01±0.004
21:5 (Heneicosapentaenoic acid)	0.01±0	<0.01
22:0 (Behenic acid)	<0.01	<0.01
22:1 (Erucic acid)	0.01±0.006	0.02±0.006
22:2 (Docosadienoic acid)	<0.01	0.01±0.01
22:3 (Docosatrienoic acid)	<0.01	<0.01
22:4 (Docosatetraenoic acid)	<0.01	<0.01
22:5 (Docosapentaenoic acid)	<0.01	<0.01
22:6 (Docosahexaenoic acid)	0.02±0.01	0.02±0.01
24:0 (Lignoceric acid)	<0.01	<0.01
24:1 (Nervonic acid)	<0.01	<0.01
Cholesterol (mg/100 g)	57.93±5.46	59.39±5.04
Minerals		
Calcium	0.01±0.003	0.01±0.002
Iron	0.00±0.0005	0.000±0.003
Phosphorus	0.18±0.082	0.16±0.082
Zinc	0.00±0.0003	0.00±0.0001
Vitamins		
Niacin (mg/100g)	10.68±1.23	10.64±1.03
Vitamin B ₆ (mg/100 g)	0.40±0.07	0.38±0.07
Vitamin B ₁₂ (mcg/100 g)	1.01±0.25	0.97±0.28
¹ Unless otherwise specified, quantities are expressed as g/100g homogenized meat.		
² Values marked with “<” indicate concentrations below the level of detection for the instrument used in the assay.		

In a study of the Jin Hua breed, Shibata et al. (2006) described carcass traits and meat quality of the offspring of cloned females. Sires were sexually produced (non-clone) Jin Hua boars. Forty-four offspring (23 male and 21 female) obtained from six litters produced by six dams were slaughtered at 70 kg body weight. Compared to non-cloned Jin Hua controls, offspring of cloned dams had shorter back and loin length (54.1 ± 0.3 cm vs. $56.0 \pm .3$ cm in controls), lower weight ratio of loin bacon (40.6 ± 0.2 percent vs. 41.9 ± 0.4 percent in controls) and higher weight ratio of ham (27.9 ± 0.3 percent vs. 26.5 ± 0.2 percent in controls). There were no differences between offspring of clones and control pigs in back fat thickness or loin area. Analysis of meat quality

indicated that meat from offspring of clones had a lower pH (5.47 ± 0.01 vs. 5.67 ± 0.03 in controls) and greater cooking loss (26.8 ± 0.3 percent vs. 24.9 ± 0.4 percent in controls). Water content, fat content, weep loss and shear value of meat from offspring of clones were not different from control values. The authors concluded that most of the meat quality characteristics of the Jin Hua breed were retained in the progeny of Jin Hua dam clones.

iii. Conclusions from Studies Evaluating the Composition of Meat and Milk from Clones and Their Progeny

The second prong of our Risk Assessment is based on the hypothesis that food products from healthy animal clones and their progeny that are not materially different from corresponding products from conventional animals are as safe to eat as their conventional counterparts. CVM has reviewed several peer-reviewed publications that have evaluated gross (e.g., milk yield, carcass characteristics) and fine (e.g., individual amino acid and fatty acid components) characteristics of meat and/milk from clones, and in two studies, their sexually-reproduced progeny. All but one of these studies indicate that none of the characteristics that we examined differed in any biologically significant way between the clone and comparator. The only exception is a preliminary study in bovine clones which provides evidence that lipid metabolism may be altered in clones, resulting in slight alterations in the fatty acid composition of milk and meat. However, without a comparison of these data to historical reference values, it is unclear whether these differences are representative of all bovine clones or are specific to the limited number of genotypes used in the study.

For swine clone progeny, a comprehensive, peer-reviewed analysis of meat from a large number of animals provides strong evidence that there are no compositional differences between meat from swine clones and meat from conventional swine, and that meat from clone progeny and their comparators is not materially different.

Therefore, in this prong of the Risk Assessment, CVM concludes that the weight of evidence indicates that meat and milk from clones and their progeny do not differ materially from meat and milk derived from their conventional counterparts, and therefore, based on compositional analysis, do not pose any additional food consumption risks compared with meat and milk from conventionally bred animals.

iv. Allergenicity and Feeding Studies in Rodents

The Report of the Japanese Research Institute for Animal Science in Biochemistry and Toxicology

In addition to data on composition of milk and meat from clones, the report of the Japanese Research Institute for Animal Science in Biochemistry and Toxicology (Japan 2002) included the results of studies on the allergenic potential of milk and meat derived from cattle clones as well as rodent feeding studies conducted with these products.

The ability to digest a protein is one index of potential allergenicity; a protein that is less digestible may be more likely to provide an allergenic response. The protein digestion rate of freeze-dried milk combined in feed consumed by rats is reported below for milk obtained from ordinary cattle, blastomere nuclear transfer (BNT) clone cattle, and SCNT clone cattle. The authors report that there was no biological difference among the groups tested.

Table VI-33: Protein Digestion of Freeze Dried Milk from BNT and SCNT Clones and “Ordinary” Cattle (from Japan 2002)		
Test Group	Number of Animals	Digestion Rate (mean ± standard deviation)
Ordinary Cattle	5	83.0 ± 2.6
BNT clone cattle	5	82.7 ± 2.0
SCNT clone cattle	5	8.13 ± 3.4

In a separate study, mice were sensitized by intraperitoneal injection to extracts of milk from clone and non-clone cows. Fourteen days later, the abdominal wall of the mice was surgically retracted and an allergic reaction induced by re-injection of the freeze-dried milk extract into the abdominal wall. Control mice did not receive the second injection of milk extract. Allergenic response was assessed based on vascular permeability as measured by the diameter of dye leakage from the site of injection. No statistically significant differences in allergenic activity were reported between groups. The data are presented in Table VI-34.

Table VI-34: Allergenic Response to Milk from BNT and SCNT Clones and Ordinary Cattle (from Japan 2002)			
Test Group	Mouse Group	Number of Animals	Diameter of dye leakage (mm) (mean ± standard deviation)
Ordinary Cattle	Control Group	7	7.0 ± 3.7
	Test Group	10	18.0 ± 2.9
BNT clone cattle	Control Group	7	4.7 ± 3.2
	Test Group	10	18.0 ± 3.9
SCNT clone cattle	Control Group	7	4.9 ± 4.6
	Test Group	10	17.9 ± 4.2

Based on these two studies, the authors conclude that there were no biologically or statistically significant differences in the allergenic potential of milk from ordinary cattle or BNT or SCNT clones.

To address possible allergenic potential of meat from clone cows, the Japanese researchers compared protein digestion rates with artificial digestive juice and in a rat model, and looked for an allergenic response following direct challenge in rats. For the *in vitro* digestion test, samples were taken from a one-day old conventional calf and a four day old clone. The rates of digestion by artificial digestive juices (artificial gastric juice and artificial intestinal juice) were compared for freeze dried meat derived from ordinary cattle, BNT clone cattle, or SCNT clone cattle (Table VI-35). No information is provided in the translation regarding the artificial digestive material. The results are presented below as the rate of protein digestion.

Table VI-35: Rates of <i>in vitro</i> Digestion of Beef from SCNT Clones or Ordinary Cattle (from Japan 2002)								
Digestive juice	Sample	Rate of digestion after the start of incubation (per cent)						
		Course	Start	0.75 hr	1.5 hr	3 hr	6 hr	12 hr
Artificial gastric juice	Ordinary beef		0	68	79	-	95	90
	Somatic cloned beef		0	59	78	-	91	90
Artificial intestinal juice	Ordinary beef		0	-	20	40	66	67
	Somatic cloned beef		0	-	28	38	67	63

It was concluded that there were no biologically significant differences in the rates of digestion for meat from ordinary beef cattle or from clone beef cattle using artificial digestive juices.

The protein digestion rate of freeze-dried meat combined in feed consumed by rats is in Table VI-36. The authors report that there was no biological difference among the groups tested.

Table VI-36: Protein digestion rate in rats following consumption of freeze dried meat from clone cattle and non-clone cattle (from Japan 2002)		
Test Group	Number of Animals	Digestion Rate (mean±s.d.)
Ordinary cattle beef	5	83.8 ± 6.6
BNT clones	5	82.3 ± 4.7
SCNT clones	5	84.9 ± 3.6

In a separate study, mice were given sensitizing intraperitoneal injections of extracts of freeze-dried beef from clone and non-clone cows. Fourteen days later, the abdominal wall of the mice was surgically exposed and an allergic reaction induced by re-injection of the freeze-dried beef extract into the abdominal wall and administered a vascular dye. Control mice did not receive the second injection of beef extract and only were administered the dye. Allergenic response was assessed based on vascular permeability as measured by the diameter of dye leakage. No statistically significant difference in allergenic activity was reported between groups. The data are presented in Table VI-37.

Test Group	Mouse Group	Number of Animals	Diameter of dye leakage (mm) (mean±s.d.)
Ordinary cattle	Control group	7	5.3 ± 5.0
	Test group	10	13.0 ± 5.9
BNT clones	Control group	7	7.0 ± 4.9
	Test group	10	12.5 ± 3.5
SCNT clones	Control group	7	5.7 ± 4.2
	Test group	10	13.1 ± 5.0

The authors conclude that there were no biologically or statistically significant differences in the allergenic potential of meat from ordinary cattle or BNT or SCNT clone cattle.

The potential for milk and meat from BNT and SCNT clone cattle to cause clastogenic¹⁰⁹ (DNA breaking) events was assessed using an *in vivo* mouse micronucleus assay. Mice were fed milk or freeze dried powdered beef from ordinary cattle, BNT clone cattle, or SCNT clone cattle at 0, 2.5, 5, or 10 percent of the diet for 14 days. In addition, a positive control group received a single intraperitoneal injection of 2 mg/kg mitomycin C, a known clastogen. The positive control group showed a statistically significant increase in the incidence of micronuclei appearance and polychromatic erythrocyte rate, and was considered a positive test. No milk- or meat-fed group, whether derived from ordinary or clone cattle, caused mutations in this assay (i.e., no group fed beef derived from ordinary cattle or clone cattle was positive in this assay for mutagenicity or clastogenicity). The report concludes that there were no biologically significant differences in the component analysis or the results of feeding milk or meat from ordinary cattle, BNT clones, or SCNT clones.

¹⁰⁹ Clastogens are often referred to as mutagens, as most DNA breaks result in mutations if they do not first kill the cell or organism. The mouse micronucleus test examines the ability of a substance to cause the chromosomes of precursors of red blood cells in mice to break. Because mammalian red blood cells lose their nuclei as they mature, if any DNA is left in the mature blood cells, it is due to pieces of chromosomes breaking away from the rest of the nucleus as it is extruded from the immature blood cell during its maturation.

In a 28-day dose-range finding study, rats were fed diets containing freeze dried milk or beef from clones and ordinary cattle at concentrations of 0, 5, 10, or 20 percent of the diet.¹¹⁰ General signs, body weight, food consumption, urinalysis, hematology, blood chemistry, and organ weights were compared between groups. English-language summary tables were provided in the original Japanese-language report and have been provided in Appendix I. No biologically significant differences were reported in rats fed milk or meat from clones compared to rats fed milk or meat from ordinary cattle. In addition, the report notes that 10 cattle fed clone milk powder at 2.5, 5, or 10 percent of the diet showed no significant differences in body weight increase, indicating that the milk did not contain anti-nutrients or other toxicants to cattle. The duration of exposure is not reported.

Finally, the report provides some data from a 14-week oral feeding study conducted in rats to determine the effects of a diet containing milk and meat derived from clone cattle. Results of this study were subsequently published (Yamaguchi et al., 2007) and are discussed in detail in the next section.

v. Peer-reviewed publications

In a review of the nutritional value of milk and meat from cattle clones, Tome et al. (2004) described a study in which Wistar rats were fed either a milk-based or meat-based diet for 3 weeks. Milk and meat were obtained from cloned animals at INRA or non-cloned control cattle. Effects on food intake, body weight gain, organ weights and fasting glucose levels were measured. No differences were observed in the response of rats receiving either meat or milk from clones compared to rats fed meat or milk from control cattle. In addition, no differences were detected for IgG, IgA or IgM subtypes for rats receiving clone- or non-clone-derived diets. Further, no specific anti-milk or meat protein IgE responses were detected in rat sera. These results provide preliminary evidence that the nutritional value of milk and meat derived from clones is not different from that of milk and meat from non-clone animals.

In 2007, Yamaguchi et al. reported the results of a long-term (14-week) feeding trial in which rats were fed various amounts of milk and meat derived from conventionally bred cattle and cattle derived from either embryonic or somatic cloning. The authors claim that this published report is the first to use standard toxicological methods to study the effects of feeding meat and

¹¹⁰ Animal feeding studies to examine the toxicity of specific components of materials contained in foods are significant elements of a toxicity assessment. It is, however, generally recognized that animal feeding studies to examine the toxicological effects of whole foods (*i.e.*, feeding the whole food from a clone to the toxicology test animal) are of limited value due to the complex nature of the whole food, inability to provide sufficiently high doses of minor components of the whole food, and limited sensitivity of the assay. (Kessler et al. 1992; Codex Alimentarius, 2003 at ftp://ftp.fao.org/codex/Publications/Booklets/Biotech/Biotech_2003e.pdf).

milk from clones. Male and female rats (n=10 per group), approximately five weeks of age, were fed diets containing 1, 2.5 or 5 percent meat powder or 2.5, 5 or 10 percent milk powder derived from Japanese Black clones, Holstein clones, or non-cloned control cattle of the same breeds. Control rats were fed the basal diet, and diets containing meat or milk powder were formulated to be equal in nutritional value to the basal diet. Doses of meat and milk powder were based on a preliminary trial in which feeding 10 percent or 20 percent meat powder or 20 percent milk powder resulted in decreased feed intake, decreased body weight gain and (undescribed) clinical and pathological findings, or the equivalent of a maximum tolerated dose (MTD), the standard starting point for setting dosing for longer term toxicological studies. The authors speculate that adverse effects of feeding high concentrations (10-20 percent) of meat or milk were due to the nutritional imbalance of the diet (see footnote 28 for a discussion on why whole food feeding studies are not recommended by the US FDA). In addition to standard toxicological endpoints (e.g., clinical signs, body weight gain, feed intake, analysis of blood and urine), rats were subjected to a functional observational battery including reflex and sensory functions and locomotor activity.

None of the animals in the study died during its course. No abnormal clinical signs or changes in body weight or feed intake associated with consumption of meat or milk from either embryonic or SCNT clones. Body weight gain decreased was observed in one male rat exposed to the low dose of milk powder. Subsequent investigation of this rat indicated that it suffered from malocclusion (abnormal position of the teeth resulting in difficulty with chewing) leading the authors to conclude that the decreased weight gain was unrelated to exposure to milk powder from the SCNT clones. Average growth rate in rats fed milk or meat from embryonic or somatic clones, as indicated by body weight change, was not different from that in controls. Estrous cyclicity, assessed by vaginal smears on days 43-57 of the study, also was not affected by feeding milk or meat from either group of cattle clones.

The authors state that results of urinalyses conducted at weeks 4, 8, and 12 were similar between control rats and rats fed milk or meat from cattle clones. Similarly, the authors report no significant differences in hematology or blood chemistry between control and treated groups.

Compared to control animals, there were no differences found at necropsy in gross pathology or organ weights in rats fed milk or meat from cattle clones. Extensive histological findings are shown for 13 or 14 organs in rats fed meat or milk, respectively (testicular histology is missing from the group fed meat). Although numerous abnormal histological findings are reported, consumption of milk or meat from either embryonic or somatic clones did not increase the

incidence or severity of pathologies in any organ. The authors therefore concluded that the histological findings represent naturally occurring events unrelated to treatment.

Frequency of vocalization was included as part of the functional observational battery. At weeks 4, 8, and 12, changes were observed in the frequency of vocalizations between controls and rats fed milk or meat cattle clones. After comparing data from weeks 4, 8 and 12 with data from other weeks (data not shown), however, these frequencies were not consistently higher or lower than controls and, the authors concluded that the changes were incidental and unrelated to treatment. The authors reported that there was no effect of feeding milk or meat from either embryonic or SCNT clones on reflex, sensory functions, or locomotor activity.

The overall conclusion from this study is that consumption of meat or milk from cloned cattle did not alter the physiological condition of the rats. The study provides no evidence for a specific risk of consuming meat or milk from cattle derived from SCNT, which are not currently consumed by humans, compared to meat or milk from clones derived from embryonic cloning technology, which have been present in the Japanese food supply for several years.

Conclusions from Allergenicity and Feeding Studies in Rodents

The second prong of this risk assessment is based on the hypothesis that edible products from healthy cloned animals and their progeny are as safe to eat as edible products from conventionally produced livestock. CVM reviewed three studies in which the rat was used as a surrogate animal model to investigate possible biological effects of eating meat or milk from cattle clones. One of these feeding studies was conducted over an extended period (14 weeks) and included standard toxicological endpoints as well as a functional observational (behavior) battery. None of these studies demonstrated any change in the physiology or pathology of the rat following consumption of meat or milk from clones. Moreover, no evidence has been found to indicate that the allergenic potential of meat or milk from cloned cattle is greater than that of meat or milk from non-cloned cattle. No behavioral changes were observed. These findings are consistent with our conclusions using the Compositional Analysis approach, *i.e.*, that meat and milk from clones and their progeny are not materially different from meat and milk derived from conventional counterparts and thus do not pose any additional food consumption risks relative to food from conventional animals.

B. Drawing Conclusions Regarding Risks Associated with Consumption of Food Products from Animal Clones

1. Approaches for Decreasing Uncertainties

The fundamental problem in determining the quantity and types of data required to reduce the uncertainties associated with a judgment of “no additional risk” has bedeviled the scientific, risk, and regulatory communities. The impracticality of proving a negative and, in the absence of its proof, determining the consequent activities to identify the conditions under which concerns have been minimized to levels considered “acceptable” becomes the goal of a comprehensive risk assessment/management process.

In fact, certainty of prediction is unattainable in science. In its absence, risk assessment can provide risk managers with a systematic approach for bounding the “risk space” in which to operate by allowing assumptions and uncertainties to be clearly identified. Especially for new technologies in which uncertainty may be high, the “bounded framework” risk assessment process allows decision makers (both risk assessors and risk managers) to draw conclusions based on the data. Then, by explicitly addressing uncertainties, identifying biases, scientifically defensible (or alternatively, policy-based) judgments can be made about acceptable risk levels. The added benefit of such a process is that interested individuals are provided with a level of transparency that allows them to judge the quality of the science and the relative merits of decisions stemming from its evaluation.

This risk assessment has provided an overview of the molecular evidence for epigenetic dysregulation as the basis for obvious and subtle hazards that may arise in animal clones, the biological reasons for why subtle changes would not persist in progeny of healthy clones, the existing data on the health of animal clones and their progeny, and information on the composition of foods derived from clones and their progeny. These data can be incorporated into four procedural steps leading about to conclusions regarding food safety:

- *Bounding the risk space*, in which the “risk hypotheses” are explicitly identified and thereby the biases that influence the weight of the evidence evaluations regarding the health of the animals and the composition of food products derived from them;
- *Performing a weight of evidence evaluation of the data to characterize the risks* contained within the risk space, in which the information on food consumption hazards posed by cloning is summarized, and drawing conclusions based on the risk hypotheses presented in Step 1;

- *Characterizing the uncertainties* associated with the data and their interpretation, including identifying important data gaps based on Critical Biological Systems and Compositional Analysis approaches; and
- *In subsequent versions of this Risk Assessment, reevaluating previously estimated risks* based on new information to make new weight of evidence determinations.

2. Bounding the Risk Space

The two underlying risk hypotheses that explicitly bound the “risk space” in which the evaluations are being made are

- **Animal Clone Risk Hypothesis 1: *Clones are the Same as Sexually-Derived Animals***
Animal clones *are* biological copies of the donor animal, and *data confirming* overall findings of animal health and food product comparability *are sufficient* to indicate that no additional risk is posed by the consumption of such food products.

- **Animal Clone Risk Hypothesis 2: *Clones are Different from Sexually-Derived Animals***

Animal clones *may appear to be* faithful biological copies of the donor animal, but subtle hazards may have resulted from incomplete or inappropriate reprogramming of the genome as part of the SCNT process. In order to avoid additional risks above those posed by consumption of foods from sexually-derived animals under this hypothesis, *comprehensive health and compositional data* must be collected and analyzed to demonstrate that the animals are healthy, and that food products derived from them do not differ significantly from sexually-derived animals.

- **Clone Progeny Risk Hypothesis: *Gametogenesis Resets Epigenetic Dysregulation***

Normal, healthy clones reproducing via sexual reproduction give rise to progeny animals that are as healthy as animals derived from any other sexual reproduction event.

3. Developing Conclusions Regarding Food Consumption Risks

The conclusions that can be drawn with respect to the safety of consuming food products from animal clones and their progeny based on the data reviewed in this Risk Assessment follow. Because risk assessment is best performed recursively, risk assessment conclusions should always be considered to apply to the dataset that was examined; each conclusion is based on the

information that was available for consideration, but if additional data become available, a conclusion may change, or the degree of confidence placed in the conclusion may be adjusted. Nonetheless, risk managers need to make decisions at particular points in time, and despite the desire for recursive assessments, decisions often include statements about the degree of certainty that accompany them.

Each conclusion is followed by a statement on whether the judgment comes from application of Hypothesis 1 (Assumes Clones are the same as Sexually-Derived Animals), or Hypothesis 2 (Assumes Clones are Different from Sexually-Derived Animals), and the reason for the selection of that hypothesis (and its implicit bias).

As previously stated, the Risk Assessment assumes that all of the laws and regulations that apply to sexually-derived animals and the food products that come from them apply equally to animal clones, their progeny, and food products that are derived from them.

Our weight of evidence risk assessment conclusions are presented on a species-specific basis, except for bovine clones, where the large dataset allows for the consideration of individual developmental nodes. The weight of evidence evaluations take into account:

- All of the observations for that species (or developmental node);
- The extent to which those observations are coherent with biological assumptions;
- The consistency with which those observations are also seen across species, including the mouse model, where applicable;
- Uncertainties that persist in the evaluation, including the source of those uncertainties; and
- The confidence level in the conclusion based on all of the preceding considerations.

Because this is a qualitative, comparative risk assessment, it does not attempt to assign quantitative values to estimates of risk or safety. The strongest conclusions that can be drawn regarding positive outcomes in risk assessments of this type are “no additional risk” because outcomes are weighed against known comparators. In the context of edible products derived from clones, a finding of no additional risk means that food products derived from animal clones will not pose any additional risks relative to corresponding products from non-clones, or are as safe as foods we eat every day. As with all risk assessments, some uncertainty is inherent either in the approach we have used or in the data themselves. For each conclusion, CVM has attempted to identify the sources and extent of these uncertainties. A more complete discussion of sources of uncertainties and their implications can be found in Chapter VII.

4. Weight of Evidence Conclusions Regarding Food Consumption Risks for Clones and their Progeny

Based on this review of the body of data on the health of animal clones, the composition of meat and milk from those animals and corresponding information on clone progeny, CVM has drawn the following conclusions:

a. Cattle Clones

Edible products from perinatal bovine clones may pose some very limited human food consumption risk.

The underlying biological assumption in place for this age cohort is that perinatal clones may be physiologically more unstable at birth due to residual incomplete or inappropriate reprogramming of the donor nucleus. Data from both the peer-reviewed publications and Cyagra are consistent with that assumption; some perinatal clones do not survive for several reasons, including poor placentation, LOS, and in some cases, frank malformations. Although the health of surviving clones can be unstable for a period of time, many survivors tend to adjust to life outside the womb within a relatively short period, either on their own or with assistance from caregivers (see Juvenile Developmental Node). The peer-reviewed literature and Cyagra data indicate that, depending on the laboratory, a significant proportion of perinatal clones survive gestation and are born without significant health problems. Laboratory measures of key physiological functions do not appear to indicate that surviving animals are very different from conventional newborns. It is therefore unlikely that food consumption risks have been introduced into these animals.

The uncertainty associated with the preceding statement is relatively high, however, for the following reasons. First, postulated differences in epigenetic reprogramming between perinatal clones and comparators suggest that some subtle hazards may have been introduced into these animals. Second, the relatively poor condition of many of these perinatal clones also precludes the conclusion that no food consumption risks, such as nutritional imbalances, are present. Therefore, given that perinatal clones may differ from comparator animals of the same age, at this time, the Center concludes that they may pose a very limited nutritional risk for consumption as food.

i. Risk Hypothesis Statement for Perinatal Bovine Clones

At this time there is insufficient information to move from Hypothesis 2 (Clones are Different) to Hypothesis 1 (Clones are the Same), even though the available data neither identify nor predict

the presence of food consumption hazards (and subsequent risks) from these very young clones. The uncertainties in the data are relatively high and lead the Center to have a relatively low degree of confidence in the safety of edible products from perinatal bovine clones. We note, however, that it is highly unlikely that clones of this age group would be consumed for food.

A possible disposal method for clone calves that die or are euthanized due to poor health during the perinatal period is rendering.¹¹¹ Rendered materials have many uses, including ingredients in animal feed (meat and bone meal). Thus, it is important to consider possible risks to food animals that consume rendered materials derived from clones, as well as the possible risk to humans who consume those food animals. Although the Center has concluded that perinatal clone calves may pose some limited risk human consumption, we do not believe that the nature of this risk is unique to cloned calves because all of the pathologies observed in these calves are also observed (at lower rates) in calves produced using other ART methods. Therefore, there is no *a priori* reason to suggest that products derived from rendering perinatal clone calves would have any unique properties compared to rendered products derived from non-clone calves. The Center concludes that rendering perinatal clone calves will not pose risks, either to animals (via animal feed) or to humans consuming animals fed rendered material derived from clone cattle.

Edible products from juvenile bovine clones pose no additional food consumption risks relative to corresponding products from contemporary conventional comparators.

The underlying biological assumption for this developmental node is that if any anomalies were found in the youngest clones and those animals survived to be healthy adults, the juvenile developmental node would be a period of equilibration and normalization. The data appear to be consistent with such a hypothesis.

Clone calves that survive the perinatal period and are not affected by congenital abnormalities appear healthy and demonstrate normal patterns of growth and development. None of the physiological measures taken, including both clinical chemistry and hematology, indicated any food consumption hazards in these animals. For some bovine clones, the health problems observed during the perinatal period appears to extend into the juvenile period, resulting in an increased risk of morbidity and mortality in calf clones during the first six months of life. This risk appears to be the result of sequelae of developmental abnormalities present at birth that persist beyond the perinatal period (*e.g.*, musculoskeletal defects, prolonged recumbency, enlarged umbilicus, respiratory distress, poor thermoregulation). However, morbid animals

¹¹¹ Rendering is a process that uses heat (cooking), extraction of moisture, and separation of fat to convert animal tissues into stable, value-added materials. Material suitable for, and commonly used in rendering includes inedible tissues from slaughtered animals, carcasses that are condemned upon inspection, and sick animals that die on farm or in transit. The high temperatures used in the rendering process effectively destroy foodborne pathogenic microorganisms (Meeker 2006).

bearing these problems are not expected to contribute to food consumption risks because they are not expected to pass the antemortem inspection required to enter the food supply. No new or additional health risks were identified during the juvenile period other than those observed during the perinatal node. Therefore, the Center concludes that healthy juvenile bovine clones are not different from comparator animals of the same age, and pose no additional food consumption risk.

ii. Risk Hypothesis Statement for Juvenile Bovine Clones

The assessment began at the position of Hypothesis 2, but the scientific evidence has moved the assessment from Hypothesis 2 to Hypothesis 1 for maturing juvenile clones. The weight of the evidence and the underlying biological assumptions lead the Center to conclude that there would not likely be any additional risk from the consumption of food from healthy juvenile clones relative to corresponding products from their conventional comparators. The consistency of these observations across all of the data for juvenile bovine clones makes the uncertainty associated with this judgment relatively low, and provides the Center with a relatively high degree of confidence in judgments regarding the health of (and consequent food safety of edible products derived from) this age cohort of bovine clones.

Edible products derived from adult bovine clones pose no additional risk(s) relative to corresponding products from contemporary conventional comparators. This conclusion is based on application of both prongs (CBSA and Compositional Analysis) of the risk assessment approach.

The body of data comprising the CBSA approach on adult domestic livestock clones is made up of two components: data and information extracted from peer-reviewed publications and the Cyagra dataset. The empirical evidence on the health of these animals is consistent with the biological prediction that there are no underlying biological reasons to suspect that healthy animal clones pose more of a food safety concern than conventional animals of similar age and species.

The data from Cyagra survey indicate that healthy clones of the oldest cohort (6-18 months) are virtually indistinguishable from their comparators even at the level of clinical chemistry and hematology. These data also confirm the observation that physiological instabilities noted earlier in the lives of the clones are resolved juvenile developmental node (see previous conclusions regarding other developmental nodes), and do not reappear as the clones age. The statements regarding the health and apparent normality of animals of this age group from the peer-reviewed literature are consistent with the data evaluated by CVM. There are some reports of early deaths of clones; as these animals would not enter the food supply, they do not

pose a food consumption risk. Data on reproductive function in cows or bulls of this age cohort indicates that that healthy bovine clones surviving to reproductive maturity function normally and produce healthy offspring. These data are consistent across studies. Given that reproduction is the most difficult “biological hurdle” placed on an organism, the observation of normal reproductive function provides an additional degree of confidence to the conclusion of the appropriate development of these animals.

The reports on the composition analysis of meat or milk from bovine clones do not indicate that there are biologically significant differences in the composition of meat milk derived from clones and conventionally bred cattle. Although these comparisons are important for determining the effects of cloning on tissue composition (and therefore food consumption risks), it is equally, if not more important to consider historical or reference values. None of the observed values that could be associated with cloning fell outside historical reference ranges, and were also observed in meat or milk from conventionally bred animals. These observations illustrate the wide variability in the composition of milk and meat in general, and indicate that cattle clones produce meat and milk that display similar ranges of variability.

No novel components have been identified in meat or milk from clone cattle, and data from one report show no difference in allergenic potential for meat or milk derived from cattle clones compared with meat or milk from non-clone comparators. Similarly, neither meat nor milk from clone or non-clone cattle induced mutations in a mutagenicity assay (Japan 2004). Finally, none of the reports identified hazards, subtle or otherwise, that could pose food consumption risks.

iii. Risk Hypothesis Statement for “Adult” Bovine Clones

The assessment began at the position of Hypothesis 2: that animal clones may appear to be copies of the donor animal, but that the process of cloning may have introduced subtle hazards that could pose food consumption risks. As presented above, however, the weight of the evidence has moved the assessment from Hypothesis 2 to Hypothesis 1 (Clones are the same as their sexually-derived counterparts). Extensive and consistent empirical evidence, including epigenetic, physiological, and health data on individual animals and compositional analysis of milk and meat derived from individual animals, indicate that adult bovine clones are biologically equivalent to their contemporary comparators. Therefore, evidence confirming the health of the animals produced via similar methods, and evidence confirming the compositional similarity of meat and milk from clone and non-clone cattle indicates that there is no additional risk from the consumption of edible products from these animals relative to sexually-derived comparators. The consistency of the observations provide the Center with a high degree of confidence in judgments regarding the health of (and food safety of edible products derived from) this age cohort of bovine clones.

We note that given the economic considerations involved, it is not likely that many adult clones would enter the food supply as meat at this stage of the technology, unless they had suffered a non-treatable injury or old age. Milk products from lactating female bovine clones, however, could be introduced into the food supply.

b. Swine Clones

Edible products from adult swine clones pose no additional risk(s) relative to corresponding products from contemporary conventional comparators.

This conclusion is based on the same underlying biological assumption as cited for adult bovine clones (*i.e.*, non-transgenic clones would not likely express toxicants, no exogenous genes, and diseased animals would not be slaughtered for food). Because the data are more heavily weighted towards adult, market sized animals, judgments regarding the safety of food products from swine clones are provided in one aggregate set of comments.

Although generating swine clones appears to pose more technical difficulties than bovine clones, once piglets are born, the vast majority appear to be healthy. The health status of perinatal and juvenile animals is generally presented as “normal” or “healthy” in peer-reviewed publications. Results of one study conducted during the juvenile period indicate that the acute phase immune response may be altered in piglet clones. However, the overall implications of this study for the health of swine clones is unclear because the observations were made in only two piglets at a single time point in the life of the animals and the results have not been confirmed in other studies. Data from Archer et al. (2003a,b) and the ViaGen dataset do not indicate that older swine clones are more susceptible to infection. Therefore, even if the acute phase response of clone piglets is altered, it does not appear that this alteration affects their continued growth during the Juvenile Node or their health during the Reproductive or Adult Nodes. Further, data from Archer et al. (2003a,b) and the ViaGen dataset do not indicate that older swine clones are more susceptible to infection than their comparators.

The most compelling argument for the normal health status of swine clones has been presented by Archer et al. (2003a,b), who evaluated the behavior and physiological status of a small cohort of relatively young (15 weeks), and approximately market age (27 weeks) swine clones relative to closely related conventional pigs. Age-related physiological measures appeared to be normal, as demonstrated by levels of measures of growth such as alkaline phosphatase, calcium, and phosphorus and measures of immune system maturity such as globulin. No significant differences were observed in either behavior, epigenetic, or physiological measurements, indicating that these animals were not materially different from the comparators.

The data on the ViaGen clones (Appendix F) are on a relatively small number of animals, reared in very unusual settings (i.e., deprivation of colostrum, initial husbandry in pathogen-free conditions, switching to commercial settings) and are therefore confounded with respect to outcome. Nonetheless, the data indicate that even though the clone barrows were subjected to a significant immunological challenge after moving from pathogen-free conditions to more standard housing conditions, most clones were able to respond appropriately to this stress. Nonetheless, carcass qualitative characteristics were similar for clones and comparators in the ViaGen Dataset. Further, reproductive performance for these clone boars appears normal. No differences were noted in semen quality between clones and comparator boars; farrowing rates and litter sizes were within national averages. No biologically relevant differences were observed in the composition of meat from clones or comparators.

i. Risk Hypothesis Statement for Swine Clones

Based on both underlying biological assumption and confirmatory data, CVM concludes that consumption of food from healthy adult swine clones would not pose an additional risk above consumption of their conventional counterparts. The data from Archer et al. (2003a,b) is particularly compelling as it includes data on behavior, epigenetic reprogramming, and physiological measurements at two time points in the development of these clones. Likewise, data from ViaGen includes information on growth and reproduction, indicating that swine clones are not materially different from age-matched, genetically related swine. Taken together, these results support a Risk Hypothesis Level of 1 (clones are the same as sexually-derived animals) because they demonstrate a relatively high certainty based on biological plausibility, consistency of observations among different and compelling datasets, and consistency with responses observed across other clone species.

c. Sheep Clones

Except by relying on underlying biological assumptions, and by inference from other species, there is insufficient information on the health status of sheep clones to draw conclusions with respect to potential risks that could be posed from the consumption of food products.

With the exception of reports on Dolly, CVM was unable to find any publicly available reports on the health status of live sheep clones. There are several studies addressing methodological issues for optimizing the generation of clones, but these do not address post-natal health. There are reports of anomalies noted in fetal sheep clones that have died or been terminated, and reports on the pathology associated with animals that do not survive. Although these are instructive for understanding the molecular and developmental pathways that may be perturbed

during the process of SCNT, these studies have limited relevance to addressing food safety because the deceased animals would not have been allowed to enter the food supply. CVM was not able to find any reports on the composition of milk or meat from sheep clones.

i. Risk Hypothesis Statement for Sheep Clones

At this time there is insufficient information to support Hypothesis 1; Hypothesis 2 must be the default position with respect to potential food consumption risks from sheep clones. CVM was not able to find any studies providing specific evidence to show that sheep generated by SCNT are healthy and normal, and would therefore pose no additional food safety concerns beyond those of their conventional counterparts.

d. Goat Clones

Edible products from goat clones pose no additional food consumption risk(s) relative to corresponding products from contemporary conventional comparators.

This conclusion is based on the same underlying biological assumption cited for the other livestock species, and a relatively small but compelling dataset. Once clone embryos are transferred to surrogate dams and pregnancies are confirmed, the “success rate” for live births is quite high. The only anomaly noted was that approximately half of the cohort of goats reported on by Keefer et al. (2001a) appeared to have poor suckling response immediately after birth, but by the second day were responding normally and nursing from their surrogate dams. The animals appear to have developed well through reproductive age. The available data indicate their physiological responses are appropriate for age and breed. The reproductive development and function of male Nigerian Dwarf goat clones demonstrate that those animals functioned appropriately relative to age- and breed-matched comparators. One male progeny goat was derived from the buck clones; this animal also appeared to function in an age- and breed-appropriate manner. No meat or milk composition data were identified for goat clones.

i. Risk Hypothesis Statement for Goat Clones

Although the assessment began at Hypothesis 2, based on the underlying biological assumptions stated for the other clone species, consistency of responses with other species of clones, and a small but relatively rich dataset, CVM concludes that Hypothesis 1 more appropriately represents the conclusions regarding the food safety of goat clones. CVM places particularly high weight on the study of reproductive function, as it is one of the most complex physiological pathways to coordinate. The consistency of appropriate reproductive function, even in a small cohort of animals, adds to the confidence that can be placed in the judgment that these animals are as normal and healthy as their sexually-derived counterparts. Based on this finding, edible products from goats are not anticipated to pose more of a food consumption risk than their

sexually-derived counterparts. Further, given the data on the normal reproductive function of these animals, and a preliminary report of normal reproductive function of one male offspring of a male goat clone, CVM has more confidence in the empirical demonstration that clone progeny are as healthy as other sexually-derived animals.

e. Clone Progeny

Edible products derived from the progeny of clones pose no additional food consumption risk(s) relative to corresponding products from other animals.

Relative to the amount of meat and milk derived directly from clones in the U.S., it is likely that more food products (both meat and dairy) will be produced by the progeny of clones. Unlike their clone parents, progeny of clones are produced by sexual reproduction. The underlying biological assumption for health of progeny animals is that passage through the process of creating the cells that ultimately become ova and sperm naturally resets epigenetic signals for gene expression. This process is thought to effectively “clear” the genome of incomplete or inappropriate signals, and thus preclude genetic transmission of abnormalities from clones to their offspring. None of abnormalities that are observed in clone cattle have been reported in their progeny. For the progeny of swine clones, peer-reviewed studies together with extensive information in the ViaGen dataset provide direct data to indicate that these progeny are healthy at all developmental stages, and show that the composition of meat from the progeny of swine clones does not differ materially from pork derived from the offspring of conventionally bred swine. Both the cattle and swine data support the underlying biological assumption that the progeny of clone animals are essentially indistinguishable from the comparable progeny of other sexually derived animals.

ii. Risk Hypothesis Statement for Clone Progeny

Supported by extensive empirical evidence in the progeny of swine clones, we concur with the high degree of confidence that the outside scientific community (NAS 2002a,b) places in the underlying biological assumption that any abnormalities observed in clones will not be transmitted to their sexually-produced progeny. CVM therefore concludes that consumption of edible products from clone progeny will not pose any additional food consumption risk(s) relative to consumption of similar products from other sexually-derived animals.

5. Summary of Risk Hypotheses

The current weight of evidence suggests that there are no biological reasons, based on empirical data and underlying biological assumptions, to indicate that consumption of edible products from cattle, pigs, or goat clones poses a greater risk than consumption of those products from their non-clone counterparts. No frank or subtle hazards that could result in food consumption risks were identified in either the studies of the health of clones or in studies of the composition of milk or meat from clones. The level of certainty for these conclusions is highest for bovine clones, followed closely in degree of certainty by swine and, and then goat clones. The lack of species specific data for sheep clones precludes an evaluation of the risk for consumption of sheep clones at this time.

Consumption of edible products from the progeny of clones poses no additional risk(s) relative to those from other sexually-derived animals, based on underlying biological assumptions (supported by evidence from the mouse model system) and compelling empirical evidence on the health of clone progeny and their meat composition.

The level of confidence that may be placed in these overall conclusions is quite high, although additional data can always increase confidence.

a. Additional Issues

In addition to the hazards and risks described in the preceding portion of this risk assessment, there are a few issues that do not fit neatly into one of the categories that have been discussed previously. Many of these are overarching issues that may also have applicability to technologies other than SCNT.

i. Potential Allergenicity

The issue of allergenicity is one that is often cited for foods that do not have a long history of consumption. Although there is no reason to suspect that cloning will cause the synthesis of new proteins in animals that appear healthy and normal, there are two possible pathways that might pose an increased allergenic risk from the edible products of animal clones. One is an increase in the relative amount of an individual protein component of milk or meat that may only be present in very low or trace amounts. Cows' milk has been associated with true allergies (Cows Milk Allergy or CMA) in approximately six percent of the US population (Bernstein 2003). Caseins, although the predominant proteins in milk, do not appear to be the key allergens associated with CMA. The other possible pathway is that processing of the proteins during their generation in the mammary gland or muscle cells somehow alters their antigenic presentation. The Center cautions that these are purely hypothetical pathways, and that there has been no demonstration that either of these actually occurs.

In theory, evaluating the relative concentrations of milk proteins in clone and comparator milk could provide information to determine if the first risk exists. The study by Tian et al. (2005) provides just such a comparison using SDS/page technology. In practice, however, even this study highlights the difficulty in establishing the appropriate comparator and minimizing variability. Milk from non-clone dairy animals may vary in relative composition due to the influences of breed, diet, number of lactations, where in the lactation cycle the milk is collected, etc. Further, the level of exposure (dose) required to elicit an allergenic response is not well understood, and has been the subject of much discussion in the scientific literature (Taylor 2002) and among international regulatory bodies (Codex Alimentarius 2003¹¹²). Nonetheless, the limited studies provided (Japan 2002) show that milk from both SCNT and BNT clone cattle showed similar digestibility characteristics both *in vitro* and in a rodent *in vivo* assay. In addition, a rodent bioassay for allergic response did not show any significant differences in response between clone and non-clone derived milk. Combined with the underlying biological assumptions, these data support the lack of a unique allergic response to milk derived from clone cattle.

Similar risks are not likely to occur for meats, as meat allergies are so much less prevalent in the population that they are almost considered idiosyncratic, and individuals likely to suffer from meat-related allergies are likely to avoid those meats entirely. In addition, freeze dried meat from clone and non-clone cattle produced no difference in response in digestibility in both an in-vitro and rodent in-vivo assay, and there was no difference in difference in response in a rodent allergenicity bioassay (Japan 2002).

Finally, it is important to remember that relative and potential allergenicity in food is an issue that vexes the scientific and regulatory communities. FDA supports further research into the overall risk factors that cause individuals to exhibit aberrant immune responses. The agency has been actively involved in the evaluation of predictive tests at the laboratory and clinical level that address changes in protein structure and presentation. Nonetheless, it is important to remember that efforts such as those undertaken by the ILSI Allergy and Immunology Institute, the International Biotechnology Council, the National Academy of Sciences, Food and Agriculture Organization and the World Health Organization and the Codex Alimentarius address the allergenicity of *novel* proteins. These proteins are either new to the food supply as the result of the introduction of new foods, or are present in different matrices, as may be the case with transgenic plants or animals.

ii. Microbiological Effects

¹¹² ftp://ftp.fao.org/codex/Publications/Booklets/Biotech/Biotech_2003e.pdf

It has been suggested that epigenetic changes in animal clones could somehow alter the rumen and intestinal microflora of the ruminants (cattle, sheep, and goats), or the intestinal microflora of monogastric species (swine) (NAS 2002b). Such alterations in intestinal flora might be considered hazards because they could, in theory, result in increased levels of an existing zoonotic pathogen or the growth of a novel zoonotic pathogen. Shedding of these pathogens in fecal material could possibly result in a higher load of undesirable microbes on the carcass at slaughter, increasing the likelihood of contamination of the edible tissues.

The use of animal drugs has been postulated to alter the intestinal flora of treated food animals, resulting in an increased load of zoonotic pathogens in the food supply. The potential for animal drugs to induce this change was considered at length by the January 2002 CVM Veterinary Medical Advisory Committee on that topic¹¹³. Most of this independent scientific advisory committee found that animal drug use was unlikely to significantly impact pathogen load (or the prevalence of zoonotic pathogens), and that pathogen load has little or no impact on public health.

The Center is not aware of any studies that have characterized the intestinal flora of livestock clones, and the complexity of the intestinal microflora makes this an extremely difficult question to address directly. Although it is possible that epigenetic reprogramming in clones may have effects on the intestinal flora, this postulate can be challenged on the basis of animal health. The data reviewed in this risk assessment indicate that the vast majority of clones studied during the juvenile, reproductive and post-pubertal phases of life are as healthy as their sexually-produced counterparts. It therefore seems very unlikely that the milieu of intestinal microflora is abnormal in these animals, and that contamination of carcasses of clones due to bacterial shedding would pose a greater food consumption risk than that posed by conventional food animals.¹¹⁴ We further note that such alterations would not be unique to clones as all animals, regardless of their method of production, are subject to alterations in epigenetic programming.

iii. Unanticipated Effects

This risk assessment has attempted to identify the range of potential hazards and risks that could be generated as the result of SCNT in domestic livestock species. Although it may be possible for a healthy clone to express some proteins inappropriately, the same argument can just as easily

¹¹³ <http://www.fda.gov/cvm/VMAC/winter2002meet.htm>

¹¹⁴ A possible exception to this conclusion may be cattle clones during the perinatal period, many of which have impaired health that could result in higher than normal fecal shedding of pathogens. However, it is unlikely that very sick calves or calves with internal abnormalities would enter the human food supply because they would not pass state or USDA inspection (as required by the Federal Meat Inspection Act; <http://www.fsis.usda.gov/home/index.asp>).

be made for sexually-derived animals. At this time, there is no validated method for determining small differences in protein constituents in foods, and even if such methodologies existed, the question would still remain as to how to interpret them--what foods would be used as comparators, and what degree of variability would be considered to pose a risk (NAS 2004)?

Finally, the issue of the hypothetical dysregulation of endogenous substances that may pose a hazard by virtue of increased dose should be addressed. The primary concern in this case is the up-regulation of small molecules that may retain bioactivity in the bodies of the human (or animal) food consumer, usually by virtue of the lack of degradation in the intestinal tract. For example, levels of endogenous substances that have posed some public concern in the past (*e.g.*, estrogen and IGF-I) have been evaluated in bovine clones, and based on those data, there is no reason to expect that the levels of these substances in clones would pose any food consumption risks for humans.

iv. Technology Changes

This risk assessment has focused on the outcomes of cloning (*i.e.*, clones and their progeny) rather than on the cloning process itself. As discussed in Chapter II and elsewhere, however, at the time this risk assessment was developed, most clone producers use the same overall technology to produce clones. Clearly, different producers and laboratories may modify the process to enhance the overall success rate of the cloning process. In general, however, the clones that were evaluated in this risk assessment were produced by very similar processes. From a risk perspective, the important constant in technology used to produce these clones is that donor nuclei and recipient oocytes (or oöplasts) are not significantly manipulated beyond the obvious steps described in Chapter II. Thus, hazards other than epigenetic dysregulation are not introduced into clones.

Significant changes in cloning technology, especially those accompanied by donor nucleus or oocyte treatment regimens introducing new hazards into the overall process, would significantly increase the uncertainty associated with our judgments regarding the degree of risk that could accompany the resulting clones and clone food products. Without a careful evaluation of the animals arising from such methods, it would not be appropriate to speculate on the relative safety of the process from either an animal health or food safety perspective.

6. How Much (Information) Is Enough?

The question of determining when sufficient data have been collected in order to allow high confidence in risk-based decisions regarding edible products from animal clones is difficult to

determine in the abstract. In practice, the answer is “it depends on what questions you ask, and how the data answer those questions.”

Because the nature of the technology has generally precluded generating large datasets on clones with good statistical power, CVM constructed a systematic approach to frame the appropriate questions (hazard identification), evaluated the available data (hazard characterization), and attempted to characterize resulting risk (probability of harm given that exposure occurs). This weight of evidence approach allows for the evaluation of the data from the CBSA and Compositional Analysis prongs of the Risk Assessment as part of an overarching whole. The conclusions from this risk assessment represent the judgment of CVM veterinarians, animal scientists, toxicologists, and risk assessors. The underlying assumptions for clones and their progeny were that the animals needed to meet all relevant federal, state, and local laws and regulations for conventional animals, and the food products derived from clones or their progeny also had to meet relevant federal, state, and local laws and regulations.

When considered across the Developmental Node spectrum, the data on the health of livestock clones were remarkably consistent across species, despite initial anomalies that appear to be species-specific. For example, although LOS may be more prevalent in cattle and sheep, most surviving animals normalize initial anomalies and become “healthy and normal.” This consistency has increased the value of even small datasets (*e.g.*, goats), and has contributed significantly to the judgments regarding the health of these clones and their suitability as food sources. In addition, CVM evaluated a number of reports on the composition of meat and milk from clones and their progeny. No biologically important or safety-relevant differences were noted when compositions were compared to standard databases or contemporary comparator controls. If anything, these data confirm the rather wide variability in the composition of meat and milk eaten on a daily basis. In summary, no toxicological hazard of concern for the human consumer has been identified in any of the reported studies. Although additional data from other sets of animals, particularly in other species routinely used for food, could be useful in increasing the confidence that may be placed in overall judgments regarding food safety, the weight of the evidence at this time is sufficient for the agency to draw the conclusions it has made in this Risk Assessment with reasonable certainty.

Chapter VII:
Summary and Conclusions

Chapter VII

Summary and Conclusions

Somatic Cell Nuclear Transfer (SCNT) is a technology still relatively early in its development. Cloning has been accomplished in relatively few species, with most of our current information stemming from studies in cattle, swine, goats, and mice. This Risk Assessment has addressed the hazards and potential risks that may be experienced by domestic livestock (*i.e.*, cattle, swine, sheep, and goats) involved in the cloning process (Animal Health Risks) and whether edible products from animal clones or their progeny pose food consumption risks beyond those of their conventional counterparts (Food Consumption Risks).

A. Methodology

This Risk Assessment used two complementary approaches, the *Critical Biological Systems Approach* (CBSA) and *Compositional Analysis Approach*, to identify and characterize potential animal health and food consumption hazards associated with cloning. It then employed a weight of evidence approach to draw conclusions regarding risks to animal health and for consumption of food products from clones and their progeny. This weight of evidence approach consisted of four steps:

- (1) *Evaluation of the empirical evidence* (*i.e.*, data on molecular mechanisms, physiological measurements, veterinary records, and observations of general health and behavior) for the species being considered;
- (2) *Consideration of biological assumptions* predicated on our growing understanding of the molecular mechanisms involved in mammalian development;
- (3) *Evaluation of the coherence of the observations* with predictions based on biological mechanisms; and
- (4) *Evaluation of the consistency of observations* across all of the species considered, including the mouse model system.

The Risk Assessment also assumed that animal clones, their progeny, and all food products derived from either clones or progeny must meet the same federal, state, and

local laws and regulations as food from conventionally bred animals.

Because no exogenous genes have been introduced into animals derived via SCNT, the underlying assumption has been that adverse outcomes observed in animal clones arise from epigenetic modifications due to incomplete reprogramming of the donor cell nucleus. Methodological and technological components (*e.g.*, selection of donor cell, cell cycle stage, *in vitro* factors associated with the SCNT process) may also affect outcomes as they do for other ARTs.

B. Conclusions Regarding Risks to Animal Health

To assess the health of animal clones for both the animal health and food consumption risk portions of this risk assessment, we used the *Critical Biological Systems Approach* (CBSA), which divides the life cycle of clones into five distinct Developmental Nodes. Available data for each species were sorted into these Developmental Nodes to evaluate the data systematically and to determine whether there are common developmental difficulties among the livestock clones or whether animals “recover” from initial infirmities related to cloning.

The results of the CBSA indicated that significant adverse health outcomes have been reported for cattle and sheep clones and their surrogate dams. These tend to include dystocia and high gestational mortality. In cattle and sheep clones, post-natal mortality tends to be concentrated in the perinatal period, and is higher in clones than in animals produced using other assisted reproductive technologies (ARTs).

To date, no adverse outcomes have been noted in clones that have not been observed in animals derived via other ARTs or natural mating. Goats and swine appear to develop without significant abnormalities. The incidence of adverse outcomes in cattle and sheep clones, however, appears to be higher than in other forms of ARTs. Common adverse developmental outcomes that have been observed in cattle and sheep generally fall under the heading of Large Offspring Syndrome (LOS), although there may be others. Newborn animals with LOS tend to be bigger than average for their breed and species, may show edema or other abnormalities of the lungs and other parts of the body, and exhibit cardiovascular and respiratory problems.

Most animal clones that survive the critical perinatal period appear to grow and develop normally. Even animals with physiological perturbations, including less severe manifestations of LOS, seem to resolve them, usually within a period of weeks. More severe complications of LOS may persist into the juvenile period, but clones do not

appear to develop any additional health risks unrelated to those that were observed during the perinatal period. Clones that reach reproductive age appear to be normal in all of the measures that have thus far been investigated, and appear to give rise to healthy, apparently normal progeny. Mature clones appear normal and healthy, and are virtually indistinguishable from their conventionally bred counterparts.

Studies that evaluated epigenetic reprogramming in live, healthy clones indicate that although there is some variability between clones and their sexually-derived counterparts, these clones have undergone sufficient epigenetic reprogramming to carry out the coordinated functions necessary for survival and normal functioning. Molecular analyses reveal relatively small methylation differences, and either the animals are tolerant of such differences, or the epigenetic differences are below the threshold that poses observable adverse health outcomes.

C. Conclusions Regarding Food Consumption Risks

In order to evaluate potential food consumption risks associated with healthy-appearing clones, we developed a two-pronged approach. The first part of the approach is based on the hypothesis that a healthy animal is likely to be safe to eat, and relied on the CBSA. The second component of our two-pronged approach, or the *Compositional Analysis Approach*, assumed that if there are no material differences between the composition of milk and meat from animal clones (and their progeny) and their non-clone counterparts, then edible products derived from clone meat or milk would be as safe to eat as corresponding products from non-clones. This assessment assumed that animal clones and their progeny would be subject to all of the same federal and state requirements for milk and meat from conventionally bred animals.

Because each clone arises from an independent event, identification and characterization of potential subtle hazards (*e.g.*, alterations in gene expression, immune function, or hormone levels) is best accomplished by the evaluation of individual animals, at as fine a level of resolution as possible. Characterization of the overall functionality of clones, however, is likely best considered by evaluating the animal as a whole, in particular assessing the degree to which highly complex functions have been integrated, for example by demonstrating normal growth and successful reproduction.

The food consumption portion of the risk assessment postulated that because the only hazards that may be present in clones would arise from epigenetic dysregulation, and because only healthy animals meeting the same standards that conventional food animals or their edible products meet would be permitted for use as food, the only hazards that

could be present in these animals would be subtle. Part of the purpose of the CBSA approach was to determine whether any such subtle hazards could be identified. Following a detailed analysis of a wide range of health data, we identified only a few examples of altered physiological parameters in clones, and these were limited to young clone calves and swine. None of these alterations were correlated with any discernable adverse effects on animal health. We therefore concluded that no subtle hazards were identified that could pose food consumption risks from cattle, swine, or goat clones. The lack of allergenic and mutagenic effects in studies designed to detect those outcomes also indicated there were no food consumption risks.

Analyses of the composition of meat from bovine and swine clones and milk from bovine clones consistently indicate that there are no biologically relevant differences between the composition of food from clones, or their close comparators. In addition, there is no material difference, based on these studies, between the composition of meat and milk from clones and historical reference ranges of the composition of food from conventionally-bred animals.

D. Conclusions Regarding Food Consumption Risks from Clone Progeny

Progeny of animal clones are not anticipated to pose special animal health or food consumption concerns, as they are the product of sexual reproduction. The production of gametes by clones is expected to reset even those residual epigenetic reprogramming errors that could persist in healthy, reproducing clones. Studies in cattle and swine indicate that the progeny of clones are healthy and indistinguishable from other sexually-derived swine comparators. Because the value of clones lies in their genes, they are most likely to be used as breeding stock, and their use as food would be incidental. Almost all of the production animals (*i.e.*, sources of meat and milk) from the overall SCNT process are therefore likely to be sexually-reproduced progeny of clones. An extensive dataset on the progeny of swine clones indicates that the composition of meat from those animals does not differ materially from that of comparator animals or historical reference ranges.

E. Weight of Evidence Evaluations

As is the case for all risk assessments, the amount of data and information available on endpoints varied in quantity, with respect to both number of studies and number of data points available to evaluate. In some cases, evaluation of one endpoint indicated different outcomes in different laboratories. This Risk Assessment took such differences into account by applying the criteria listed above to consider the overall weight of evidence—that is, the extent to which the data supported each other, and where they did not, to

provide a framework for determining the underlying basis for the differences. Because the weight of evidence considers all of the information in one framework, it does not require any particular number of studies on any particular endpoint in order to be valid; rather, by considering all of the information together, it is able to develop a coherent perspective that takes into account biological assumptions, mechanisms, and empirical evidence. It also allows for the identification of uncertainties, and provides a science-based path for further investigations to resolve those uncertainties.

The judgment that cattle, swine, and goat clones meeting the same federal and state requirements as conventionally bred food animals would not likely pose food consumption risks, of course, contains some residual uncertainty. The source(s) of the uncertainty may be sorted into three categories:

1. Uncertainties associated with *empirical observations*. Uncertainties are lowest for those individual clones whose health has been thoroughly evaluated and, by inference, other clones produced using the same methodology. The uncertainties associated with the evaluation of empirical observations can be a function of the size, consistency, and quality of the data being evaluated. For example, the degree of confidence that can be placed in judgments arising from a well-conducted, consistent, and extensive dataset is much higher than from a small, poorly designed, and highly variable dataset. Further, because datasets tend to arise from an individual laboratory or producer, the uncertainties associated with that producer and method are lower than for other laboratories or producers for which less information is available.
2. Uncertainty stemming from *biological sources* can be minimized by the evaluation of the clones themselves. The most important factor in this evaluation is the healthy survival and functionality of individual clones, indicating that either the animal has minimal epigenetic dysregulation, or that any initial epigenetic dysregulation has been resolved. Uncertainty would be the lowest for individual clones demonstrating successful reproduction.
3. Uncertainties stemming from *technological or methodological* grounds encompass the degree to which judgments regarding clones arising from technologies in use when this risk assessment was conducted can be applied to modifications of the technology. These may only be resolved by the evaluation of the outcomes of those technological changes (*i.e.*, the actual clones).

Thus, our overall conclusions are:

For Animal Health: SCNT results in an increased frequency of health risks to animals involved in the cloning process, but these do not differ qualitatively from those observed in other ARTs or natural breeding. Cattle and sheep exhibit a set of clinical signs collectively referred to as LOS that do not appear to be present in swine or goats. Surrogate dams are at risk of complications from birth if the fetus suffers from LOS, or from accumulation of fluid in the placenta (hydrops). Clones exhibiting LOS may require additional supportive care at birth, but can recover and mature into normal, healthy animals. Most clones that survive the perinatal period are normal and healthy as determined by physiological measurements, behavior, and veterinary examinations. Progeny of animal clones also have been reported as normal and healthy.

For Food Consumption Risks: Extensive evaluation of the available data has not identified any food consumption risks or subtle hazards in healthy clones of cattle, swine, or goats. Thus, edible products from healthy clones that meet existing requirements for meat and milk in commerce pose no increased food consumption risk(s) relative to comparable products from sexually-derived animals. The uncertainties associated with this judgment are a function of the empirical observations and underlying biological processes contributing to the production of clones. Uncertainty about the health of clones decreases as they age and have more time to exhibit the full range of functionality expected of breeding stock. Edible products derived from the progeny of clones pose no additional food consumption risk(s) relative to corresponding products from other animals based on consistent empirical observations, underlying biological assumptions, and evidence from model systems.

Glossary

The following terms are defined as they are used within the current risk assessment. Unless otherwise indicated, definitions provided are the commonly accepted use of the term(s) at the Center for Veterinary Medicine, and may have been derived from various sources.¹¹⁵

allele	Any alternative form of a gene that can occupy a particular chromosomal locus.
anal atresia	Abnormally closed anal opening.
analyte	A substance undergoing analysis.
aneuploid	Describes a cell or organism which has an abnormal total number of chromosomes and where numbers of individual chromosomes are out of proportion with the numbers of the other chromosomes. Too many chromosomes is called hyperploidy; too few is called hypoploidy.
animal clones	Animals derived via somatic cell nuclear transfer techniques. The terminology employed in this assessment did not use “cloned animals.” The phrase “cloned animals” does not clearly differentiate between the animal serving as the source of genome being propagated, or the animal that has been generated from a particular source. For example, the sentence “That field contains several cloned animals” does not specify whether the animals had been used as a source of material for SCNT or whether they had been generated by that technology.
ARTs	Assisted reproductive technologies.
biallelic	Referring to expression of two alleles at the same time.

¹¹⁵ The various sources used for these definitions include: Dorland’s *Medical Dictionary*, 30th Ed., W.B. Saunders Company, Philadelphia, 2003; *Dictionary of Epidemiology*. 3rd Ed. John M. Last. Oxford University Press, 1995; [HTTP://bioethics.gov](http://bioethics.gov); <http://biotech.icmb.utexas.edu>; *Large Animal Internal Medicine*, 2nd Ed., Smith, B.P., Ed., Mosby – Year Book, Inc., St. Louis, 1996.; *Veterinary Medicine: A Textbook of the Diseases of Cattle, Sheep, Pigs, Goats and Horses*. 7th Ed. Blood, D. C. and O. M. Radostits, Philadelphia: Bailliere Tindall Company, 1989. *The Merck Veterinary Manual*, 8th Ed. Online Version. C.M. Kanh and S Line, Ed. Merck & Co., Inc, NJ, 2003; and *The American Heritage® Dictionary of the English Language*, 4th Ed., Houghton Mifflin Company. 2002.

bioengineered animals	The broadest category of animals associated with molecular biology techniques, including animal clones and all genetically engineered animals.
blastocyst	An early stage in the development of mammalian embryos, when the embryo is a spherical body comprising an inner cell mass that will become the fetus and an outer ring of cells, the trophoctoderm, that will become part of the placenta.
blastomere	Any one of the cells formed from the first few cell divisions in animal embryology. The embryo usually divides into two, then four, then eight blastomeres, and so on.
Blastomere Nuclear Transfer (BNT)	An assisted reproductive technique in which a blastomere is used as a donor for nuclear transfer into enucleated oöplasts.
capacitation	The process of sperm maturation (or activation) that occurs post-ejaculation. Allows the spermatozoa to go through the acrosomal reaction in which factors in the sperm head that allow it to penetrate the egg are released and fertilize an oöcyte.
caruncle	The site of attachment in the maternal uterus of the ruminant for the placental cotyledon (<i>see cotyledon</i>).
centromere (centromeric)	A specialized chromosome region to which spindle fibers attach during cell division (mitosis) that is genetically inactive. This is constricted region of a mitotic chromosome that holds sister chromatids together—the crossing point in the “X” often used to depict chromosomes.
chimera	An organism or recombinant DNA molecule created by joining DNA fragments from two or more different organisms.
chondrocyte	A mature cartilage cell.
chorion	The outermost membrane enclosing the fetus. It is formed from tissues on the outside of the embryo such as the trophoblast, and the part of it attached to the uterus wall eventually develops into the placenta.
chromatid	One of the two daughter strands of a duplicated chromosome.
chromatin	The network of fibers of DNA and protein that make up the chromosomes of the eukaryotic nucleus during interphase.

chromosome(s)	A structure composed of one very long molecule of DNA and associated proteins (<i>e.g.</i> histones) that carries hereditary information.
cleavage	The series of mitotic divisions by which a fertilized animal ovum changes, without any overall change in size, into a ball of smaller cells constituting the primitive embryo.
clone	A group of cells or individuals that are genetically identical as a result of asexual reproduction including nuclear transfer.
cloning	Asexual reproduction of animals using somatic cell nuclear transfer (SCNT).
coherence	The extent to which a hypothesized causal association is compatible with preexisting theory and knowledge.
colostrum	The first fluid secreted by the mammary glands at the time of birthing that is rich in antibodies and nutrients, and precedes the production of true milk. Its ingestion confers passive maternal immunity on the offspring of some species.
Comprehensive Veterinary Exam (CVE)	Systematic approach for examining domestic livestock animals and making informed judgments as to their health. The CVE contains both objective and subjective information and is performed by a veterinarian.
congenital	Existing at, and usually before, birth; referring to conditions that are present at birth, regardless of their causation.
consistency	Close conformity between findings in different studies conducted by different methods or different investigators.
cortisol	The major natural glucocorticoid hormone synthesized in the zona fasciculata of the adrenal cortex; it affects the metabolism of glucose, protein, and fats. It also regulates the immune system and affects many other functions.
cotyledon	A lobule structure in ruminant placentae that form contact points between the fetal-derived placental tissues with the maternal caruncles (attachment points) of the uterus to form the functional units called placentomes. It consists mainly of a rounded mass of villi.
cryptorchid	A male animal with one or both testicles retained within the body cavity.

cull	To remove unwanted members or parts from a herd.
cytoplasm	The living contents of the cell, exclusive of the nucleus, consisting of an aqueous protein matrix or gel, and where essential membranes and cellular organelles (mitochondria, plastids, etc.) reside.
de novo	Literally means “anew.” Beginning a process from its origin without prior plans.
dermatitis vegetans	A hereditary disease of the skin in swine (<i>see hyperkeratosis</i>).
differentiation	The process whereby relatively unspecialized cells, <i>e.g.</i> embryonic or regenerative cells, acquire specialized structural and/or functional features that characterize the cells, tissues, or organs of the mature organism or some other relatively stable phase of the organism’s life history.
diploid	Having two sets of chromosomes.
DNA	Abbreviation for deoxyribonucleic acid; one of the two types of nucleic acids that constitutes the genetic material of most known organisms; usually in double helix form.
DNA polymerase	The enzyme responsible for copying DNA. Common name for either of two categories of enzymes that catalyze the synthesis of DNA from deoxyribonucleoside triphosphates in the presence of a nucleic-acid primer.
ductus arteriosus	The blood vessel between the pulmonary artery (carries blood from the heart to the lungs for oxygenation) and the aorta (carries oxygenated blood to the rest of the body). During gestation the ductus arteriosus bypasses the fetal lungs, and is normally sealed after birth.
ductus venosus	The blood vessel between the umbilical vein and the caudal vena cava (carries oxygenated blood from the dam, bypassing the liver, through the vena cava to the heart of the fetus). It is normally sealed shortly after birth.
dysregulate	Abnormal or impaired control of gene expression.
dystocia	Abnormal or difficult labor.

ectoderm	The outermost layer of tissue in a developing embryo that will eventually become the skin and/or other outer surface of the organism, the outermost parts of the nervous system, and various other outer and external organs depending on the organism.
embryo	In mammals, the term is restricted to the structure present in the early part of gestation that develops into a fetus.
embryo cloning	Another term for blastomere nuclear transfer.
empirical	That which can be seen or observed alone, often without reliance on theory.
endoderm	The innermost layer of tissue in a developing animal embryo that will eventually become the digestive tract, respiratory tract, and various other things depending on the organism.
enucleate	Removal of an organ or mass from its supporting tissues.
epigenetic	Describing any of the mechanisms regulating the expression and interaction of genes, particularly during the development process. These include changes that influence the phenotype but have arisen as a result of mechanisms such as inherited patterns of DNA methylation rather than differences in gene sequence: imprinting is an example of this.
epigenetic reprogramming	In the case of somatic cell nuclear transfer (SCNT), the process of altering the instructions governing the expression of genes in the chromosomal DNA of the donor cell such that embryonic or totipotent (able to differentiate along any line or into any type of cell) gene expression conditions are reestablished.
epigenetic variation/effects	Non-hereditary, phenotypic changes in the expression in a single gene.
estrous	Pertaining to estrus. (Adjective)
estrus	The recurrent, restricted period of sexual receptivity in female mammals other than human females, marked by intense sexual urge. (Noun)
euchromatin	One of two types of chromatin seen during interphase of the cell cycle. It is genetically active (transcription occurs in it) and less condensed than heterochromatin (the other type of chromatin).
eukaryote	<u>An organism</u> whose <u>cells</u> have a true nucleus, <i>i.e.</i> , one bounded by a

nuclear membrane, within which lie the chromosomes, combined with proteins and exhibiting mitosis; eukaryotic cells also contain many membrane-bound compartments (organelles) in which cellular functions are performed.

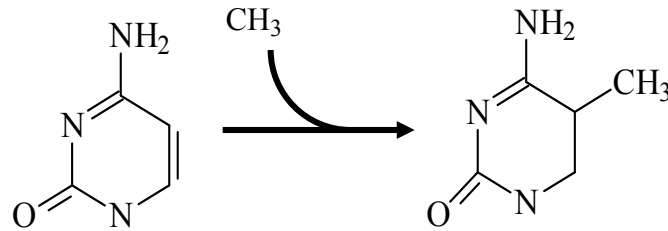
F₁	Abbreviation for filial generation 1 (first generation). The initial hybrid generation resulting from a cross between two parents.
farrow	In swine, the process of giving birth. Also used to describe a litter of pigs.
fat cow syndrome	A multifactorial disease condition often occurring in dairy cows following parturition; associated with excessive mobilization of fat to the liver in well-conditioned cows. This mobilization is induced by the negative energy balance and hormonal changes. Presenting signs usually include depression, anorexia, weight loss, and weakness that can lead to recumbency.
fecundity	The physiological ability to reproduce, as opposed to fertility.
fertility	The <u>capacity</u> to conceive or <u>induce conception</u> .
foramen ovale	A hole in the fetal heart between the right and left atria, for the purpose of bypassing the lungs. It is normally sealed shortly after birth.
founder animal	An organism that serves as the progenitor of a particular lineage.
freemartin	A sexually maldeveloped female calf born as a twin to a normal male calf. The reproductive tract hypoplasia results in an infantile uterus that does not develop appropriately with the growth of the rest of the calf and fails to respond to puberty. It is commonly sterile and intersexual as the result of male hormones reaching it through shared placental blood vessels.
gamete	A mature reproductive cell capable of fusing with a cell of similar origin but of opposite sex to form a zygote from which a new organism can develop. Gametes normally have haploid chromosome content. In animals, a gamete is a sperm or egg.
gametogenesis	The process of the formation of gametes.
gene expression	The process by which a cell transcribes the information stored in its genome to carry out the functions of life.
genetic	The process of rearranging the genome of the nucleus to restore a

reprogramming	cell's totipotency so it can differentiate into different types of cells and develop into a whole organism. Also known as de-differentiation.
genetically engineered animals	A subset of animals associated with molecular biology techniques. Includes transgenic animals, animals subjected to gene therapy and mosaic animals. This subset does not include animal clones.
genome	The full set of genes in an individual, either haploid (the set derived from one parent) or diploid (the set derived from both parents).
genotype	The entire genetic constitution of an individual.
germ cell	A reproductive cell such as a spermatocyte or an oöcyte, or a cell that will develop into a reproductive cell.
gilt	A female pig that is intended for breeding but has not yet given birth.
gonadotropin	Any hormone that stimulates the testes or ovaries.
haploid	An individual or cell having only one member of each pair of homologous chromosomes.
harm	An adverse outcome.
hazard	Something that can produce harm.
heifer	A female bovine that has not yet produced a calf.
hematology	The branch of medicine that deals with the blood and blood-forming tissues.
hemogram	A written record or graphic representation of a detailed blood assessment such as the complete blood count or differential leukocyte count.
hermaphrodite	An individual characterized by the presence of both male and female sex organs. The condition is caused by an anomalous differentiation of the gonads: an animal with ambiguous genitalia, typically a penis with ovaries or a vulva with testicles
heterochromatin	The condensed and genetically inactivated portion of a chromosome.
histones	Chromatin proteins commonly associated with the DNA of somatic

cells in eukaryotes and they are involved in packaging of the DNA and the regulation of gene activity.

hormone	A chemical substance produced in the body by an organ, cells of an organ, or scattered cells, having a specific regulatory effect on the activity of an organ or organs. The term was originally applied to substances secreted by endocrine glands and transported in the bloodstream to distant target organs, but later it was applied to various substances having similar actions but not produced by special glands.
hydroallantois	Abnormal fluid accumulation in the allantoic cavity of the placenta. (<i>See hydrops.</i>)
hydrops	Edema. Hydrops refers to a set of conditions relating to abnormal fluid accumulation in one or more compartments of the placenta and/or the fetus itself, and are alternatively referred to as hydroallantois, hydramnios or hydrops fetalis, depending on where the edema occurs.
hyperkeratosis	Characterized by lesions of the superficial layers of the epidermis. These lesions rapidly become covered with scales, and then develop hard, dry crusts with deep fissures. Generally referred to as parakeratosis in swine.
hypoplasia	Incomplete development or underdevelopment of an organ or tissue.
hypospadias	A developmental anomaly in which the urethra opens inferior (below) to its usual location; usually seen in males with the opening on the underside of the penis or on the perineum.
imprinted genes	Those genes whose degree of expression is determined by their derivation from either the dam or the sire.
<i>in vitro</i>	Outside the organism, or in an artificial environment. This term applies, for example, to cells, tissues or organs cultured in glass or plastic containers.
<i>in vivo</i>	Literally means "in life;" a biologic or biochemical process occurring within a living organism.
inner cell mass	The group of cells in a blastocyst that are destined to form the fetus.
inner cell mass (ICM)	A cluster of cells within the blastocyst. The inner cell mass will form all of the tissues of the organism and these cells are pluripotent.

ketonuria	Ketone bodies in the urine, as in diabetes mellitus; called also acetonuria and hyperketonuria.
ketosis	A metabolic disease of lactating dairy cows characterized by weight loss, decreased milk production, and neurologic abnormalities that usually occur during the first 6 weeks of lactation.
Large Offspring Syndrome (LOS)	A morphologic syndrome presumably expressed at the molecular and physiological level due to some alterations in embryonic gene expression. Animal clones with LOS may experience difficulties in developing and maintaining the placenta. An LOS fetus is unusually large for its species, has longer than usual gestation periods, and often has immature lungs or heart abnormalities. Kidneys and liver may also be affected.
leukocytosis	A transient increase in the number of leukocytes (white blood cells) in the blood.
leukopenia	A reduction in the number of leukocytes in the blood.
locus	The specific site of a gene on a chromosome.
long terminal repeats	A double-stranded sequence, generally several hundred base pairs long, at the two ends of the genetic sequence of retroviruses.
mastitis	Inflammation of the mammary gland or breast.
meconium	First stool in the intestine of a full-term fetus.
meiosis	The process in which a single diploid cell becomes four haploid cells in two consecutive divisions of the nucleus of an eukaryotic cell. In multicellular higher organisms this occurs only in the progenitors of sex cells and never in somatic cells.
methylation	The addition of a methyl group (-CH ₃) to a larger molecule (e.g. cytosine methylation)
	cytosine 5-methyl cytosine



metritis	Inflammation of the uterus.
mitosis	The division of a eukaryotic cell nucleus to produce two daughter nuclei that contain identical numbers of chromosomes and that are identical genetically to the parent nucleus except where crossing over or mutation has occurred.
monozygotic twin	One of a pair of twins derived from a single fertilized egg or zygote. <i>Synonym:</i> identical twin.
morphology	The form and structure of an organism, organ, or part.
morula	The solid mass of blastomeres formed from the cleavage of a fertilized ovum or egg.
murine	Pertaining to or affecting mice or rats.
neoplasia	Abnormal and uncontrolled cell growth that often produces a tumor (a <i>neoplasm</i>) that may or may not be cancerous (<i>i.e.</i> , capable of spread or metastasis).
nuclear transfer	Transferring the nucleus with its chromosomal DNA from one (donor) cell to another (recipient) cell.
nucleic acids	A large molecule composed of nucleotide subunits. DNA (deoxyribonucleic acid) and RNA (ribonucleic acid) are examples.
nucleoside	A molecule composed of a purine or pyrimidine nitrogenous base attached to the five-carbon sugar. This glycosylamine is a component of nucleic acids.
nucleotide	A molecule composed of a purine or pyrimidine nitrogenous base attached to the five-carbon sugar which also has a phosphate group attached to it. It is the constitutional unit into which nucleic acids are broken down by partial hydrolysis and from which they are built.

nucleus	The most conspicuous organelle of a eukaryotic cell; it contains the chromosomes and is the site of genomic DNA replication and or RNA synthesis in the cell.
oöcyte	A cell of an animal ovary that undergoes meiosis to form an ovum.
oöplasmic remodeling	After nuclear transfer, the cytoplasm of the oöcyte (oöplasm) alters the morphology of the nucleus, so that it more closely resembles the nucleus of an embryo.
oöplast	The remaining portion of the oöcyte following enucleation.
oviduct	A tube from the ovary to the uterus through which ova (eggs) may pass.
ovum	The female reproductive cell which, after fertilization, becomes a zygote that develops into a new member of the same species. Also called an egg.
parakeratosis	A nutritional deficiency disease of 6- to 16-wk-old pigs that is characterized by lesions of the superficial layers of the epidermis. It is a metabolic disturbance resulting from a deficiency of zinc or an excess of calcium in the diet.
parity	The condition of having given birth.
parthenogenesis	The development of a new individual from an unfertilized female gamete.
parturition	The act or process of giving birth to offspring.
patent ductus arteriosus	The failure of the ductus arteriosus to close after birth resulting in extra blood flow to the lungs and recirculation of oxygenated blood to the lungs rather than the rest of the body.
patent urachus	The failure of the urachus to close during parturition, resulting in the inability to excrete urinary waste.
phagocytosis	The uptake of extracellular materials by the formation of a pocket from the cellular membrane and its subsequent pinching off.
phenotype	The totality of the observable functional and structural characteristics of an organism as determined by its genotype and its interaction with its environment.

phytate(s)	A form of phosphorus commonly occurring in grain products, which is indigestible in non-ruminant species.
placentomes	Placental junctures consisting of the uterine caruncle and the placental cotyledon, which permits vascular transport of nutrients into and waste out of the fetal environment.
ploidy	Degree of repetition of the basic number of chromosomes.
pluripotent	Capable of differentiating into more than one cell type.
polar body	A small cell containing little cytoplasm that is the by-product of oöcyte meiosis in female animals.
polycythemia	An increase in the total red cell mass of the blood.
polymorphism	Describes a substance that can take on several different forms. Can refer to subtle differences in DNA sequences among individuals. It also may refer to a protein which can be coded by several different sequences; these variations do not ruin the protein's function.
polyploidy	The state of a cell having more than two times the haploid number of chromosomes in its nucleus.
portal	Anatomical nomenclature pertaining to an opening, especially the site of entrance to an organ of the blood vessels and other structures supplying or draining it.
predation	The capturing and consumption of prey as a means of maintaining life.
pregnancy toxemia	A pathologic metabolic disturbance of pregnancy that results when fetal carbohydrate or energy demand exceeds the maternal supply during the last trimester of pregnancy. Specific to sheep and goats.
preimplantation	A period very early in embryo development, before the embryo attaches to the uterus.
progeny	An animal derived from sexual reproduction that has at least one cloned animal as a parent (but could result from two cloned animals mating).
promoter	A sequence of the DNA molecule to which RNA polymerase will bind and initiate transcription.

promoter	A segment of DNA acting as a controlling element in the expression of a gene.
promoter-enhancer sequence	A control element that can increase expression of a gene.
pronucleus	The pronucleus is the structure that contains the haploid genome of the sperm or ovum after fertilization occurs, but before they fuse to make the nucleus of the zygote, or the single-celled diploid organism.
p-value	A measure of the probability that a difference between groups during an experiment happened by chance.
recumbancy	Lying down.
rendering	Reducing, converting, or melting down animal by-products by heating; a cooking and drying process that yields fat of varying grades, both edible and inedible (depending on raw material source), and animal protein that is useful for animal feeds and fertilizer.
risk	A set of conditions that links an exposure to the likelihood of an adverse outcome.
risk assessment	The methodology used to characterize potential risks and the conditions that result in the potential to experience risk.
risk management	The set of activities applied to identify and evaluate alternative strategies (often regulatory), and select among them on the basis of economic, political, scientific, ethical and social conditions or criteria.
RNA	Abbreviation for ribonucleic acid that serves to carry information from DNA to other parts of the cell or that has other functions. The generation of messenger RNA is a critical step in gene expression.
RNA polymerase	An enzyme that transcribes the information in a DNA sequence into RNA.
ruminant	Animals having a rumen - a large digestive sac in which fibrous plant material is fermented by commensal microbes, prior to its digestion in a "true" stomach (the <i>abomasum</i>). Common farm ruminants are cattle and sheep.
SCNT	Acronym for Somatic Cell Nuclear Transfer. The process of generating a live organism asexually by transferring the diploid

	nucleus of a somatic cell from a donor animal to the enucleated embryo of a recipient animal.
scours	Severe diarrhea in farm animals.
senescence	The process or condition of growing old in which cells, tissues, and organisms deteriorate and finally die.
sequellae	Morbid conditions occurring as a consequence of another condition or event.
sexual reproduction	The production of offspring by the fusion of male and female gametes (in contrast to 'asexual reproduction').
somatic cell	Any cell of an organism other than a germ cell.
stem cell	A totipotent or pluripotent cell that can replicate indefinitely and which can differentiate into other cells; stem cells serve as a continuous source of new cells.
stochastic	Pertaining to a random process, used particularly to refer to a time series of random variables. Arrived at by skillful conjecture; <i>e.g.</i> a stochastic model, a stochastic process.
superovulate	To produce numerous ova at one time.
telomerase	A DNA polymerase enzyme that maintains the structure of the telomere by adding the required repetitive sequences to the ends of eukaryotic chromosomes.
telomere	The structure that seals the end of a chromosome.
tetraploid	An organism or cell containing four haploid sets of chromosomes (see polyploidy).
totipotent	Capable of becoming any cell type in the body.
transcription	The process by which a single-stranded RNA with a base sequence complementary to one strand of a double-stranded DNA is synthesized.
transgenic	Contains heritable DNA from another source. A transgenic animal is one that has been intentionally altered using molecular biology techniques that result in heritable changes (insertions, deletions or rearrangements) in the nucleic acid sequence of the nucleus or mitochondria, and includes any offspring that inherit those changes.

translation	The second major step of gene expression in which the particular sequence of bases in the transcribed mRNA determines the sequence of amino acids in the proteins (or polypeptides) being synthesized (see transcription).
transposable element	A genetic element that has the ability to move (transpose) from one site on a chromosome to another.
trophectoderm	The group of cells in the blastocyst that form the placenta and other non-fetal tissues.
trophoblast	A layer of extra-embryonic ectodermal tissue on the outside of the blastocyst. It attaches the blastocyst to the endometrium of the uterine wall and supplies nutrition to the embryo.
urachus	A structure through which a fetus excretes urinary waste. In normal development, this structure would close at the time of parturition.
ventricle (ventriculus)	A small cavity or chamber within a body or organ, especially: (a) the chamber on the left side of the heart that receives oxygenated arterial blood from the left atrium and contracts to force it into the aorta; and (b) the chamber on the right side of the heart that receives deoxygenated venous blood from the right atrium and forces it into the pulmonary artery.
villi	Microscopic vascular protrusions from the surface of a membrane.
wild type	The phenotype that is characteristic of most of the members of a species occurring naturally and contrasting with the phenotype of a mutant.
xist	Enzyme that deactivates one of the two X chromosomes in female embryos.
zona pellucida	The thick, transparent, non-cellular outer layer surrounding an oöcyte and fertilized ovum.
zygote	The diploid cell that results from the union of a sperm cell and an egg cell.

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Appendix A:

Risk and Safety Assessment Primer for Animal Cloning

Appendix A: Risk and Safety Assessment Primer for Animal Cloning

A. How has Risk Assessment Evolved?

Although the overall process of dividing risk assessment into operational steps has been altered to address the nature of the substances or processes being evaluated, the fundamental components of the risk assessment process have remained relatively constant. Thus, for any particular etiologic (causative) agent or process,

- (1) the universe of potential outcomes that may be causally associated with exposure are identified and characterized;
- (2) the relationships between exposure and outcome are described;
- (3) estimates of potential exposure are made; and then,
- (4) the qualitative and quantitative (when available) components are integrated into an estimate of the likelihood of the potential outcomes to occur given that exposure also occurs.

Because information for decision-making is often incomplete, risk characterization also must take into account the degree of uncertainty associated with any of the steps in the overall process, as well as the cumulative contribution(s) that such uncertainties may make to the overall risk estimate.

At various times, the National Academy of Sciences (NAS) has attempted to describe risk analysis in different ways (Table A-1). The 1983 NAS report “*Risk Assessment in the Federal Government*,” first attempted to consolidate the risk assessment procedures practiced in the US regulatory agencies (primarily FDA’s Bureau of Foods, which subsequently became the Center for Food Safety and Applied Nutrition) into four coherent steps. At that time, these steps were appropriate to the nature of the substances on which risk assessments were performed *e.g.*, radiation and chemical carcinogens.

Chief among the shared characteristics of these substances was the ability to describe dose in discrete units, allowing for the relative precision of exposure and dose-response estimates. By the time of the publication of the NAS’s 2002 report “*Animal Biotechnology: Science-Based*

Concerns” (NAS 2002b), the description of the risk assessment process had evolved to be more accurately suited for the potential risks associated with animal biotechnology. The most important differences reflect the change of etiologic agents from radiation and chemicals to biological agents or processes. These differences are most obviously manifested in the hazard assessment and dose-response sections, where the range of potential adverse outcomes (harms) can differ in kind from radiation and chemical damage, and the concept of dose must accommodate biological potential. Biological potential can be thought of as the ability for the substance or organism being evaluated to either grow, replicate, die, or perform a catalytic function so that dose is no longer a constant (or possibly decreasing) amount.

Table A-1: Risk Analysis Steps as Described by the National Academy of Sciences	
1983 “Red Book”	2002 Animal Biotechnology Report
<ul style="list-style-type: none"> ○ Hazard Identification ○ Exposure Assessment ○ Dose Response Evaluation ○ Risk Characterization 	<ul style="list-style-type: none"> ○ Identify potential harms ○ Identify potential hazards that might produce those harms ○ Define what exposure means and the likelihood of exposure ○ Quantify the likelihood of harm given that exposure has occurred

B. Thinking About Risk

Qualitatively, risk may be thought of as some function of the combination of exposure and the intrinsic properties of the substance or process under consideration by linking an exposure to the likelihood of an outcome. The “risk equation” was first derived for the condition of carcinogen exposure and written as:

$$\text{Risk} = (\text{exposure}) \times (\text{potency})$$

where potency was estimated from an evaluation of the relationship between exposure and outcome (*i.e.*, the dose-response evaluation). More generally, however, the risk equation is best thought of as some function of exposure and some function of the biological properties of the agent causing the outcome:

$$\text{Risk} \propto f_{\text{outcome}}(\text{exposure}, \text{hazard})$$

In cancer risk assessment, the function of outcomes was often referred to as the “cancer potency” and was derived from the slope of the dose-response curve for tumor formation. For animal

cloning, outcomes may be thought of as the adverse health effects resulting from cloning such as Large Offspring Syndrome, or for edible products of clones, a lack of expected nutritional content of milk from animal clones.

Thinking about risk from the perspective of an “equation” is useful, even when performing qualitative analyses, because it allows the equation to be “solved” for any of the variables that have been defined. Often we ask the “forward” or prospective question: given that some process or exposure has occurred, what is the likelihood of a particular outcome (*e.g.*, how likely is exposure to a particular contaminant in milk to cause gastrointestinal distress?). Alternatively, the question can be asked in the “backwards” or retrograde form: given that an outcome has occurred, what etiologic agent under which exposure conditions is responsible for that outcome (*e.g.*, given gastrointestinal distress, did consumption of milk contaminated with x amount of y substance cause that effect? or how much of x do you have to consume before gastrointestinal distress is experienced?).

When performing a risk analysis, it is critically important to distinguish between a *hazard* and the potential *risk(s)* that may result from exposure. A *hazard* can be defined as an act or phenomenon that has the potential to produce an adverse outcome, injury, or some sort of loss or detriment. These are sometimes referred to as *harms*, and are often identified under laboratory conditions designed to maximize the opportunity to detect adverse outcomes. Thus, such observational summaries are often referred to as “*hazard identification*” or “*hazard characterization*.” Risk, as previously discussed, is the conditional probability that estimates the probability of harm given that exposure has occurred. In a qualitative assessment such as this, however, risks can be discussed only within a qualitative context, and no quantitative interpretations should be made.

Another important question to consider is who experiences the risk. At its inception, risk assessment tended to be anthropomorphic; all risks were evaluated in the human sphere, and were expressed in units of the individual, that is, the probability of a person being exposed to a hazard and experiencing a harm over a lifetime. That individual was defined as the *receptor*. Human risks could also be expressed at the population level, or the probability of x individuals in the population experiencing the harm. For animal cloning issues, the receptor can be considered to be the surrogate dam carrying a fetal clone, the animal clone itself, or humans or other animals consuming edible products of clones (*e.g.*, milk and meat).

C. How Do We Think About Safety?

For purposes of the Draft Risk Assessment, *safety* may be best thought of as the condition under which risks would be considered unlikely, rather than the condition of no risk (as such conditions do not exist for any scenario). It implies that a risk analysis has been performed, and the “risk equation” is solved for the condition that Risk \square 0 (i.e., the conditions under which risk approaches zero). When considering food from animal clones, this risk assessment has approached the issue of safety from a comparative perspective. Because one of the basic questions that the food consumption portion of this risk assessment asks is whether animal clones are materially different from their conventional counterparts, the risk question that is asked is whether edible products from animal clones or their progeny pose an increased risk relative to the same products from conventional comparators. Likewise, for animal safety, the question that is asked is whether animals involved in the cloning process are at greater risk for any adverse outcome relative to other assisted reproductive technologies.

One of the difficulties with any safety assessment is “proving the negative.” Because in practice the universe of conditions under which some risk may be encountered cannot be explored, there are always some conditions under which the null hypothesis (i.e., exposure to y μ g/liter of Substance X will pose no significant risk) will not be disproved. Thus, a careful risk/safety assessment defines the boundaries of its investigation and expresses its conclusions within those particular limits (i.e., clones born after a carefully monitored pregnancy under closely supervised conditions are at a slightly increased risk of dying than animals derived via *in vitro* fertilization, or artificial insemination, or, for food safety, milk from dairy cow clones that meets existing regulatory standards and is not significantly different from Grade A bulk tank milk is as safe to drink as milk meeting existing regulatory standards from Grade A bulk tank milk derived from non-clone dairy cows).

Appendix B:

Overall Reproductive Efficiency and Health Statistics for US Animal Agriculture

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Overall Reproductive Efficiency and Health Statistics for US Animal Agriculture

In order to gain a better understanding of the animal safety issues associated with SCNT, it is helpful to review statistics on animal health and reproduction under current agricultural practices. This section draws data from reports published by USDA/APHIS National Animal Health Monitoring System (NAHMS at <http://www.aphis.usda.gov/vs/ceah/ncahs/nahms/index.htm>) and the National Agricultural Statistics Service (NASS at <http://www.nass.usda.gov/census/census02/volume1/us/index1.htm>).

A. Dairy cattle

The Dairy 2007 Part I: Reference of Dairy Health and Management Practices in the United States report (USDA/NAHMS 2007) surveyed a total of 2,194 dairy operations in the United States. According to the USDA/NASS census, in 2002, there were 9,109,600 milking dairy cows in the United States. The predominant breed of dairy cattle in the US is Holstein, comprising 93.4 percent of the national herd. The next most popular breed is the Jersey, comprising about 3.6 percent of dairy cattle in the US. Other “colored” breeds (Guernsey, Brown Swiss, Ayrshire, and others) make up the remaining minority, and numbers for these breeds are more variable. Individual dairies vary in size from fewer than 100 to as many as 10,000 cows. Due to the variability in size of dairies, the USDA/NAHMS report broke dairies down into three groups: fewer than 100 cows, 100-499 cows, and greater than 500 cows. The report does not supply statistics for individual breeds of dairy cattle.

According to the USDA/NAHMS 2007 report, the most commonly reported causes of cow illness for all operations were clinical mastitis, lameness and infertility problems (failure to conceive by 150 days postpartum). Incidence of retained placenta was also a commonly reported problem (7.8 ± 0.2 percent), and may have contributed to incidence of reproductive problems. Incidence of clinical mastitis was similar across operations, and averaged 16.5 percent of all cows. Table B-1 presents data on causes and incidence rate of illness for operations responding to the survey.

Table B-1: Most commonly reported health problems contributing to morbidity, mortality and culling of US dairy cattle			
Cause	% morbidity¹	% mortality²	% culled³
Clinical mastitis/udder problems	16.5 ± 0.5	16.5 ± 0.7	23.0 ± 0.6
Lameness	14.0 ± 0.4	20.0 ± 0.8	16.0 ± 0.4
Reproductive			
Infertility	12.9 ± 0.3		26.3 ± 0.7 ⁵
Retained fetal membranes	7.8 ± 0.2		
Other (dystocia, metritis)	4.6 ± 0.3	15.2 ± 0.7 ⁴	
¹ Expressed as percentage of all cows ± standard deviation of the mean. ² Expressed as percentage of cows dying ± standard deviation of the mean. ³ Expressed as percentage of cows culled ± standard deviation of the mean. ⁴ Mortality attributed to dystocia ⁵ Culling for all reproductive problems			

The percent of dairy cows dying in 2002 was 5.7 ± 0.1 percent, and did not differ by size of operation. The most frequently reported causes of death for all dairy cows in this report were lameness or injury (20.0 ± 0.8 percent), mastitis (16.5 ± 0.7 percent), and difficult labor, also known as dystocia (15.2 ± 0.7 percent).

Mastitis may cause death by acute toxicity, or cows may be euthanized as a result of severe or persistent mastitis caused by treatment-resistant pathogens such as *Staphylococcus aureus* or *Mycoplasma* species. The percent of cows culled for mastitis or other udder problems in 2007 was 23.0 ± 0.6 percent for all cows culled, and represented one of the most common reasons for culling. Culling due to reproductive problems was an equally common reason given by producers in this report (26.3 ± 0.7 percent of all cows culled), with poor production not due to illness as the next most common reason (16.1 ± 0.7 percent of all cows culled). On average, 23.6 ± 0.3 percent of dairy cows were culled in 2002, with culling rate slightly higher on smaller dairies (24.1 ± 0.6 percent for herds with fewer than 100 cows) compared to medium dairies (23.7 ± 0.5 percent for dairies with 100 to 499 cows) and 23.4 ± 0.7 percent for large operations (more than 500 cows).

Mortality for dairy cattle varied by age, with unweaned heifers having the highest death rate (8.7 ± 0.2) and weaned heifers having the lowest death rate (1.9 ± 0.1 percent). Smaller operations appeared to have a higher death loss among unweaned heifers compared to operations with more than 500 milking cows (9.1 ± 0.4 and 9.4 ± 0.3 percent for operations with less than 100 cows and between 100 and 500 cows, vs. 7.7 ± 0.5 percent for operations with greater than 500 cows).

Table B-2 presents data on causes of death and incidence rate for weaned and unweaned heifers for all operations responding to the survey.

Cause	Unweaned	Weaned
Diarrhea	56.5 ± 1.3 ¹	12.6 ± 1.0
Respiratory	22.5 ± 0.9	46.5 ± 1.7
Dystocia	5.3 ± 0.7	NA

¹ Percentage of deaths ± standard deviation of the mean.

In the US, most dairy cattle are bred by AI, although many dairies still maintain bulls for cows that do not conceive to AI. According to the USDA/NAHMS 2007 report, 51.7 percent of dairies surveyed maintained one or more bulls. Embryo transfer has been promoted as a commercially feasible assisted reproductive technology (ART) for dairy cattle, particularly for dairies interested in using their best cows to improve herd genetics (Webb and Drost 1992). Embryos are also sold nationally and internationally to increase genetic advancement and overall herd production. The International Embryo Transfer Society, a professional society whose membership includes breeders and researchers, estimates that a total of approximately 670,711 *in vivo* derived bovine embryos were transferred worldwide in 2006 (Thibier 2007). Cows with less desirable genetics or production levels may be used as recipients of higher genetic merit embryos. However, ET is not a predominant means of reproduction in dairy cattle. *In vitro* fertilization has been less successful than *in vivo* fertilized ET, and is not commonly practiced. The developmental competence of cultured bovine embryos remains low (Betts and King 2001), with less than half of bovine IVF embryos developing to blastocysts, and even fewer survive to attachment in the uterus.

In cows bred by AI, pregnancy may be diagnosed by ultrasound 35 days after insemination or by palpation approximately 40 to 45 days after insemination. Average pregnancy loss following a positive pregnancy diagnosis for all cows across operations of different sizes was 4.5 ± 0.2 percent. Pregnancy loss was highest on larger operations (5.3 ± 0.3 percent for operations with greater than 500 head; 3.7 ± 0.1 percent for operations with less than 100 head; 3.7 ± 0.3 percent for operations with 100 to 499 head) (USDA/NAHMS 2007).

B. Beef Cattle

Beef cattle in the US are managed under various systems, depending on the intended use of the animals. Beef cattle destined for slaughter may change hands several times before final disposition. Breeding stock and young nursing animals may be maintained on range or in pasture. These are generally referred to as “cow-calf” operations. Following weaning, animals destined for slaughter may go directly to feedlots or may be maintained for a brief period on high quality pasture, a stage referred to as “back-grounding” or “stocker.” In the US, most cattle are slaughtered between 15 to 18 months of age.

The 1997 Beef Cow-Calf Health and Health Management Practices report (USDA/NAHMS) surveyed 2,713 beef cow-calf operations throughout the United States, representing an estimated 34,280,000 head of cattle. According to the survey, approximately 1.5 ± 0.1 percent of breeding cattle, including weaned replacement heifers, cows and bulls, died or were euthanized due to various causes in the previous year. Mortality rate was higher on small operations with less than 50 cattle, compared with larger herds (2.4 ± 0.3 percent). Approximately 20 percent of these losses were due to unknown causes. The largest single category (27 percent) of losses for beef breeding cattle was “other known” causes, most of which producers attributed to old age. The next highest categories (after “unknown”) were weather (18.0 percent) and calving problems (17.0 percent). Table B-3 presents the leading known causes of death, where a specific cause was named, for cattle that died.

Table B-3: Causes of death for beef breeding cattle (cows, bulls and weaned replacement heifers) that died	
Cause	Percent \pm SE
Digestive	6.1 ± 0.1
Respiratory	6.0 ± 1.0
Weather	18.0 ± 3.9
Dystocia	17.0 ± 1.9

Relatively few breeding females in cow-calf herds experienced health problems, according to the 1997 survey. In general, replacement heifers experienced a higher percentage of illnesses compared to mature cows. Pinkeye was the most commonly reported illness, and occurred in 1.3 percent of female breeding cattle. With the exception of pinkeye, illness rates for breeding females appeared fairly similar among herds of different sizes. Pinkeye incidence was reported highest in small herds (less than 50 head, 2.3 percent) than in large herds (more than 300 head 0.6 percent). There was no difference in incidence rate of retained placenta or uterine infections between small and larger operations (0.2 ± 0.0 percent for operations with less than 50 or more

than 300 head). Incidence of pregnancy loss was also small and not significantly different between breeding females in different sized herds (0.2 ± 0.1 percent in herds with less than 50 head; 0.3 ± 0.0 percent in herds with greater than 300 head). Major causes of health problems in breeding female beef cattle are listed in Table B-4.

Conditions	Replacement Heifers	Cows	All Females
Respiratory Disease	0.9 ± 0.3^2	0.3 ± 0.0	0.4 ± 0.1
Scours	1.0 ± 0.2	0.4 ± 0.1	0.5 ± 0.1
Pinkeye	1.9 ± 0.4	1.2 ± 0.1	1.3 ± 0.1
Cancer eye	0.0 ± 0.0	0.3 ± 0.0	0.2 ± 0.0
Foot rot	0.8 ± 0.2	0.8 ± 0.1	0.8 ± 0.1
Mastitis	N/A	0.2 ± 0.0	0.2 ± 0.0
Retained placenta/metritis	N/A	0.4 ± 0.0	0.3 ± 0.0
Spontaneous abortion	0.3 ± 0.1	0.3 ± 0.0	0.3 ± 0.0
Neurologic problems	0.0 ± 0.0	0.1 ± 0.0	0.1 ± 0.0

¹ Expressed as average percentage of all breeding females in 1996 cattle inventory.
² Percentage of females by category \pm SE.

Average mortality rate of unweaned calves was approximately 3.4 ± 0.1 percent of all calves born during 1996, and there were no appreciable differences among operations of different sizes for calf mortality. Two of the most common causes of death, when death could be attributed to a cause, were respiratory problems and dystocia. The leading causes of calf mortality according to producers surveyed are expressed in Table B-5.

Cause	Percent \pm SE
Digestive	14.4 ± 1.0
Respiratory	16.3 ± 1.2
Weather	20.2 ± 1.4
Dystocia	13.9 ± 1.3
Unknown	17.5 ± 1.4

The leading cause of morbidity in calves was scours (diarrhea) affecting 2.4 ± 0.2 percent of all calves three weeks old or younger. Older, but still unweaned calves had a slightly lower

incidence of scours (1.7 ± 0.2 percent). Diarrhea, in part, may have contributed to death losses due to digestive problems. Causes of morbidity in unweaned calves are listed in Table B-6.

Table B-6: Causes of morbidity in unweaned calves¹		
Cause	3 Weeks or Less	Over 3 Weeks Old
Respiratory	0.5 ± 0.1^2	0.8 ± 0.1
Scours	2.4 ± 0.2	1.7 ± 0.2
Pinkeye	0.1 ± 0.0	1.1 ± 0.1
Foot rot	N/A	0.2 ± 0.0
¹ Based on all calves in survey.		
² Mean Percentage \pm standard error.		

C. Swine

The total US swine population was estimated at 59,848,000 head in 2000 (USDA/NAHMS 2001). Most US swine operations are fully integrated. This means that swine remain on the same operation under the same general management throughout their lives. Animals are usually maintained under full confinement in highly biosecure facilities, to minimize disease transmission and for other economic and management reasons. Sows generally farrow (give birth) twice a year. Piglets remain with their dams for approximately 21 days, and then are weaned and moved to a nursery, where they are housed in small groups in raised pens for 6 to 8 weeks. They progress through “grower” and “finisher” phases, depending on weight, and are generally maintained in the same groups throughout the process.

The most complete survey of swine health and management practices in the United States was published in 2001. This section derived data from Part I: Reference of Swine Health and Management in the United States, 2000 and Part II: Reference of Swine Health and Health Management in the United States, 2000 (USDA/NAHMS 2001). A total of 2499 producers were surveyed for the report. In order to qualify for the report, operations must have had at least 100 head of swine at the time of the survey.

A total of 3.3 ± 0.1 percent of all breeding females died and 17.5 ± 0.7 percent were culled between December 1999 and May 2000. The most common reasons cited for culling were age, lameness, performance and reproductive failure. Measures of poor performance in this survey included small litter size, high pre-weaning mortality and low birth rate. Other reasons for culling included upgrading herd genetics, poor body condition and liquidation of the breeding

herd for financial reasons. Table B-7 presents the reasons for culling and the relative percentages of swine culled for those reasons.

Cause	Percent of culled females \pm SE	Percent of all females \pm SE
Age	41.9 \pm 1.8	7.3 \pm 0.4
Lameness	16.0 \pm 1.2	2.8 \pm 0.3
Performance	12.0 \pm 0.7	2.1 \pm 0.1
Reproductive failure	21.3 \pm 1.3	3.7 \pm 0.2
Other	8.8 \pm 1.6	1.6 \pm 0.3

The two most commonly reported health problems in breeding females were roundworms (an intestinal parasite) and Porcine Reproductive and Respiratory Syndrome (PRRS). Swine dysentery was the only health problem more commonly reported on small operations (less than 250 swine) compared to large operations. Other diseases occurred at a higher rate on larger operations. Unfortunately, no data were presented to indicate number or percent of animals affected by disease. Problems at farrowing and other reproductive problems were not reported.

D. Sheep

Most sheep in the US are raised for the production of both wool and meat. In the Eastern US, most sheep are raised on farms in fenced pasture, and may be supplemented with grain. In the Western US, it is more common to maintain sheep on open range. Lambs are generally born in late winter or early spring. Age at slaughter is variable, depending on the price of lamb compared to the price of grain and other inputs.

The National Agricultural Statistics Service (NASS), USDA reported 66,100 sheep operations with a total national herd of 6,965,000 head as of February 2002. The 2001 Reference of Sheep Management in the United States (USDA/NAHMS) reported that 23.8 \pm 1.0 percent of rams and 18.3 \pm 0.5 percent of ewes in all flocks were culled in 2000, and 5.0 \pm 0.1 percent of all sheep and lambs died. Sheep raised on farms had a marginally higher death loss compared to open or fenced range sheep (5.6 vs. 4.5 and 4.7 percent, respectively). Data on culling rates by type of operation were not available. Table B-8 presents primary reasons for culling by sex for animals culled in 2000.

Table B-8: Primary reasons to cull for all rams and ewes culled		
Reason for Culling	Rams	Ewes
Age	47.7 ± 2.1 ¹	47.9 ± 1.8 ²
Teeth problems	0.8 ± 0.3	5.3 ± 0.5
Poor mothering	N/A	3.3 ± 0.3
Mastitis	N/A	3.3 ± 0.2
Failure to lamb	N/A	5.5 ± 0.4
Ram breeding soundness	13.8 ± 1.4	N/A
Other reproductive	3.6 ± 1.1	1.2 ± 0.4
¹ Percent of all culled rams ± SE		
² Percent of all culled ewes ± SE		

Predators (23.5 ± 1.0 percent), dystocia (12.3 ± 0.5 percent) and old age (15.4 ± 0.8 percent) accounted for 51.2 percent of all adult sheep that died or were lost in 2000. Other problems included respiratory disease, other diseases, digestive and metabolic problems (including milk fever and pregnancy toxemia), poisoning/toxicity, weather, and theft. Table B-9 presents data on major causes of death for adult sheep and lambs that died in 2000.

Table B-9. Causes of death for adult sheep that died in 2000		
Cause	Sheep	Lambs
Predators	23.5 ± 1.0 ¹	44.1 ± 1.1 ²
Digestive	6.7 ± 0.6	9.9 ± 0.6
Respiratory	7.0 ± 0.8	11.7 ± 0.7
Metabolic	3.7 ± 0.4	1.0 ± 0.1
Dystocia	12.3 ± 0.5	NR ³
Other disease	3.0 ± 0.2	2.0 ± 0.3
¹ Based on percent of all sheep that died ± SE		
² Based on percent of all lambs that died ± SE		
³ Not reported		

As for swine, incidence and causes of morbidity in sheep was presented as percentage of operations reporting the problem. Data on number or percent of animals affected by illness were not presented in the USDA/NAHMS report. The most commonly reported health problems were stomach or intestinal parasites, clostridial infection, contagious ecthyma (sore mouth), and foot rot. Respiratory and reproductive problems were not reported as causes of illness in sheep or lambs in this report.

E. Goats

Statistics on goat production in the US were not available through USDA/NAHMS. According to the Agriculture Databases for Decision Support (ADDS), there are approximately 2 to 4 million goats raised in the US (http://www.adds.org/CGI-BIN/om_isapi.dll?clientID=23885&infobase=National%20Goat%20Database&softpage=Browse_Frame_Pg). However, no reliable or comprehensive statistics on goat numbers or their production in the US could be found. Goats are generally divided into three distinct types for meat, dairy or fiber (mohair or cashmere) production. Goats grown for meat or fiber are raised predominantly in large herds on open range, while dairy goats are raised in smaller herds on limited acreage with grain feeding. Intestinal parasites and respiratory diseases appear to be the most common illnesses reported in goats, although actual data were not available (ADDS Goat Handbook 1993).

Appendix C:

Comparison of Outcomes Among Assisted Reproductive Technologies (ARTs)

Appendix C: Comparisons of Outcomes Among Assisted Reproductive Technologies (ARTs)

Although there have been several studies comparing the outcomes of somatic cell nuclear transfer (SCNT) with various other assisted reproductive technologies, it is important to note that most of these evaluated data once other technologies had matured and were well-integrated into agricultural practice. The following summary provides an overview of several studies comparing the outcomes of four key ARTs. Comparison of success rates from SCNT with these ARTs may not be entirely appropriate due to the relative newness of SCNT technology. However, a review of the available studies indicates a trend of increasing adverse outcomes with increasing technological assistance; specifically, the increased rate of pregnancy failure, late gestational complications and problems associated with Large Offspring Syndrome (LOS) are most commonly associated with *in vitro* manipulation of the embryo. Table C-1 presents outcomes noted in various studies of artificial insemination (AI), *in vivo* produced embryo transfer (ET), *in vitro* produced embryos (IVP), blastomere nuclear transfer (BNT), and SCNT.

Table C-1. Outcomes noted among studies for various ART in cattle, swine and sheep.					
Developmental Node¹	Gestational Period	ART	Outcome	Reference	Comments
Node 1	Early conceptus, early embryo prior to completion of organogenesis (gd 42 in cattle)	IVP, BNT	Higher rate of embryonic death than AI or <i>in vivo</i> produced embryos	Reichenbach et al. 1992; Kruip and den Daas 1997; Wells et al. 1998; Hasler 2000;	Cattle and sheep
		IVP	Pregnancy loss following transfer of IVP or <i>in vivo</i> produced embryos prior to gd ² 21 or within 2 weeks of transfer	Farin and Farin 1995 McMillan et al. 1998	
		IVP	Increased total length of conceptus from IVP embryos 2X that of <i>in vivo</i> produced at gd 12 and 17	Farin et al. 2001 Lazzari et al. 2002	

Developmental Node ¹	Gestational Period	ART	Outcome	Reference	Comments
		IVP	Gd 16 IVP conceptuses shorter than <i>in vivo</i>	Bertolini et al 2002	Likely reflects survival status during critical time of maternal recognition
		IVP	19% of conceptuses from IVP blastocysts degenerated by gd 17	Farin et al. 2001	
		IVP	Altered embryonic disc morphology affected by culture medium for IVP embryos	Fischer-Brown et al. 2005	
		IVP	Pregnancy rates 45% or higher in dams receiving IVP embryos	Hasler 2000; van Wagtendonk-de Leeuw et al. 2000; Lane et al 2003	Factors affecting outcome include embryo culture system, embryo quality, embryo evaluator, number of embryos implanted, synchrony with dam's estrus cycle, fresh vs. frozen embryos
		ET	Higher embryo survival rate when embryos are transferred fresh rather than frozen-thawed	Spell et al. 2001	
		AI	Embryo loss ~30% by gd 30 in beef and dairy cattle	Smith et al. 1982; Sreenan and Diskin 1983; Dunne et al. 2000; Santos et al. 2004	
		AI	Embryo loss by gd 21 associated with high plasma estrogen levels on day of insemination	Shore et al. 1998	Possible estrogenic effect of legume in diet
Node 1	Late embryonic/early fetal period (days 30-90)	ET, IVP	Embryo loss for <i>in vivo</i> produced embryos < 5% (from 2 months – term)	King et al. 1985 Hasler et al. 1987	
		IVP	Embryo loss for IVP embryos higher <ul style="list-style-type: none"> - 13% after gd 40 - 10.7-13.1% - 24% total between gd 53-calving, with more between gd 50-80 	Hasler et al. 1995; Agca et al. 1998; Hasler 2000; Block et al. 2003	Depending on medium

Developmental Node ¹	Gestational Period	ART	Outcome	Reference	Comments
		AI, ET, IVP	Pregnancy rates at gd 22 not different among groups. At gd 42, pregnancy rates similar between AI and ET, but increased embryo loss in IVP compared to AI and ET	Drost et al. 1999	
		IVP	Abnormal development of allantoic membranes and cavity in placentae of IVP embryos gd 30-90	Peterson et al. 2000	Abnormal placental development and reduced placental blood membrane development
		IVP	Abnormal placentome and blood vessel morphology between gd 70-222	Miles et al. 2004 Miles et al. 2005	
		IVP	Gd 61 and older fetuses heavier than ET fetuses; altered fetal organ growth; excessive amniotic fluid	Sinclair et al. 1999	
		IVP	Gd 70 altered angiogenesis and placental morphometry; modified synthetic oviductal medium (mSOF) compared with medium with serum had fewer placentomes, low placental fluid volume and lower fetal weight: placental weight ratio; Placentomes (cotyledon tissue) had decreased density of blood vessels, decreased expression of angiogenic factor mRNA and vascular endothelial growth factor (VEGF)	Miles et al. 2005 Farin et al. 2006	
		AI	Fetal loss by gd 44 30-40% in swine pregnancies	Vonnahme et al. 2002	Fetal survival related to placental efficiency

Developmental Node ¹	Gestational Period	ART	Outcome	Reference	Comments
		AI	Embryonic/fetal loss varies from 10 to 20% between gd 28 and 80 in beef and dairy cattle	Pope and Hodgson-Jones 1975; Kummerfeld et al. 1978; Bulman and Lamming 1979; Lucy 2001	Progesterone levels in dams' milk may be normal through first 30 days of pregnancy, followed by sudden drop
		AI	Embryonic/fetal loss 11 to 44% by gd 50 in beef cattle	Bulman 1979	Attributed to bull
Node 1	Late gestation	ET, IVP	Compensation in vascular beds of IVP bovine embryos; Compared with <i>in vivo</i> embryos, IVP had decreased fetal villi, binucleate cell volume densities in placentomes. Proportional volume of blood vessels in maternal caruncles increased in IVP group. Ratio of blood vessel volume density: placentome surface area increased.	Miles et al. 2004	Theorized to compensate for increased fetal size and need for increased nutrients and gas exchanges, but increased vascular blood network at level of placentome
		IVP	IVP fetuses show increased glucose and fructose in fetal plasma levels; increased placental surface area	Bertolini et al. 2004	
		IVP	Hydroallantois frequency in IVP pregnancies (1/200) higher than in "normal" pregnancies (1/7,500)	Hasler et al. 1995	
		IVP, SCNT	Pregnancy loss higher in SCNT than IVP embryos; 50-100% loss gd 30-60; placentae hypoplastic and reduced cotyledonary development	Hill et al. 2000; Chavette-Palmer et al. 2002; Heyman et al. 2002; Edwards et al. 2003; Lee RS et al. 2004	

Developmental Node ¹	Gestational Period	ART	Outcome	Reference	Comments
		BNT	Late gestation abortions, stillbirths, underdeveloped fetuses for gestational age, edema, hydronephrosis, testicular hypoplasia, skull and heart malformations; lack of udder development in dams	Wells et al. 1998	
		IVP, SCNT	Broad distribution of fetal and neonatal body weights for both IVP and SCNT-derived embryos; shifted to “heavy” relative to <i>in vivo</i> embryos	Wilson et al. 1995 Kruip and de Daas 1997 Farin et al. 2001 Miles et al. 2005	Two competing explanations: (1) “normal” for these animals may be heavier than for <i>in vivo</i> produced embryos, or (2) a proportion of animals shifts weight distribution of population
			Adaptation to small changes in biochemical parameters and morphology	Sangild et al. 2000	
		IVP, ET	Increased gestation length, dystocia, perinatal mortality, fetal edema, altered organ development, abnormal limbs	Kruip and den Daas 1997; Behboodi et al. 1995; van Wagendonk-de Leeuw et al. 1998; Farin et al. 2001; Bertolini and Anderson 2002; Edwards et al. 2003; Rerat et al. 2005	Frequency and severity of abnormalities: IVP>ET>AI
		AI	55.9% of abortions due to infection	Santos et al. 2004	
Node 2	Perinatal	IVP, BNT	Increased birth weight, Increased crown-rump length; increased mortality and physical deformities	Behboodi et al. 1995; Wilson et al. 1995; Walker et al. 1996; Rerat et al. 2005	
		IVP	Perinatal mortality in IVP ranges from 2.4-17.9%, due to dystocia associated with large fetuses	Hasler et al. 1995; van Wagendonk-de Leeuw et al. 1998; Block et al. 2003	Lower in heifers than in cows

Developmental Node ¹	Gestational Period	ART	Outcome	Reference	Comments
		IVP, SCNT	IVP and SCNT fetuses have cerebellar hypoplasia, respiratory distress, and heart enlargement	Schmidt et al. 1996; van Wagendonk-de Leeuw et al. 2000; Chavette-Palmer et al. 2002	
		IVP	Altered expression of mRNA for non-imprinted myostatin and glyceraldehydes-3-phosphate in IVP fetuses	Crosier et al. 2002	
		IVP, SCNT	Altered expression of mRNA or protein in IVP and SCNT placentae for VEGF, peroxisome proliferators activated receptor γ , leptin, bovine placental lactogen, transforming growth factor (TGF) β 1, 2, 3, TGF- β receptor, major histocompatibility class I antigens	Davies et al. 2004; Miles et al. 2004; Ravelich et al 2004; Miles et al 2005; Ravelich et al. 2005	
		IVP, SCNT	Expression of demethylating enzymes DMT 1, 3a altered in IVP and SCNT preimplantation embryos	Wrenzycki et al. 2004	
		BNT	Birth weight range 26.4 to 67.3 kg; slow to stand, poor suckling behavior, flexor tendon deformities, hypoxemia, hypoglycemia, acidosis, hypothermia; altered metabolic hormones (thyroxine, triiodothyronine, and insulin)	Garry et al. 1996	
		BNT	Calving rate ~50% using high quality embryos; some very large calves (up to 70.5 kg); contracture of limbs and spine, cardiac and skull deformities noted in a few calves; high rate of dystocia (52/100); hydroallantois observed in four cows	Willadsen et al. 1991	

Developmental Node ¹	Gestational Period	ART	Outcome	Reference	Comments
		IVP, SCNT	Increased incidence of dystocia and C-section deliveries for IVP pregnancies compared to AI/NM; lack of contractility and other signs of labor in ewes; higher mortality among IVP and SCNT compared to AI/NM	Ptak et al. 2002	Sheep
		AI	Heat stress reduces birth weight and passive transfer of immunity and results in low IgG concentration in calves	Collier et al. 1982	High levels of glucocorticoids accelerates “gut closure”
Nodes 2-3	Postnatal	IVP	Increased feed intake and growth rate	Rerat et al. 2005	
		IVP	Altered glucose and electrolyte metabolism compared to AI persisting through early juvenile period	Rerat et al. 2005	
¹ For the purposes of this table, Developmental Node 1 is divided into three stages of pregnancy: early embryo, late embryo-early fetal, and late gestation. ² Gd= gestation day or day of pregnancy.					

A. Successes and Failures of AI, IVP, and ET

Success of AI depends on a variety of factors, including health of the female and timing of insemination relative to ovulation. In dairy cattle, conception rates to AI following spontaneous estrus have declined from approximately 55 percent in the 1950s to 45 percent in the late 1990s. The use of hormones to synchronize estrus for timed AI has further reduced conception rates to approximately 35 percent. The reasons for this apparent reduction in dairy cow fertility are not clear, although a number of factors have been cited as possibly contributing to the phenomenon, including increased milk production (resulting in increased stress and reduced availability of nutrients for reproductive function), increased average herd size (resulting in fewer person-hours spent observing cows for estrus behavior), nutrition, herd health, inbreeding, and environmental pollution (Lucy 2001). Embryo loss has been estimated to occur at a rate of 10 to 20 percent in dairy cattle (Lucy 2001) and as high as 30 percent in beef cattle (Dunne et al. 2000), and generally occurs prior to 30 days gestation. Fetal losses in swine pregnancies can be as high as 40 percent following AI (Vonnahme et al. 2002). The reasons for these losses *in utero* are not

always apparent. Lucy (2001) noted that embryo loss may occur even in cases where the developing embryo appeared normal. However, in swine, fetal loss appears to be related to the size and efficiency of the placenta (Vonnahme et al. 2002).

Betts and King (2001) noted that the developmental competence (an embryo's ability to progress through normal cell division and development) of IVP and cultured embryos was low. Using *in vitro* procedures (as published up to 2001), less than half of inseminated bovine oocytes reached blastocyst stage, and of those that did, many did not implant or attach following transfer. Betts and King (2001) noted that chromosomal abnormalities such as aneuploidy and polyploidy played a fundamental role in most of these embryonic deaths.

The evolution of IVP technology in cattle can be observed by comparing early studies (conducted prior to 2002) with more recent publications. Studies using IVP embryos during the mid- to late 1990s (Behboodi et al. 1995; Farin and Farin 1995; Hasler et al. 1995; Walker et al. 1996; Drost et al. 1999; Sinclair et al. 1999) noted relatively high rates of embryo loss and LOS among fetuses and neonatal calves. In contrast, several more recent studies using IVP embryos have indicated few or no problems (Chavatte-Palmer et al. 2002; Heyman et al. 2002; Bertolini et al. 2004; Rerat et al. 2004). However, high embryonic mortality and placental abnormalities may still be observed with IVP in some labs (Miles et al. 2004; Miles et al. 2005).

Embryo transfer, in which oocytes are fertilized *in utero* then removed and transferred to surrogates, has become a commercially viable technology (See Chapter II), and is generally more successful than IVP. In a study by Drost et al. (1999), initial pregnancy rates, as determined by blood progesterone levels at gestation day 22, were similar among cows bred by AI, ET or IVP. However, by gestation day 42, embryo loss among cows receiving IVP embryos was higher than either AI or ET, while pregnancy rate was similar between cows bred by AI compared to those receiving ET embryos. The success of ET may be affected by treatment of the embryo prior to transfer and synchrony between the surrogate and the embryo donor (Pope 1988; Spell et al. 2001). According to Spell et al. (2001) fresh embryos had a higher rate of survival than embryos that had been frozen then thawed prior to transfer. Embryo survival was also higher when surrogates had been in estrus within 12 hours of the embryo donor (Spell et al. 2001).

In order to follow fates of client-owned pregnant cows carrying IVP-derived pregnancies in a commercial ET operation, Hasler et al. 1995 noted that for the first 100 transfers, 24 ended in pregnancy loss before 100 days of gestation. The success rate improved the subsequent year, however, with only 7 percent of IVP-derived pregnancies spontaneously aborting. They compared these results to 5.3 percent of ET pregnancies aborted between two and seven months of gestation in an earlier study.

In a comparison of AI and IVP, Behboodi and coworkers (1995) noted an increased incidence of dystocia and Cesarean sections (C-section) for IVP derived pregnancies compared to AI in a small group of cattle (8/13 IVP-derived pregnancies vs. 7/71 AI pregnancies requiring C-section). Birth weights of calves derived from IVP embryos were higher than calves produced by AI, likely contributing to the observed increase in dystocia among dams carrying IVP-derived pregnancies. Sinclair et al. (1999) also observed large IVP-derived fetuses with altered development and excessive amounts of amniotic fluid. In that study, nine of 13 fetuses (69 percent) derived from embryos co-cultured with granulosa cells (a type of cell found in the ovary) and one of six embryos (17 percent) incubated in synthetic oviductal fluid (SOF) plus steer serum were oversized, while embryos that had been incubated with SOF alone produced normal sized fetuses. Bovine embryos cultured for three to five days post-fertilization also were associated with increased dystocia due to oversized calves in a study by Walker et al. (1996). (See discussion of influence of culture conditions on success rates in Chapter IV).

Farin and Farin (1995) collected bovine IVP and ET fetuses from beef heifers at seven months gestation and compared development between the two groups. Fetuses from the IVP group were heavier than their ET counterparts (18.6 ± 1.1 vs. 15.4 ± 0.8 kg), had greater heart girths (56.5 ± 1.2 vs. 52.4 ± 0.9 cm) and weights (139.7 ± 8.3 vs. 116.2 ± 5.8 g), and greater long bone lengths (23.1 ± 0.6 vs. 21.3 ± 0.4 cm). When organ and skeletal measures were compared on a per kilogram body weight basis, however, IVP fetuses had consistently smaller skeletal measures than ET fetuses. Internal organ weights per unit of body weight were not different between the two groups of calves. The authors concluded that IVP fetuses were undergoing abnormal and disproportionate development compared to ET fetuses. It should be noted that the most rapid period of prenatal growth in cattle is during the last two months of gestation (months 8 and 9) (NRC 2001), which would have occurred after these pregnancies were terminated.

Young and Fairburn (2000) noted that both IVP and embryo culture have resulted in abnormal phenotypes, including up to two-fold increases in birth weight (LOS), excess amniotic fluid, hydrops fetalis¹¹⁶, altered allometric organ growth¹¹⁷, advanced fetal development, placental and skeletal defects, immunological defects, and increased perinatal death.

Markette et al. (reviewed by Farin et al. 2001) observed that 54.7 percent of ET recipients were pregnant at 60 days gestation, with the majority of pregnancies lost prior to day 24 of gestation.

¹¹⁶ Accumulation of fluid in the entire body of the newborn.

¹¹⁷ Allometric growth refers to differences in the rate of growth of a particular organ or part in relation to the rest of the organism. An example of normal allometric growth is the legs of a newborn foal (horse) in proportion to its body size; the legs are long and out of proportion to the rest of the body. As the foal ages, the body grows (and fills out) more rapidly than the legs, so that in adult horses the legs appear proportional to the rest of the body. Altered allometric growth in the context of ARTs has resulted in enlarged hearts and undersized kidneys, as well as other organs, which are not appropriately proportioned to the rest of the body.

In a large study, King et al. (1985) reported that the incidence of pregnancy loss in 1,776 embryo transfer recipients was 3.15 percent from 2 to 3 months of gestation, and 2.14 percent between 3 to 7 months. These mid- and late-gestation spontaneous abortions were not influenced by embryo age, embryo quality, time between embryo collection and transfer, asynchrony of recipient with donor estrus, donor age, ovarian response to gonadotropin treatment, or whether or not the donor had a history of infertility, according to the authors. In most studies, pregnancy loss during the fetal period (day 42 to 280 of gestation in cattle) was greater following transfer of embryos produced *in vitro* than that for embryos produced *in vivo*. Mid- to late-gestation spontaneous abortion of about 7 to 13 percent has been reported for recipient cattle carrying fetuses derived from IVP embryos, and in some studies pregnancy loss has been considerably higher (Farin et al. 2001).

Conversely, Bertolini et al. (2004) compared fetal development in *in vivo* and IVP cattle pregnancies and reported no significant difference between groups for pregnancy rates (20/53 and 36/112 for control and IVP groups respectively) and fetal losses after day 45 (2/20 and 3/36 for control and IVP groups respectively). They did report that fetal losses between gestation days 30 and 44 were 3.4-fold higher ($P < 0.05$) in the IVP group (17/36) than in controls (4/20). Also in contrast to earlier studies, Bertolini et al. (2004) reported that their measurements of conceptus physical traits for both *in vivo* produced controls and IVP pregnancies on days 90 and 180 demonstrated allometric proportionality between fetal body size and body weight with no physical deformities observed in any fetus.

In a review of research on early embryo development, Gardner and Lane (2005) stated that the environment of the preimplantation embryo has a profound effect on the physiology and viability of the conceptus. Among the many factors that can influence development of IVP embryos, they cite the use of serum products as an important contributor to developmental abnormalities in cultured embryos. These authors state: "*Mammalian embryos are never exposed to serum in vivo...Rather, serum is a pathological fluid, the composition of which is greatly undefined and varies enormously with source...serum induces premature blastulation in domestic animal embryos...affects embryo morphology...and leads to perturbations in ultrastructure...and energy metabolism.*" Other factors that may influence development of embryos *in vitro* include ammonia, oxygen, inadequate nutrients, and freezing (Gardner and Lane 2005).

Rerat et al. (2004) compared the perinatal health characteristics of IVP and AI cattle and observed no differences in post-natal mortality or viability. Calves in this study were generally healthy with the health status of IVP calves at birth and during the first 112 days of life similar to that of AI calves. Clinical traits such as heart rate, rectal temperature, and respiratory rate were nearly identical in both groups. At birth, measurements indicative of growth performance such as potassium, 3,5,3'-triiodothyronine (a metabolic hormone), and thyroxine concentrations were

lower in IVP than in AI calves. Postnatally, IVP calves had a faster growth rate than AI calves under conditions of identical nutrient intake.

Sakaguchi et al. (2002) induced twinning in Japanese beef cows by transferring one or two *in vivo* fertilized embryos into AI bred cows. Fetal dystocia occurred in 7 of 14 twin parturitions, in which some twin calves appeared to enter the uterine cervix at the same time, but no single parturition was accompanied by dystocia. The incidence of retained placenta was significantly higher in the twin parturitions (10/14; 71 percent) than in the single parturitions (2/22; 9 percent). These complications are known to occur with natural twins in cattle, however, and may not be directly related to ET technology. The incidence of retained placenta in healthy, single calf-bearing dairy cows is approximately 5-15 percent, (slightly lower in beef cows) and is increased when there are twins. The expected incidence of dystocia is 10-15 percent in first-parity animals, and 3-5 percent in mature cows (Merck Veterinary Manual Online 2002).

B. Outcomes for BNT, Fetal- and Adult-Cell SCNT

Although success rates for various types of cloning have improved, they are still highly variable across studies. In earlier studies, generally less than 10 percent of all NT embryos transferred to recipients were born alive (Wells et al. 1999). Some of these early studies noted that both blastomere and somatic cell NT clones appeared to have the same low success rate and exhibited many of the same problems, such as poor or dysfunctional placentation and LOS (Stice et al. 1996; Wells et al. 1998). Stice et al. (1996) reported that no fetuses derived from BNT survived beyond day 60 of gestation. Wells et al. (1998) reported a 64 to 80 percent pregnancy loss during the attachment phase for clone fetuses derived from an embryonic sheep cell line, while a further 43 percent of pregnancies were lost in the last trimester, such that 11 percent of embryos survived to term (12/112). In contrast, Le Bourhis et al. (1998) reported 9/30 transferred male bovine BNT clones developed to calving, while 6/27 female BNT clones resulted in live calves. Heyman et al. (2002) compared development and survival of BNT, fetal and adult NT clones to IVP-derived embryos under the same culture conditions. Pregnancy loss from 90 days of gestation to calving were 43.7 percent for adult and 33.3 percent for fetal SCNT, compared to 4.3 percent for BNT clones, while none of the IVP-derived pregnancies were lost. Pace et al. (2002) reported 75 percent pregnancy loss of adult (some transgenic) SCNT embryos throughout pregnancy.

Results from these studies may reflect the evolution of NT technology over time. Embryonic or BNT cloning and IVP success rates appear to have improved. Although losses remain high for the newer SCNT technology, success rates for this technology also have improved over time. It remains to be seen what progress may be made in further reducing pregnancy loss and other risks associated with SCNT.

C. Conclusions regarding outcomes for ARTs

Based on the studies reviewed, there appears to be a general trend indicating that the frequency of embryo/fetal loss and abnormal pregnancy outcomes increases with increasing manipulation of the embryo and *in vitro* culture. This trend is evident, even when maturity of the technology is considered. Causes of embryo/fetal loss are not always evident, but late gestational complications (hydrops and dystocia) and fetal/neonatal abnormalities (skeletal and organ deformities, oversize, metabolic alterations) have all been noted in ET, IVP, BNT and SCNT. The frequency of these outcomes varies somewhat among laboratories, but has the general trend ET<IVP<BNT<SCNT. These data support a conclusion that SCNT falls on a continuum of ARTs, and that the adverse outcomes noted with SCNT are not unique, but are of concern due to their increased frequency.

Appendix D:
Transgenic Clones

Appendix D

Transgenic Clones

A. Issues

The current risk assessment is limited to address the risk of clones from non-transgenic cells. Although not within the purview of this analysis, the results of a number of studies that address either transgenic animal clones or transgenic and nontransgenic animal clones (Hill et al. 1999; the “ACT series” including Cibelli et al. 1998, Lanza et al. 2000, and Lanza et al. 2001 for cattle; McCreath et al. 2000 and Denning et al. 2001 in sheep; Baguisi et al. 1999 and Keefer et al. 2001a in goats; Carter et al. 2002, Lai et al. 2002, and Lee GS et al. 2003 in swine) are presented here to clarify the relative utility of such studies for assessing potential risk(s) associated with non-transgenic somatic cell nuclear transfer (SCNT).

Many reviews of SCNT outcomes cite these papers as indicative of the severity of adverse outcomes associated with cloning, or a demonstration of the positive outcomes that can come from cloning. Hill et al. (1999), in particular, is often cited as the seminal “adverse outcome” paper for cloning. On the other hand, if only the “final report” paper of the ACT series, Lanza et al. 2001, were read, it would not be possible to know that the cells from which the cattle were cloned were indeed transgenic. This distinction only becomes apparent when the earlier papers are also reviewed. Most recently, headlines were generated when Pearson (2003) reported on “sudden death syndrome” in pig clones; CVM reviewed the paper that describes their generation and noted that these pig clones carried two distinct transgenes (Lee GS et al. 2003).

Because these animals are transgenic clones, it is not possible to determine whether adverse outcomes result from the direct effect of the expression of the transgenic construct, pleiotropic effects resulting from insertion of the construct, the SCNT process, or some interaction of any or all of these processes. For example in a comparison of 34 cell lines, Forsberg et al. (2002) reported that when otherwise similar cells are used as donors for SCNT, those that are transgenic (for a selectable marker gene) result in lower pregnancy initiation (22 percent vs. 32 percent) and calving (3.4 percent vs. 8.9 percent) rates. The authors hypothesize that the additional culturing required to generate transgenic cells, selection of transgenic lines, or the DNA construct itself, could be responsible for the lower rates.

CVM thus assumes that transgenic clones occupy a different “risk space” from “just clones.” Conversely, an argument can be made that if no adverse outcomes are detected, then for these animals, neither process sufficiently perturbed development to induce anomalies. We have

included those studies in the overall risk assessments if such results were obtained. Nonetheless, because these studies occupy such a large segment of the cited literature, a few are presented here to illustrate the range of responses noted, with the appropriate caveats for interpretation.

B. Cattle

Hill et al. 1999

Hill et al. 1999 reports on a group of 13 transgenic clones of a Holstein bull. Twelve Brangus cows carrying 13 fetuses cloned from Holstein cells were originally included in the study, although three of these transgenic clone fetuses died prior to the perinatal period (defined in this paper as two weeks prior to anticipated delivery and a few days thereafter), and one cow aborted at eight months of gestation. Two cows developed hydroallantois and were delivered by C-section; four others were also delivered by this method due to subjective judgment regarding fetal size. The two remaining pregnancies delivered vaginally. Birth weights of the transgenic clones ranged from 44-58.6 kg (average Holstein male calf weight is in the range of 40-50 kg), and cited as within the weight range of *in vitro* produced embryos. Five of the eight live born clones were judged to be normal within four hours of birth based on clinical signs and blood gas measurements. Three of the eight were immediately diagnosed with neonatal respiratory distress. One of these calves died from pulmonary hypertension, pulmonary surfactant deficiency, and elevated systemic venous pressure at day 4. The other three animals recovered. Two of the five fetuses that did not survive to birth also exhibited signs of pulmonary hypertension and placental edema at necropsy. Another clone died at 6 weeks of age with signs of respiratory distress; subsequent field necropsy suggested dilated cardiomyopathy, although no definitive diagnosis could be made.

The Advanced Cell Technology Series

The series of papers from the Advanced Cell Technology group (Cibelli et al. 1998, Lanza et al. 2000, and Lanza et al. 2001) on the health of clones are similar to that of Hill et al. (1999) in that the animals presented are clones that are derived from transgenic cells. Interpretation of any adverse outcomes is thus also confounded by the potential role of the transgene and its insertion.

The results of these studies are summarized in Lanza et al. (2001), in a short overview with accompanying supporting documentation provided by the journal in electronic form. Of 30 fetal transgenic clones that developed to term, 24 were reported healthy at 1-4 years of age, but five died within 7 days of cardiopulmonary difficulties that the authors speculated were secondary to placental insufficiencies. The sixth animal died at day 149 due to enteric disease, lymphadenopathy, and exhibited mild placental edema and high fever at birth. Problems observed at birth included placental edema, including edematous cotyledons (attachment sites of the placenta to the uterus), labored breathing, froth and fluid in the lungs, pulmonary edema,

pneumonia, high fever, septicemia, lethargy, abdominal distention, masses in the abdomen, liver damage due to hypoxia, and heart abnormalities.

Birth weights of the survivors were reported as 45 ± 2 kg (this paper cites normal as 43 kg). An unspecified number exhibited pulmonary hypertension and respiratory distress at birth. Presumably, they received supportive care at that time. Another unspecified number were also reported as experiencing fever following vaccination. This is not an atypical response among calves receiving vaccinations, as stimulating a potent immune response is likely to produce at least a mild local and systemic (fever) reaction in the animals (Roth 1999).

Physical and veterinary examination of surviving animals aged 1-3 years were reported as normal and included temperature, pulse, and respiratory rate. No abnormalities were detected in general appearance, on auscultation (listening to breathing, heart beat, and digestive sounds), and behavior appeared normal. Puberty onset was reported to occur at the expected time, and fertility appeared to be normal. At the time of publication (2001), two of the animals had delivered apparently normal progeny.

Clinical chemistry parameters evaluated for these animals included electrolytes, urea, creatinine, glucose, bilirubin, aspartate aminotransferase (AAT), sorbitol dehydrogenase (SGT), albumin, globulin, and total protein. Globulin and total protein measurements were reported in the publication as “slightly below normal.” All other measurements were reported to be within normal range. Hemograms (analysis of cellular components of blood) were all reported as normal: hematocrit, hemoglobin, red blood cells, mean red cell volume, mean red cell hemoglobin concentration, and white blood cell numbers and differentials were within normal ranges. Blood gases were also within normal ranges. To examine immunocompetence in the clones, peripheral blood lymphocytes from the transgenic clones and conventional Holsteins were compared to determine whether the same ratio of cell surface markers were present, and if the transgenic clone cells responded to mitogen challenge in the same way as cells from conventional Holsteins. No significant differences were observed between the cell surface markers or cellular responses of cells from conventional animals or clones.

In the early spring of 2003, an interview of an ACT executive reported in the lay press indicated that two animals from this cohort had developed significant health problems. One animal was reported to have developed a tumor, and the other was diagnosed as having neurological problems. The first animal apparently died during surgery to remove the tumor, and no further information is available on the potential causes of the tumor. The second animal was later diagnosed as being positive for Johne’s disease (*Mycobacterium paratuberculosis*), an infectious, chronic, progressive disease that often presents with chronic diarrhea and eventual cachexia (general physical wasting and malnutrition). It is therefore unlikely that this animal’s symptoms

were due to either cloning or transgenesis. We are unaware of any other adverse outcomes associated with these animals.

C. Swine

Carter et al. 2002

Carter et al. 2002 reported on the overall health status of transgenic swine clones produced from cells transfected with green fluorescent protein (GFP). The 10 transgenic piglet clones from three litters were followed for the first six months of life.

Five of the ten transgenic swine clones died or were euthanized during the study. Two piglets died of congestive heart failure at 7 and 35 days of age, two others died from bacterial infections at 3 and 116 days of age. The fifth animal died at 130 days of age, following a history of chronic diarrhea, decreased growth and vitamin E deficiency. The remaining five piglets were reported as healthy and growing similarly to conventional animals housed in the same facility at the conclusion of the study. Behavior was reported as “consistent with pigs of their age group.”

Average birth weight of the transgenic clones (1,312 g) was similar to average birth weights of conventional piglets from similar genetic background (1,450 g). Average daily weight gain for transgenic clones through the first 16 weeks was (461 g) relative to the herd average (594 g), which the authors considered as within the normal range.

Some of the piglets displayed physical defects. These included two piglets with contracture of the flexor tendons, another piglet with five digits on a forelimb (four digits are normal) and an enlarged dewclaw. Another piglet with low birth weight was described as having short legs and a large, round chest.

Hematology and blood clinical chemistry data were collected beginning at 2 days of age and every two to four weeks until 24 weeks of age. Most hematological variables were similar to the comparator group, except for hemoglobin, hematocrit, and plasma total protein. Mild anemia and low blood protein concentration were observed for the first four weeks, but both these conditions resolved by eight weeks of age. The authors stated that decreased hematocrit and hemoglobin values are common in piglets reared in confinement, and that these symptoms are generally treated with iron dextran. Similarly, clinical chemistry results indicated decreased levels of albumin and globulin during the first four weeks in the transgenic clones relative to comparators, but these values were back within the normal range by eight weeks of age. The authors attributed the decreased protein and globulin values to the decreased colostrum intake of the newborns as the surrogate sow bearing them did not initiate normal lactation, and piglets were dosed with colostrum at some unspecified point after birth.

Seven of the transgenic clones were evaluated for cardiac function. Although no physical defects were found, one piglet had evidence of mitral insufficiency (a condition in which the mitral valve of the heart does not close all the way during contraction, resulting in regurgitation of some of the blood in the left ventricle), and dilation of the left atrium and ventricle. This piglet and two other clones had reduced cardiac output values compared to control piglets, but did not display clinical signs of cardiac disease. Although similar cardiac abnormalities have been noted in conventional swine, the incidence is reported to be very low (Carter citing Hsu et al. 1982). These developmental defects appear to be similar to those noted in cattle clones (see Critical Biological Systems discussions).

Lai et al. 2002

This study was reported in a brief communication, and a limited amount of data was presented. Piglets were generated from cell lines (derived from inbred miniature pigs) in which the α -1-3-galactosyltransferase gene was interrupted by the insertion of a gene sequence in order to create α -1-3-galactosyltransferase “knock-outs.” The α -1-3-galactosyltransferase gene codes for a protein that causes hyperacute rejection of swine organs when transplanted into primates. “Knocking out” the expression of this gene increases the suitability of these animals to be used as donors of organs for human transplant patients.

Six piglets were born from two litters. All but one of the piglets had low birth weights compared to the breed average (115 to 650 g vs. 860 g). One piglet from each litter died shortly after birth from what the authors termed “respiratory distress syndrome.” A third piglet died at 17 days of age during a routine blood draw, and was diagnosed at necropsy with a dilated right ventricle and thickening of the heart wall. Other abnormalities noted in these surviving transgenic piglets included flexor tendon deformities in three animals; abdominal ascites, enlarged right ventricle, pulmonary hypertension in one animal; and ocular defects and lack of patent ear canals in another animal. The authors attributed these abnormalities to failures in reprogramming during the SCNT process rather than the genetic engineering process, as they did not see a consistent phenotype across the piglets.

Lee GS et al. (2003)

Recently, Pearson (2003) reported that the University of Connecticut laboratory that had generated four transgenic swine clones had announced that the three (of four) surviving piglets died suddenly of heart failure at less than six months of age. The fourth piglet died at three days due to infection and abnormal spine development (Lee GS et al. 2003). Because of the transgenic nature of the animals (they carried genes for human clotting factor IX and porcine lactoferrin, an iron transport protein found in blood), it is not possible to attribute the deaths solely to cloning. It

is unknown whether any cardiac abnormalities were detected in these animals prior to their deaths, or if any measurements of cardiac function were made.

D. Sheep

Denning et al. 2001

Denning et al. 2001 were unsuccessful in producing viable knock-out sheep lacking either the α -(1,3)-galactosyl transferase (GGTA1) or the prion protein (PrP) gene using gene targeted fetal fibroblasts and SCNT. Reconstructed embryos were either incubated for six days (n=48) or overnight (n=93) in synthetic oviductal fluid with bovine serum albumin (concentration not specified). Embryos incubated overnight *in vitro* were then embedded in 1 percent agar chips in phosphate buffered saline and transferred to the ligated oviduct of an estrus-synchronized ewe for six days. A total of 120 morula or blastocyst stage embryos were transferred to 78 estrus-synchronized Finn Dorset ewes as final recipients. It is not clear from this paper how many of the transferred embryos had been incubated *in vitro*. Although 39 pregnancies were diagnosed at gd 35, only eight were maintained to term, resulting in four live births. Three of the four live-born lambs died shortly after birth. The fourth lamb survived 12 days before it was euthanized after developing dyspnea (difficulty breathing) due to pulmonary hypertension and right-sided heart failure. The authors attributed the abnormalities observed to the nuclear transfer procedure, as they were similar to results obtained with non-transgenic NT lambs.

McCreath et al. 2000

McCreath et al. 2000 inserted a promoter-less neomycin selectable marker between the ovine α 1(I)-procollagen translational stop and polyadenylation signal¹¹⁸ in male and female ovine fetal fibroblast cultures. Four transgenic female fibroblast cultures were selected as nuclear sources for SCNT, due to their vigor and normal chromosome number. A total of 80 morula and blastocyst stage embryos were transferred to recipient ewes. No description of post-fusion incubation or estrous cycle status of recipient ewes was provided in this report. Fourteen lambs were born alive; seven of these lambs died within 30 hours of birth. Four more lambs died between 3 days and 12 weeks of age. Three lambs survived and were described as thriving at one year of age. Necropsy of lambs that died *in utero* or after birth revealed a number of abnormalities including a high incidence of kidney defects (frequently renal pelvis dilation) and liver and brain abnormalities (not specified). The authors attribute these abnormalities to either cell treatment or the NT procedure, because the necropsy findings were similar to a previous nuclear transfer study using the same cell lines.

¹¹⁸ These are regions of the DNA construct that provide instructions for the appropriate processing of information in order to make functional proteins.

E. Goats

Baguisi et al. 1999

In this study from Genzyme Transgenics, six cell lines were established from 35- and 40-day old fetuses that resulted from the mating of a transgenic buck (carrying a human antithrombin III (hAT) gene with a goat β -casein promoter) to a non-transgenic doe. This study differs slightly from several other transgenic cloning studies reported here, in which the gene was inserted into the cell lines before the cultures were established. Clone embryos were cultured on goat oviduct epithelial cells for 48 hours (2-16 cell stage) before being transferred to estrus synchronized recipient does. Although overall cloning efficiency was low (3/112 embryos transferred resulted in live births), all pregnancy losses occurred prior to 60 days of pregnancy. There were no stillbirths and no abnormalities observed in the live-born kids. Kids weighed between 2.35 and 3.5 kg, within the normal birth weight for dairy goats, and are reported as healthy.

Keefer et al. 2001a

In this study from the Nexia Biotechnologies laboratory, goat fetal fibroblasts were transfected¹¹⁹ with green fluorescent protein (eGFP) and neomycin resistance genes. These are commonly used as markers to demonstrate that transgenes have been inserted and are being expressed. Twenty seven NT embryos were produced with the transfected cells, and an additional 70 non-transgenic NT embryos were constructed and transferred into 13 estrus synchronized recipient does. The authors did not specify how many embryos (transgenic or non-transgenic) were transferred to each doe. Five non-transgenic male clones and one transgenic female clone were born alive. Three of the non-transgenic clones died of bacterial infections, but the single female transgenic clone lived and showed no signs of abnormalities. The kids were all within the normal birth weight range (1.5 to 3.1 kg) for goats at that facility, and no abnormalities were observed in the placentae.

Behboodi et al. 2004

The authors compared development of embryos cultured with oviductal cells *in vitro* vs. embryos cultured *in vivo*. Embryos were constructed using skin fibroblasts of transgenic goats. Only embryos cultured *in vivo* resulted in pregnancies. Two of these pregnancies were lost early in gestation (after 30 days gestation), and four other pregnancies were carried to term. Two surrogate does delivered stillborn kids 2-3 days after their due dates; the other two does delivered healthy kids (one per each doe) at term. The two live clones weighed 3.8 and 4.1 kg at birth, and were within the normal birth weight range for their breed (Saanen). Clones were weaned at 8 weeks of age, and had similar growth rates compared to age-matched AI derived Saanen kids born at the same facility (14.5 and 18.1 kg for clones vs. 14.88 ± 1.98 kg for AI comparators).

¹¹⁹ Transfection is the process of introducing exogenous DNA into cells without the use of viral vectors. Common methods include co-precipitating DNA with salts and polymers.

Pathology on the dead fetuses indicated diffuse atelectasis (lung collapse) and the presence of amniotic fluid in the lungs. No bacterial or viral cause for the deaths of these clones could be identified. The presence of amniotic fluid in the lungs suggests that the clones attempted to breathe prematurely, a sign of fetal stress which sometimes occurs around the time of birth.

Behboodi et al. 2005

The authors evaluated health, growth, reproduction and lactation in four female goat clones generated from two transgenic fetal cell lines (one cell line coding for glycosylated and the other for non-glycosylated protein). A total of seven clones were carried to term. One clone from the glycosylated group was still born with evidence that the umbilical vessel had ruptured. Two clones died at birth (one from each of the transgenic lines) after failing to breathe on their own, despite attempts at manual ventilation. Thus, two clones from each transgenic line survived to adulthood. There were no differences in birth or weaning weights among the four surviving clones or their age-matched comparators. Transgenic clones exhibited enlarged umbilical stumps (two live and one stillborn kid), “tendon laxity” (three of the four live-born clones), and minor generalized edema (number of clones affected not indicated). These conditions resolved without intervention. The four does were bred and produced nine kids, compared to five kids produced by comparators. Clones expressing the glycosylated version of the protein lactated only briefly, but the does expressing the non-glycosylated protein had normal lactation length and milk yields.

This study is the only one we encountered that presented hematology and blood clinical chemistry data for four goat clones. These data are presented in comparison to four age-matched comparators and values from the literature (Pugh 2002). It is unclear whether or not the comparators in this study were also transgenic, whether they were the same breed as the clones, or how they were generated. Hematology values were similar between clones and comparators, and all hematology values fell within the published range. For clinical chemistry, 18/24 values were not significantly different between clones and their age-matched comparators. Of the 19 clinical chemistry values for which published ranges were available, 18 of the values for clones and comparators fell within the published range. The one value out of the published range was creatine kinase (244.6 vs. 204.4 IU/L for clones and comparators). However, values between clones and comparators were not statistically different. The study does not specify the age of the goats at time of blood sampling, so it is difficult to interpret the high values for CK in these animals compared to the published range.

This study is unique among reports of goat clones because it is the first to indicate possible signs of LOS in goat clones (enlarged umbilici, failure to initiate breathing, tendon problems). It is interesting to note that similar signs have not been noted in non-transgenic goats. We should also note that clinical signs in the four surviving clones resolved, and their health, growth,

reproduction, and hematology, clinical chemistry values indicate that even these transgenic clones are apparently normal.

Landry et al. 2005

The authors reported on growth (weight gain, wither and hip height change) and endocrine profiles of two lines of transgenic goat clones. Group 1 consisted on five does carrying the AT-III gene with a β -casein promoter inserted into cells of a female Toggenburg (dairy breed). The gene inserted into the second line of goats (Group II; n=2) was not identified, but the cells used for cloning were from a female Saanen (also a dairy breed). Non-transgenic, non-clone comparators (n=7) were Boer X Spanish crossbred meat-type does (Group III). The authors did not report on overall health of the clones. One female in each group of clones died prior to the end of the study; one died due to an accident, the other due to a ruptured abomasum. Neither death appears to be related to cloning. Both groups of clones were within range for their breed for birth weight, and appeared to grow normally. Interpretation of hormone profiles (GH, IGF-I, T3, and T4) is difficult due to the fact that the clones and comparators were of different breed (purebred vs. crossbred) and type (dairy vs. meat) backgrounds. However, for most of the hormones assayed, the values for clones fell within the range of values for comparators. The one exception is insulin, which resulted in an extremely low value in blood samples of comparators, and may have been the result of difficulties with the assay.

Melican et al. 2006

With the long-term goal of producing caprine milk containing recombinant therapeutic proteins, the authors demonstrated successful reproduction and lactation in transgenic dairy goats produced by SCNT. Does were produced using primary fetal cells harvested from day 35-40 fetuses. These cells were co-transfected with DNA fragments encoding the heavy and light immunoglobulin chains of three different monoclonal antibodies, plus neomycin resistance as a selectable marker. Two transgenic does were hormonally induced to lactate at two to three months of age and produced small amounts of milk (10-20 ml per day) for 30-40 days. These does were subsequently bred and, at 18 months of age, gave birth to 6 kids (4 were transgenic). Following parturition, the 2 transgenic does produced an average of 2.2 liters of milk per day for 3-6 months. A third transgenic doe, at age 23 months, also lactated following the birth of one transgenic kid, producing 2.6 liters of milk per day for one month prior to being dried off. The milk of all 3 transgenic does contained the antibody for which they were transgenic. These results demonstrate reproductive capacity in cloned female goats, and the ability of these goats to produce normal quantities of milk following parturition.

F. Conclusions Regarding Transgenic Clones

The experience of these cohorts of transgenic clones can be summarized as follows:

- A relatively large fraction of transgenic fetal bovine clones in cohorts surviving to late gestation presents with severe and often fatal difficulties. Some of these are qualitatively similar to those observed in cattle and sheep clones that are not derived from transgenic cells. Due to the many other variables that have been altered in the generation of these animals, at this time it is not possible to attribute these abnormalities to either of the processes (cloning or transgenesis) or their combination.
- Some animals in both cattle cohorts are born with varying degrees of initial respiratory or other physiological distress. Supportive care appears to allow most of these animals to survive to adulthood, although some animals that initially survive can succumb to possible sequelae up to six weeks later.
- Animals surviving to adulthood in the ACT cohort that appear to be healthy on visual inspection also exhibit physiological values that generally fall within normal ranges. CVM is unaware of an update of the health status of the Hill et al. cohort.
- Animals in the ACT cohort surviving to reproductive maturity appear to be capable of bearing normal offspring, although it is not clear whether the offsprings' health has been examined in a rigorous manner.
- Two severe adverse outcomes have been noted for the ACT cohort. Both cloning and transgenesis can likely be ruled out as causes for one (Johne's disease) and no causal agent or process has been associated with the neoplasm found in the other.
- The appearance, behavior, and physiological function of the animals that survive suggest that even the "riskiest" set of clones (*i.e.*, transgenic clones) can develop into normally functioning animals. These results are consistent with the analysis of non-transgenic clones, and provide additional confidence that rigorous monitoring and responsible husbandry of such animals can allow for the selection of animals that are healthy.
- Abnormalities for transgenic sheep clones appear similar to reports for non-transgenic sheep and cattle clones.

- Goats appear to suffer fewer adverse effects compared to sheep and cattle. Of the reports reviewed, only one cohort exhibited clinical signs of LOS.
- Abnormalities reported for transgenic swine clones are similar to those reported for transgenic and non-transgenic cattle clones. CVM is aware of only one report in non-transgenic swine clones (Park MR et al. 2005) in which clones exhibited similar health problems; however, *in vitro* methods used in this study likely influenced the outcome of swine clones in this study.

Appendix E:
The Cyagra Dataset

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Appendix E: The Cyagra Dataset

A. Response to CVM Data Requests

The Center for Veterinary Medicine (CVM) has presented its proposed risk assessment approach at several public venues since the fall of 2002. As part of the proposed approach, we have requested that investigators engaged in cloning cattle, swine, sheep, or goats share data that they might have on the health status of these animals with the Center. The intent of the data request was to supplement the published data with unpublished data generated by the developers of these animals. We thought that data on the health status of animal clones would likely be in the hands of the private sector, which might have less impetus to publish than academic laboratories. The Center promised producers that, to the extent allowed by law, if they wished, their identities could be kept confidential by FDA, and that we would not publish the specific identity and location of the animals.

We discovered that there are very few datasets describing the health of animal clones. In general, clones are monitored closely for the first few weeks of life (or through weaning). They are often then moved from “research/hospital” facilities to “farm-like” facilities, where they are often reared with conventional animals. Most producers kept fairly cursory veterinary records unless the animal was in distress. Further, because of technical issues associated with generating successful pregnancies, only a few clones tend to be delivered at one time or from one cell line. The result is that aggregating and analyzing data becomes difficult unless publications are planned in advance.

As many of the clone producers either have primary employment as academics, or continue to maintain academic appointments, there may be data available that have not yet been shared with CVM because of the investigators’ desire not to jeopardize their ability to publish in peer reviewed journals. Because of CVM’s pledge to be fully transparent in this risk assessment, we determined that all data submitted would be made public through the risk assessment. We obtained the express permission from the submitters of data for the public release of this data. “Publishing” this information in the assessment could preclude formal publication in a peer reviewed journal, as most high quality peer reviewed journals have a policy of being the site of first publication.

As our risk assessment methodology evolved, it was presented at public fora (the Pew Initiative for Agricultural Biotechnology’s September 2002 Biotech in the Barnyard conference, the April 2003 American Registry of Professional Animal Scientists meeting in Maryland, and the FDA

Science Forum of April 2003). Subsequent discussions between clone producers and agency staff resulted in investigators returning to the field to try to collect existing data, or, in one exceptional case, to generate *de novo* data on the health status of clones. Without exception, every clone producer or investigator contacted was willing to answer questions on aspects of clone production, gestation, delivery, and care. Many have provided data or information that we have incorporated into this risk assessment or will use in future iterations. In order to issue the risk assessment in a timely manner, however, we have had to put off our analysis of some of the datasets until the next revision of the risk assessment. We are very grateful to those producers and owners who voluntarily expended significant time, effort, and in some cases, capital, to provide information to us.

B. Cyagra Dataset

One clone producer, Cyagra Inc.,¹²⁰ has been engaged in the production of cattle clones since 1999. In the late spring of 2003, Cyagra submitted an extensive database to CVM for use in the animal health component of our food consumption and the animal health risk assessments. These data were made available for CVM to use in our risk assessments with no restriction, except to protect to the extent allowed by law the identity and location of the animals, and their current owners. In order to accommodate this request, CVM issued each animal in the study a unique identification number. These numbers have been employed throughout this analysis.

Cyagra has asserted that they have provided data on all of the clones that they can trace, including those that died, or were euthanized or culled. Animals were divided into three age cohorts by Cyagra: neonates (within 24 hours), 1-6 month age cohort (between 30 and 175 days of age), and 6-18 month age cohort (187-557 days of age).

The age spread among these animal cohorts reflects key stages in physiological development of cattle. For example, digestion differs significantly among different age groups: a 2-month-old calf is just starting to use its rumen, while a 6-month-old calf is a fully developed, cud-chewing ruminant. In this case, these two calves have been grouped together even though they have substantive physiological differences, because they have more in common than, for example, a neonate and a six month old calf. For the sake of accuracy, we have classified this group as 1 to 6 months old. A 6-18 month old calf is not quite old enough to be considered an adult, as it is still growing, and the younger animals in this group will still be pre-pubertal. We have therefore decided to classify this group simply as “6 to 18 months.” The distribution of animals in the cohorts is found in Table E- 1: Distribution of Cattle Clones and Comparator Populations.

¹²⁰Cyagra Inc. is a privately held biotechnology company commercializing SCNT technology for the agricultural sector.

Comparators were approximately age-matched animals reared on the same farms or facilities as clones. The comparators were not born at the same locations, and do not represent the same distribution among breeds as the clones. Comparator animals were not clones, but were produced by either artificial insemination (AI) or natural breeding, from either primiparous (heifers) or multiparous dams, and were all delivered vaginally. Blood samples from neonatal comparator animals were taken after colostrum administration, while neonatal clones were sampled prior to receiving colostrum.

These animals do not provide a strict biological comparator that has experienced the same treatments and conditions as the clones. For example, the culturing conditions in the embryonic phase for cloned embryos could be more closely compared with those encountered by animals generated by *in vitro* fertilization. These comparators are not, strictly speaking, “control” animals.

Further, given the approximate age- and breed-matching, this dataset should not be evaluated in the same manner as a tightly controlled prospective “laboratory” experiment. Rather, our opinion is that this dataset should be viewed as an attempt to compare health and laboratory test values between clones and conventional animals comprising part of national dairy and beef herds. These data were not generated or collected under “Good Laboratory Practices,” and we have not attempted to audit the data except insofar as we have detected errors or requested clarification(s) from Cyagra.

Table E- 1: Distribution of Cattle Clones and Comparator Populations for Blood Analyses				
	Number of Animals			
	neonates	1 to 6 months	6 to 18 months	Totals
Clones	10	46	18	67
Comparators	17	47	21	83

In Table E-1, 7 (of the 46) 1-6 month clones and 2 (of the 47) 1-6 month comparators were sampled in the neonatal group. The 1 to 6 month and 6 to 18 month cohort information was collected within a relatively short time frame. These data may best be thought of as a “snapshot” view of the animals during their development, rather than a longitudinal study in which the same animals are followed over some period of time. In fact, only nine animals were sampled or examined at more than one time point (at birth and weaning), and of those, seven were clones and two were controls (Clone ID# 71, 72, 73, 78, 79, 119, and 132; Control ID# 135 and 162).

1. Description of Clones

All clones were derived from actively dividing cells from skin biopsies; recipient oocytes were obtained from commercial abattoirs. After 7 or 8 days of *in vitro* culture, morula or blastocyst stage reconstructed embryos were implanted into recipient Holstein heifers. Pregnancies were monitored closely, and with few exceptions, clones were delivered via Caesarean section (C-section) to reduce the risk associated with birth. Blood samples were drawn from the neonates prior to colostrum administration.

Table E-2 summarizes the information on samples taken from calves within the first 24 hours of birth. Some of the animals in the Cyagra dataset required some supportive care immediately after birth (*e.g.*, glucose, warming, or supplemental oxygen), and many (n=29 out 134) received umbilical surgery after birth. Enlarged umbilical vessels which do not close naturally after birth are an identified hazard for clone calves, and many of these calves received surgery to prevent complications such as umbilical infections and bleeding (see subsequent discussion on veterinary examinations and health status). This appears to be a fairly common problem in clones, and may be associated with poor placentation. However, no direct causal attribution can be made at this time to any particular developmental pathway causing the umbilical problems.

Health anomalies noted in surviving animals for which there are no additional follow-up data include diarrhea, fever, anemia, heart murmur, and slight contracture of the flexor tendons (referred to as “contracture”).

Of the 134 clones in this review, 28 were stillborn, died, or were euthanized within 48 hours of birth, leaving 106 animals (or 79 percent) alive two days after birth. At the time that data were collected on these animals (late March 2003), 67 were alive (64 percent of those surviving to 48 hours, or 50 percent of those born or delivered). Eleven (10 percent) of the animals alive at 48 hours died within approximately one and a half years later. These data are summarized in Table E-2. Of the eleven deaths between 48 hours and one and a half years later, Cyagra considers two deaths not related to cloning, and the other nine as “related, possibly related, or questionably related” to cloning. Of those fitting the “related (to some degree) to cloning” category, one was clearly a fetal developmental anomaly: flexor tendon contracture (“contracture”); three experienced difficulties with the umbilicus ultimately leading to death either via infection or adhesions; two had gastrointestinal problems with bloat or adhesions; two had circulatory problems; and one animal was euthanized for “failure to thrive.”

Animal Number	Birth Weight (kg)	Age at Death (days)	Problems Noted	Cause of Death
3	NP ¹	0	Abnormal delivery	Stillborn
6	NP	16	NP	Accident; hung in stall
11	NP	0	Abnormal delivery	Euthanized
12	NP	0	Ruptured uterus in recipient	Stillborn
13	NP	0	NP	Unknown
14	33.2	13	Contracture ² , umbilical infection	Septicemia
16	50.0	2	Slack abdomen, umbilical problems, breathing difficulties	Failure to transition to neonatal circulation
18	68.2	0	Polycystic kidneys	Stillborn
19	69.1	0	Umbilical problems, flaccid abdomen	Stillborn
20	NP	0	Abnormal development	Euthanized
23	45.5	0	Abnormal development, internal bleeding, umbilical problems	Euthanized
28	NP	0	NP	Stillborn (C-section)
29	NP	0	Abnormal development	Euthanized
31	76.8	0	Abnormal renal development	Euthanized
34	NP	0	NP	Stillborn (C-section)
43	NP	1	Diarrhea	Rotavirus
47	NP	0	NP	Stillborn (C-section)
48	54.5	0	NP	Stillborn (C-section)
49	NP	0	NP	Stillborn (C-section)
51	NP	0	Flaccid abdomen, "bulldog"	Stillborn (C-section)
52	NP	0	NP	Stillborn (Fetotomy)
54	59.1	0	Reverted to fetal circulation, cardiac, neurological problems	Euthanized
57	NP	23	Ruptured abomasum	Ruptured abomasum
63	NP	60	Loss of hair, appetite, muscle	Euthanized/failure to thrive
65	61.4	3	Lethargic	GI transit; adhesions from umbilical bleeding
66	54.6	149	Contracted tendons, recurring bloat, large umbilicus requiring surgery	Bloat/GI motility problems
68	NP	0 (2 weeks	Pericarditis	Unable to determine

Animal Number	Birth Weight (kg)	Age at Death (days)	Problems Noted	Cause of Death
		premature)		
77	NP	47	Umbilical problems	Severe contracture, unresponsive to therapy
80	NP	1	Diarrhea	Rotavirus
86	NP	0	Severe contracture, fluid filled belly	Euthanized.
92	NP	0	Depressed, pus in umbilicus	Unable to determine
95	NP	0	Severe contracture	Euthanized
97	NP	0	Severe contracture, fluid filled belly	Euthanized
105	45.5	0	Severe twisting of neck, contracture	Euthanized
107	NP	2	Hypoxemia, rapid deterioration	Euthanized
109	NP	0	Abnormal development	Euthanized
113	NP	22	Nephritis	Pyelonephritis ³ / umbilical infection
123	NP	9	Contracted front fetlocks	Pyelonephritis/ umbilical infection
125	NP	0	Severe contracture, rotation	Euthanized

¹NP = Not provided
²Contracture is a condition in which muscles have a fixed, high resistance to stretching due to fibrosis of the tissues supporting the muscles or joints, or from disorders of the muscle fibers.
³Pyelonephritis is an inflammation of the kidney due to bacterial infection.

2. Evaluations Performed

Several types of information including veterinary records, clinical chemistry measurements, hemograms,¹²¹ and urinalysis are provided in this dataset. Not every collectable data point has been provided for each animal. Some information is unavailable because use of the data in a review such as this was not anticipated at the time the data were collected. In addition, dispersal of clones to their ultimate owners limited data collection to the degree to which owners made information or animals available. Nonetheless, this is the largest collection of information on the health status of non-transgenic clones of which we are aware, and the most detailed with respect to health status and laboratory tests.

¹²¹ A hemogram is a panel of measurements characterizing the nature of the circulating blood in an animal or human.

The dataset includes information on the following:

- Breed from which donor cells were collected
- Gender of the donor
- Birth date of the clone
- Birth status (alive, stillborn)
- Birth weight
- Perinatal health status and veterinary/supportive care provided
- Health status of animals between two and twelve months of age
- Veterinary care, including treatment with drugs, surgery, or other therapeutic interventions
- Standard blood chemistry assays (Large Animal Panels)
- Assays for serum Insulin-like Growth Factor-1 (IGF-1), estradiol-17 β , amylase, cholesterol, and bile acids
- Complete blood counts (CBC) and differentials
- Standard urinalysis

Comprehensive veterinary examinations were performed by licensed cattle veterinarians. Blood samples were drawn within a few hours of birth, or at the time of veterinary examination. For CBC, blood was collected into standard EDTA-treated collection tubes; additionally, two unstained and unfixed air-dried smears were provided. For chemical analyses, whole blood was collected, allowed to clot, and the serum fraction separated by centrifugation. Laboratory analyses were all performed at the Cornell University's Animal Health Diagnostic Laboratory.

3. CVM's Analysis of Cyagra Data: Method

Our goal in evaluating the Cyagra dataset has been to determine whether extensive interrogation of the health status of the clones, including clinical chemistry and hematology, could

- (a) Distinguish clones from comparators;
- (b) Determine whether the health status of the clones was inferior to conventional animals and offer a predictor of a successful outcome; and
- (c) Determine whether any of the information indicated concerns regarding animal health or food safety.

We note that this was not a "blinded" analysis of the provided data. No attempt was made to disguise the identity of the animals, and whether they were clones or comparators. CVM personnel engaged in performing the evaluation included veterinarians, animal scientists,

toxicologists, and risk assessors, with extensive training in evaluating clinical and physiological measurements of animals traditionally consumed as food in the US.

For the overall health status of animals, the veterinary records were reviewed for notations indicating therapeutic interventions (including administration of colostrum, vaccines, dehorning, surgeries, drug therapies, etc.). Clinical and hematologic data were compared to both reference ranges provided by the testing laboratory and to the comparator animals. Additionally, laboratory values from the comparator animals were also compared to the testing laboratory to determine the degree to which the comparator group was represented by the testing laboratory's reference range (see Results). In general, urinalysis data were only used qualitatively as confirmation of outcomes noted in the clinical chemistry (e.g., glucose, BUN or creatinine levels). Table E-3 provides a summary of the analyses performed and tabulated in the Charts indicated.

Outcomes were reviewed on an analyte basis across a cohort of animals (analyte evaluation), and on a per animal basis across analytes (animal evaluation). The questions asked for each animal and analyte tested were “How many of the total animals tested exhibited values outside the comparator/testing laboratory reference range for Analyte X?” and “How many values outside the comparator/testing laboratory reference range does Animal Y exhibit?”

Table E-3. Summary of Charts Describing Comparisons			
	Clones: Reference Range	Clones: Comparators	Comparators: Reference Range
<i>Clinical Chemistry:</i> 6 to 18 months 1 to 6 months neonates	Chart 300 Chart 200 Chart 100	Chart 301 Chart 201 Chart 101	Chart 302 Chart 202 Chart 102
<i>Hematology:</i> 6 to 18 months 1 to 6 months neonates	Chart 310 Chart 210 Chart 110	Chart 311 Chart 211 Chart 111	Chart 312 Chart 212 Chart 112

The Charts are a graphical summary of CVM's analyses. For each chart, the unique identification number associated with each animal (“ID#” or “animal number”) is listed in columns horizontally across the top of the table; the analysis performed is listed in rows vertically down the side of the table. If the value being evaluated fit within the comparison range being used for that interrogation, a black rectangle was recorded in the cell corresponding to the animal column/analyte row pair (■). If the value was outside the comparator range, but judged to be not clinically relevant, a gray rectangle (■) was recorded. If the value recorded was above or below the clinically relevant range, an arrow indicating whether the value was greater or less than the

range was inserted ($\uparrow\downarrow$). Values that were considered to be so far out of range as to be physiologically incompatible with a healthy animal but unsupported by related clinical measurements were deemed artifact and labeled “X.” For example, a calf with a blood glucose level of 4 mg/dl would be comatose or dead. If the sample came from an animal that was not comatose or in distress, and there were no other related clinical measurements normally associated with abnormal blood glucose, we assumed that the measurement was an artifact. Missing values were represented by an asterisk (*).

4. CVM’s Analysis of Cyagra Data: Results

a. Comprehensive Veterinary Examinations

Comprehensive Veterinary examinations were performed on 53 clones and 2 non-clones, and included explicit evaluations of the following:

- Demeanor
- Posture
- Gait
- Body Condition
- Skin and coat
- Vocalization
- Lungs (Auscultation)
- Nerves
- Integument
- Musculo-skeletal system
- Cardiovascular system
- Oral/Pharyngeal region
- Urine
- Gastrointestinal system
- Genitals
- Neurological examination
- Peripheral lymph nodes
- Responsiveness of pupils to light
- Corneas and eyelids
- Umbilicus
- Weight
- Heart Rate
- Respiration rate
- Temperature
- Feces

The calves in this study were examined by veterinarians specializing in cattle at roughly 1-6 months of age or at 6-18 months of age. The most consistent abnormality reported for clones was umbilical surgery, often described as umbilical hernia surgery. In some instances, the records stated, “umbilicus – had surgery.” Some other comments on the umbilicus were: “had umbilical hernia surgery,” “ventral hernia,” and “1 ½” hernia,” “fluid filled mass,” “umb. stump.” In the initial submission of 58 animals, 26 animals had umbilical surgery. Other abnormalities reported included two clones with musculo-skeletal abnormalities, one with slight precocious (early) mammary development, two with harsh lung sounds, three cryptorchid (undescended testicles)

bull calves, and one with premature ventricular contractions (PVCs, a form of cardiac arrhythmia) every 5 – 10 heartbeats.

The two clones with musculo-skeletal abnormalities included a Holstein heifer (ID# 79) with thick withers, enlarged left carpus, and leg that deviated laterally, and an Angus heifer that was a dwarf tending to gastro-intestinal bloat (Clone #108). These are obvious abnormalities and the animals were culled. The calf with slight mammary development was a 4½ month old Jersey (Clone #87). This age is young for mammary development but the phenomenon sometimes occurs in conventional heifers if they are overfed. There is no notation of follow up to determine if the calf continued to develop precociously.

The two clones with harsh lung sounds were a Holstein heifer (Clone # 41) and an Angus heifer (Clone #58). Both also had umbilical surgery. A note at the bottom of the Angus heifer's exam sheet stated that the heifer "may not return home due to permanent lung damage." There is no indication as to whether this animal was culled. Three Holstein bull clones derived from the same cell line were diagnosed with a retained testicle (cryptorchid) (Clones #128, 130, 131). Although cryptorchidism is not common in bull calves, it is thought to be heritable and is seen with some regularity. Bulls exhibiting cryptorchidism would fail their breeding soundness exams, and would not be used for breeding,¹²² but would not be refused by an inspector at slaughter.

A Holstein bull calf clone (Clone #126) was diagnosed with premature ventricular contractions from a single exam, but no subsequent follow up is provided to determine whether the animal outgrew the condition or whether the animal was culled. The frequency of cardiac arrhythmias in conventional calves is unknown. Thoracic auscultation (listening to the chest with a stethoscope) or more elaborate procedures are needed to detect cardiac arrhythmias. Calves are rarely examined with thoracic auscultation unless they show signs of illness.

b. Conclusions from Veterinary Examinations

The adverse physical exam findings noted in this limited sample of clones do not present a food safety issue for several reasons. One of the precepts of this risk assessment is that animals found to have a disease or condition that would render them adulterated (e.g., unfit for food, unhealthful, unwholesome) are excluded from the food supply, as normally happens with conventional animals. Dwarf animals from conventional breeding would likely be culled

¹²² Cryptorchidism is undesirable because of its heritability, its adverse effect on fertility, and potential for the development of testicular cancer in animals living long enough to allow neoplasia to develop. From a veterinary standpoint, however, testicular neoplasia is more of an issue with companion animals, as they are generally longer-lived than farm animals.

depending on the extent of the physical abnormality. Pre-pubescent mammary development, lung sounds, cryptorchidism, and cardiac arrhythmias are not conditions that typically exclude animals from food use. If the disease process had progressed to an extent sufficiently severe to cause systemic changes (e.g., liver congestion, enlarged heart, edematous lungs), the carcass would be condemned on inspection at the slaughtering plant. In fact, all of these conditions occur in conventional animals.

With respect to animal safety, these conditions may pose some cause for concern. Our review of these data indicates that the clone cohort appears to exhibit a higher incidence of abnormalities than might be expected in a random sample of conventional calves. There is, however, an absence of data on the prevalence of these outcomes in contemporary cattle. As some of these defects (e.g., dwarfism, cryptorchidism) likely have a hereditary component, in the absence of information on the donor cattle and their individual histories, we cannot determine whether the defects result from the cloning process, the selection of the donor nucleus, or some combination of those factors. The clustering of cryptorchidism in clones from one cell line, for example, implies that heredity may indeed be a contributing factor in the appearance of that outcome. Comparison with datasets on animal health from other clone producers would be instructive in determining whether these health problems are common among clones generated by different methods and multiple cell lines.

c. **Laboratory Values: Selection of Most Appropriate Comparator**

Two comparators were available for evaluating the Clones: the Cornell Animal Health Diagnostic Laboratory (“Reference Range”) and approximately age-matched, and breed-distributed cohort of animals contemporarily reared at the same farms as the clones (“Comparator Population” or “Comparators”). The Reference Range population from the Cornell Laboratory is described as follows:

*“We establish reference intervals by collecting blood from at least 50 **adult healthy** animals. These healthy animals are obtained from a variety of sources (e.g., student- or faculty-owned). Therefore, our reference intervals are only applicable for adult animals and not young animals. Results from young animals may fall outside our reference intervals because of age-dependent changes in their analytes. For example, phosphate concentrations and alkaline phosphatase activity are higher in young animals and decrease to within reference intervals at about one year of age.”*

(<http://www.diaglab.vet.cornell.edu/clinpath/reference/>)

Follow-up conversation with the laboratory indicates that the animals used to establish the laboratory’s reference range were exclusively dairy cows, and thus do not represent the beef

breeds that are included in the Cyagra clone cohort or comparator cohorts, and may not include bulls. In addition, it is important to remember that the reference range is selected as a statistical distribution containing about 95 percent of the normal samples. As a result, as many as 5 percent of the test values will likely fall outside that range. Statistically, when numerous tests are run on the same animal, the chance of obtaining one or more results outside the “normal range” rises based on chance alone and not a disease state.

Table E-3a: Fraction of Blood Values Within Comparison Range							
Animals	Analysis	Clones: Cornell Reference Range		Clones: Comparator Population		Comparator Population: Cornell Reference Range	
		Chart	FCWR	Chart	FCWR	Chart	FCWR
Peripubertals (6-18 months)	Clinical Chemistry	300	0.75	301	0.99	302	0.73
	Hematology	310	0.73	311	0.99	312	0.71
Juveniles (1-6 months)	Clinical Chemistry	200	0.62	201	0.96	202	0.71
	Hematology	210	0.59	211	0.96	212	0.61
Neonates (<48 hours)	Clinical Chemistry	100	0.31	101	0.90	102	0.36
	Hematology	110	0.61	111	0.90	112	0.62

FWCR= Fraction contained within range of comparison, calculated by determining the number of out of range analytes of potential clinical relevance to the total number of measurements collected in each Chart.

Table E-3a provides a summary of the Charts evaluating the clinical chemistry and hematology tests performed on the Cyagra clones compared with the comparator population, and the Cornell Reference Range. In addition, the comparator population was compared to the Cornell Reference Range. First, as cautioned by the Cornell Laboratory, the Reference Range is not a good comparator for young animals. A number of the clones and comparators fall outside the Reference Range¹²³ but the similarity to the Reference Range increases with age for both clone and comparator populations. Approximately half the animals in the older cohort were less than one year of age, however, and all clones and comparators were less than two years of age. All of the animals in the older cohorts were still growing and thus do not match the laboratory reference adult cattle population well. Clearly, then the most relevant comparison for the clone cohorts in this review is the comparator population.

d. Conclusions Regarding Clone and Comparator Population Cohorts in Aggregate

¹²³ It should be noted that because the reference range represents only 95 percent of the animals used in its derivation, even comparison of the animals used for the derivation will not fit exactly within the distribution. Thus, if the reference range were expanded to include those values outside the 95 percent distribution, it is likely that the clone and comparator populations would show a higher degree of “fit” than is observed in this analysis.

Review of the degree to which the clone cohorts have laboratory values that fit within those of the comparator population cohorts indicates the following:

1. Even at birth, 90 percent (107 of 119 measurements) of the hematology values, and 90 percent (272 of 324 values) of the clinical chemistry values lie within the values of the comparator population (Table E-3, Charts E101 and E111). This is particularly instructive, considering that many of the clones required some assistance immediately after birth (no similar records were kept for the comparators, but we assume that no extraordinary measures were taken, and were informed that all comparators were born vaginally). Further, clones had blood samples drawn before colostrum administration, while the comparators had blood samples drawn after colostrum administration, but within 24 hours after birth. Colostrum consumption (quantity and quality) influences certain laboratory values (*e.g.*, globulin, total protein, GGT).
2. The 1 to 6 month age cohorts are even more similar to each other than the neonatal cohorts: both the clinical chemistry and hematology values have 96 percent and 95 percent concordance respectively (707 of 742 of the hematology measurements and 1,404 of 1,462 of the clinical chemistry measurements for clones are within the clinically relevant ranges) (Charts 201 and 211).
3. The 6 to 18 month cohorts are almost superimposable with respect to laboratory values (Charts 301 and 311). Only three of the 294 hematological values and seven of the 592 clinical chemistry measurements were outside the clinically relevant ranges, significantly less than would be expected by chance alone.

Based on clinical chemistry and hematology values, it is not possible to distinguish between these two cohorts (clones and comparators). The superimposability of the laboratory values and the absence of any significant health observations in the clones (based on the limited number of explicit veterinary exams) leads to the conclusion that the health of these animal clones during the 6-18 month period is not inferior to that of conventional animals.

Because we have concluded that the comparator group is the appropriate basis for comparison for the clones, all subsequent discussion regarding clinical and hematological values will be considered in that context.

e. Animal and Analyte Specific Analyses

In addition to evaluating the overall status of the clone and comparator cohorts, individual animal and analyte data were reviewed to determine if more detailed evaluations could provide either confirmation of the overall health of the animals, or to serve as indicators of potential health problems that might be present in the animals that were not detected on the comprehensive veterinary examinations. For each Chart, the following two questions were asked:

1. “For *all* of the clones in this age cohort, how many of the values for each analyte were out of the range established by the comparators? (*i.e.*, looking across each row, how many arrows or grey rectangles were present?), and
2. “For each clone in this cohort, how many of the analytes were out of the range established by the comparators?” (*i.e.*, looking down each column of the Chart).

There are three overall issues addressed by this evaluation:

1. Whether the laboratory values of the clones were similar to those of the comparator population on an animal-by-animal level, or whether it would be possible to distinguish between the two populations based on the clinical chemistry and hematology data. A finding of similar laboratory values would provide confidence that there were no material differences in metabolic, immunologic, and hematopoietic (blood producing) functions between clones and conventional animals;
2. Whether the clones respond to the internal (growth and maturation) and normal external (stressors, disease) environments appropriately (analyte based approach); and
3. Whether the individual values can be used to predict the long-term viability of that animal or that cohort (analyte and animal approaches combined).

A description of the parameters that were evaluated and their relation to physiological status is provided in Appendix F: The Comprehensive Veterinary Examination.

Clinical chemistry and hematology responses are best evaluated in the context of the whole animal, including its age, species, breed, husbandry, geographic location, reproductive status, and the laboratory performing the analysis. Laboratory findings complement the subjective physical diagnosis of the patient by providing objective information for the process of differential diagnosis, monitoring treatment, and formulation of a prognosis (see Appendix

F). “Abnormal” laboratory measurements and examinations are often defined as those values lying outside the limits of the reference range. Determining what constitutes “normal” is more complex than simply comparing an individual value to a reference range derived from a sample of a representative population.

Laboratory tests are designed to *support* a clinical diagnosis based on the patient’s history and clinical findings, with all of the information contributing to the final decision. Comparing clinically normal clone and comparator populations only on laboratory measurements, in the absence of disease or injury, is, at best, an exercise in attempting to identify subtle hazards that may be hypothesized to exist in animal clones. The statistical or biological significance of any value that may lie outside the comparison ranges available for this review in the absence of corroborating information on the health status of the individual animals or cohorts would be difficult to justify on scientific grounds.

Further, it is important to remember that the evaluation is only as good as the sample provided. One of the terms used to group the inconsistent outcomes that result from sample mishandling or processing errors is “artifact.” Erroneous conclusions can result if the artifact is accepted as a true sample result. An artifact is suspected when the laboratory data are inconsistent with the clinical assessment or there is an inappropriate relationship between tests. For example, low blood glucose values can indicate hypoglycemia, with the accompanying clinical signs of lethargy or seizure. However, low blood glucose values in a blood test can also result from not separating the red cells from the serum in a timely fashion after drawing blood. In that case, the glucose level will be artifactually reduced. Other artifacts that can influence blood variables are lipemia (the presence of excess fats and fatty acids in the blood), hemolysis (the breakage of blood cells releasing their contents into the sample), poor collection technique, and storage for too long a time at an inappropriate temperature.

Therefore, a single value outside of a “normal range,” or even a number of values outside a “normal range” in one animal does not necessarily mean (or even imply) that an animal’s health is at risk. Rather, these values should serve as signals for the further investigation by experienced veterinarians and animal scientists who exercise professional judgment regarding that value in the context of weighing available evidence.

In the Analyte Evaluation, similarities and differences in analytes across animals and within age-matched cohorts were considered. One or two values outside the range could be considered normal and acceptable variation in the absence of other physiologically based evidence. Because this review attempts to use clinical values to identify potential hazards, even those few values outside the range were examined in the context of other physiological characteristics to determine if those values implied some anomalous trend. If more than two variables were out of

the comparator range, we returned to the animal's entire record in an attempt to understand those values in the context of the animal, and to attempt to determine if they represented a concern for the health of the animal and therefore the safety of edible products derived from it.

In the Animal Evaluation, similarities and differences within individual animals were evaluated. For each animal, we first determined the total number of analytes that were considered sufficiently out of range to imply clinical relevance, thereby triggering further scrutiny. Those analytes were evaluated for internal consistency with other values to rule out artifacts, and then considered within the context of the animal's health records for clinical consistency between the lab work and the clinical picture (based on available veterinary exam records) of the animal.

In the following sections, we have divided our analyses into the three age cohorts: within 24 hours of birth, 1-6 months, and 6-18 months. Results that are outside the comparator range are presented on an analyte and animal-specific basis. The exception to this format is the first subsection that discusses growth-related phenomena that span age groups.

v. Growth-Related Phenomena

Because young animals are growing rapidly, measures of bone growth such as calcium, phosphate, and alkaline phosphatase might be expected to be elevated relative to adults. (Alkaline phosphatase is an enzyme that reflects a number of physiological parameters, and in young animals represents the activity of bone growth and development). This is, in fact, observed in the clones and comparators. Review of Charts E300 and E302 (6 to 18 months old), E200 and E202 (1 to 6 months old), and E100 and E102 (within 24 hours of birth) clearly indicates that all of the alkaline phosphatase levels, and a high proportion of calcium and phosphate levels, are elevated in both cohorts relative to the Cornell Reference range. Review of Charts E301, E201, and E101, however, reveals that clone alkaline phosphatase values are almost entirely within the range of the comparators (0/18 for 6 to 18 months, 8/46 for 1 to 6 months, and 0/10 (within 24 hours of birth) values out of range). For those animals in the one to 6 month cohort, the increased levels of alkaline phosphatase occurred in the youngest animals, a finding consistent with higher rates of growth in younger animals relative to older animals.

Total protein, globulin, and albumin reflect, among other things, the immune status of the organism, which varies with age. Immediately after birth, globulin levels, which are largely comprised of immunoglobulins, are derived almost entirely from colostrum (the antibody rich first "milk" secreted by mammals). "Passive immunity" is conferred by the ingestion and intestinal absorption of immunoglobulin-rich maternal colostrum. In the two to four months after birth, a calf's own immune system begins to ramp up its production of immunoglobulins, as the circulating supply of maternally-derived immunoglobulins in milk wanes. This phenomenon can

be observed in Charts E200 and E202 (Clones: Reference Range (1 to 6 months), and Comparator Population: Reference Range). Clone and comparator calf globulin values are low relative to the Cornell lab reference range because that reference range is derived from adult animals. The clone and comparator calves have not fully started to produce antibodies from their own B-lymphocytes. Review of Chart E201 (Comparison of Clones to Comparator Population), however, indicates that there were few differences between the clone and comparator populations, reflecting the appropriate age-related lag between the decrease in passive acquired immunity and endogenous immunoglobulin production.

The globulin levels that are different between clones and comparators also reflect this age-related physiological phenomenon. Clones #72 and 73 were among the youngest in the 1 to 6 month old group, and thus would be expected to have lower globulin levels. For example, comparison of the globulin value for clone #100 (174 days of age, globulin of 4.6g/dL) with clone #72 (48 days of age and globulin level of 1.6 g/dL) clearly demonstrates the age-related changes in the analyte, and appropriately reflects the normal developmental increase in endogenous globulin production.

In summary, clones and comparators exhibited entirely appropriate developmental stage responses in those laboratory values that reflect age-specific alterations. At least with respect to this dataset, clones respond appropriately to the internal signals guiding normal development and maturation.

f. Animals with Measurements at Different Developmental Nodes

There is a small sub-cohort of seven clones for which laboratory values are available for both the earliest (neonatal) developmental group and early in the second developmental node group (1-6 months). The seven calves are 71, 72, 73, 78, 79, 119, and 132.

More variability was observed among the clinical chemistry values generated from six of seven of these clones relative to the comparators than in the hematology values (for one clone, #119, no differences were noted relative to the comparators). Compare Charts E101 vs. E201 for chemistry and Charts E111 vs. E211 for hematology. In particular, gamma glutamyl transferase (GGT) values appeared lower for four of the seven neonatal clones. This observation is entirely consistent with the difference in treatment that the animals received with respect to the timing of the blood draws. For clones, blood samples were drawn prior to the administration of colostrum, while the comparators had blood samples drawn following its administration. As colostrum has been shown to have high intrinsic GGT activity, the difference between the two groups could well be due to the reflection of absorbed colostrum by the comparators (Meyer and Harvey 2004). GGT values normalized by the second time of measurement for three of the clones. Calf

#73 continued to demonstrate slightly lower GGT activity (4U/L vs. comparator range of 5 - 32 U/L) at Day 48, but this value is unlikely to have clinical significance. We would have expected the globulin level to be lower also, because of the young age of this clone. There was one comparator calf with a low enough globulin level (1.3 g/dl) to allow all of the clone calf values to fall within the range, had it been included in establishing the range.

Six of the seven clones exhibited lower aspartate amino transferase (AST) values at birth relative to the comparators, but these values normalized by the time of the second blood draw (Chart E101 for neonates v. E201 for 1-6 months). Five of the seven clones had low bile acid or cholesterol levels at birth. All three of these analytes are produced by the liver, and their relatively low values may be a reflection of the changeover from fetal hepatic circulation (which bypasses much of the liver) to neonatal circulation in which the liver becomes more fully perfused. Bile acids normalized by the second measurement. The initial, relatively low creatine kinase (CK) (Clone #72), total iron binding capacity (TIBC) (Clone #73), and elevated iron (Clone #132) values resolved by the time of the second blood draw. The cholesterol level for clone #79 was low at birth, but was elevated relative to comparators in the 1-6 month blood sample.

A few measurements appeared elevated in five of the clones at the time of the second measurement. Glucose, alkaline phosphatase, phosphorus, creatinine, and the A/G ratios were elevated relative to the comparators, and anion gap, globulin and total protein were decreased relative to the comparators. These variations from the comparator range are discussed more fully in the Animal and Analyte Review portions of this Appendix.

For hematological data, complete blood count information is only available for four of the seven neonatal clones (Chart E111). Of those, only Calf #78 exhibited any values outside the range of the comparators. At birth, that animal exhibited decreased lymphocytes and decreased platelets, which normalized by the time of the second blood draw. The second blood sample (Chart E211) showed increased banded (immature) neutrophils, whose only biological significance here is that it demonstrates that the clone's bone marrow can produce normal immature neutrophils. The stimulus for the release of the banded neutrophils in this case is not known. No neonatal hematological values were available for clone #78. At the time of the second blood draw (Day 84) clone #79 exhibited decreased total protein as measured by refractometer (TP-ref), increased mean platelet volume (MPV) and decreased red cell distribution width (RDW) relative to the comparators. These variations (increase in bands for clone #78 and decreased total protein (TP) and RDW and increased MPV) are discussed in the animal and analyte review sections. Clone #79 was eventually culled (see discussion in Animal and Analyte Section).

In summary, with the exception of one clone (Clone #73, GGT), none of the values measured at birth that were out of range of the comparator group persisted into the second developmental cohort. Initial laboratory measurements taken at birth differed in clones versus comparators. Some of that difference was likely attributable to the difference in timing or source of colostrum administration between the two groups. Clones #71, 72, and 73 were derived from the same cell line. As discussed in the Animal and Analyte Review sections, most of the values that were out of range at the time of the second blood draw can either be attributed to the age of the animals or do not appear to have clinical significance.

vi. Age Range within 24 hours of birth (Charts E101 and E111)

There were 10 live clones and 17 live comparators in this age cohort.

(c) Analyte Evaluation

Despite an expectation of substantial differences in laboratory measurements between clones and comparators at birth, Chart E101 shows 27 of the 33 analytes were very similar between the groups: they had either one difference or no differences. The remaining six analytes tended to be more variable between clones and comparators.

The values out of range in the four analytes related to liver function (AST, GGT, cholesterol, and bile acids (hBA)) were low relative to comparators. AST levels, in particular, were low in 9 of 10 animals. Although these parameters can indicate zinc or vitamin B₆ deficiencies, (Duncan and Prasse 2003, Meyer and Harvey 2004) have reported that “*reduced AST activities (below the reference range) are noted with relative frequency in dogs and rats during pre-clinical drug trials....*” We are uncertain whether similar observations have been made in cattle. Colostrum possesses an intrinsic high GGT activity (Meyer and Harvey 2004) that is passively transferred to neonates. Given that the clones did not receive colostrum until after blood samples were drawn, while comparators did, lower GGT levels in clones are not unexpected.

Low cholesterol is associated with porto-systemic shunts (abnormal liver blood circulation) in young animals. Fetal circulation provides for the bypass of the bulk of the liver tissue through a vessel called the *ductus venosus* in a loop including the placenta, umbilical vein, and the *ductus venosus*. At birth, the *ductus venosus* closes off; the liver is then fully perfused with blood from the hepatic artery and other components of the hepatic portal circulation. Were these low cholesterol levels to continue into the next developmental age group, there might be cause for concern. Given that the cholesterol levels appear to normalize, based on information from the seven calves that were sampled at both the neonatal and weaning time periods, and review of the overall 1-6 month cohort, there is little reason to think that the lower values in these very young

animals pose a health risk. The low levels at birth are more likely a reflection of the changeover from fetal to adult circulation, possibly exacerbated by the clones' unusually large umbilical vessels, which often required surgical correction (see 1-6 month cohort discussion). The lower bile acid values observed may also be related to the transition from fetal to neonatal circulation, and are not likely indicative of any disease state.

Table E-4: Summary of Laboratory Values from Subcohort of Clones with Laboratory Measurements Taken at Two Developmental Nodes					
Calf ID#	Parameters	Charts E101/111: Birth	Age at Second Blood Draw (Days)	Chart E201/211: 1-6 Months	Current Status
78, Holstein ♀	Clinical Chemistry	↓AST, ↓GGT (pa)	54	↑glucose, ↑alk phos	Presumed healthy
	Hemogram	↓lymphocytes, ↓platelets		↑banding	
79, Holstein ♀	Clinical Chemistry	↓AST, ↓hBA	65	↑creatinine, ↓globulin, ↑A/G, ↑alk phos, ↑chol	Culled
	Hemogram	No data		↓rdw, ↑MPV, ↓TP-ref	
71, Holstein ♀	Clinical Chemistry	↓AST, ↓hBA, ↓GGT (pa)	48	↑P, ↑glucose, ↑alk phos	Presumed healthy
	Hemogram	within range		within range	
72, Holstein ♀	Clinical Chemistry	↓AST, ↓hBA, ↓GGT, ↓CK	48	↓anion gap, ↑P, ↓TP, ↓glob, ↑A/G, ↑glu, ↑alk phos, ↑lipemia	Presumed healthy
	Hemogram	No data		↑MPV	
73, Holstein ♀	Clinical Chemistry	↓AST, ↓GGT (pa), ↓TIBC, ↓hBA, ↓cholesterol	48	↓anion gap, ↑P, ↓TP, ↓glob, ↑A/G, ↑glu, ↓GGT,	Presumed healthy
	Hemogram	within range		↑MPV	
119, red Holstein ♂	Clinical Chemistry	within range	36	within range	Presumed healthy
	Hemogram	within range		within range	
132, Holstein ♂	Clinical Chemistry	↓AST, ↓chol, ↑iron, ↑% saturation	Not available	within range	Presumed healthy
	Hemogram	No data		within range	

pa = presumed artifact
 Other abbreviations as described in Appendix F: Comprehensive Veterinary Examination
 Within range = within the range of values for the comparator population

The hemograms for the neonatal clones (Chart E111) also were very similar to the comparators: 15 of the 17 analytes had either no values out of range or just one value out of the comparator range. With the exception of clone #43, which was infected with rotavirus and died at one day of age, all red blood cell analytes were within the range of the comparator group. Three clones had lymphocyte counts lower than the comparator range.

(d) Animal Evaluation

In general, clones appeared to be more fragile perinatally than comparators. Review of Table E-2 indicates that many of the animals did not survive parturition. Of those that did survive, three were infected with rotavirus, and two, clones #43 and #80, died from rotavirus-induced diarrhea.¹²⁴ Clone #43 exhibited lower AST, cholesterol, GGT, and bile acids than the comparator range, and platelet counts higher than the comparator range. Clone #80 had sodium levels that slightly exceeded the comparator range (149 mg/dL, relative to the highest comparator value of 146 mg/dL), elevated iron, and relatively high percent saturation. The elevated sodium level may have been related to electrolyte disturbances that occur with diarrhea.

Calf #78, a Holstein heifer, also had WBC within comparator range at birth and developed normally through Day 54 of age. She had low lymphocytes and low platelets, but did not present clinically with infection, and survived. Low lymphocyte counts (lymphopenia) can result from severe systemic bacterial and viral infections, disruption of the lymphatic drainage (ruptured thoracic duct), or hereditary disorders in which the production of the lymphocytes is impaired. It may also be a function of suppressed immune function in calves. In the absence of additional information, we think that these data should not be overinterpreted. Of the three clones with lymphopenia, two died from infection, but the third survived, indicating that although perinatal lymphocyte count may be a useful parameter to monitor, it is not predictive of outcome.

Clones #71, 72, 73, 78, 79, 119, and 132 were discussed in aggregate in the preceding section of the subcohort with laboratory measurements taken at two time points. Clone #79 was a Holstein heifer that was culled for poor posture and gait at 54 days of age. In addition to low AST and low bile acids, she was noted to have “very thick withers, and a general build resembling a beef calf. Her left carpus was enlarged, and her left leg deviated laterally.”

Clone #75 was a Holstein heifer with no follow-up data at a later age. She exhibited AST, cholesterol, GGT, and bile acid levels that were lower than the comparator range. Clone #132 was a Holstein heifer initially presented with AST and cholesterol levels lower than the

¹²⁴ Rotavirus is a common enteric pathogen in cattle to which many calves are exposed; some succumb. There is a commercial vaccine for the dam to increase immunity to rotavirus in the colostrum to passively immunize calves in order to protect them.

comparator range and iron and TIBC above the comparator range, but that appeared to be thriving at 50 days.

(e) Conclusions for Perinatal Period: Animal Safety

In general, clones appeared to be more fragile than comparators. Three of the ten calves died or were culled. Two of these died at one day of age of rotavirus infection, and one was culled for poor posture and gait. The major classes of adverse outcomes noted for neonates included stillbirth, umbilical bleeding/abscess/management, colostrum/passive transfer problems, and euthanasia for defects (renal, circulatory, tendon contracture, placental abnormalities, cardiac, abomasal, and ascites (increased fluid in the abdomen)). These problems also occur in conventional animals, although the incidence of the adverse outcomes appears to be higher in clones. Animals with these outcomes are readily identifiable.

The laboratory values indicated a variety of anomalies in the clones. The liver values (AST, GGT, cholesterol, bile acids (hBA)) were decreased in several animals, for reasons likely related to the placental/umbilical abnormalities. GGT levels were also low relative to the comparators, likely related to blood sampling prior to colostrum intake. Although both red and white blood cell counts are more variable than in older cohorts, both systems appear to be functioning normally in response to environmental stress (rotavirus).

Based on these data, it is not possible to use either a particular analyte or analyte profile to predict whether an individual animal, or indeed the entire cohort, will develop into normal, fully functioning healthy animals. The health and laboratory data are consistent with the hypothesis that animals that look and behave normally are most likely normal with respect to laboratory values.

(f) Food Safety

Healthy clones of this age are unlikely to be used for human food, given their potential value as breeding stock. It is also highly unlikely that clones of this age group would be fed to animals except through rendering of dead clones that occurred at parturition or by accident. In any event, the laboratory values do not appear to indicate that these animals are materially different from conventional newborns, but their physical condition at birth seems to indicate that they are more fragile than comparators, and by inference, other conventional animals.

vii. Age Range 1 to 6 months (Charts E201 and E211)

There were 46 clones and 47 comparators in this age cohort.

(g) Analyte Review

Chart E201 shows glucose values were higher for clones than comparators in six of the 42 accepted measurements (four of the values are so low as to be incompatible with life (< 2mg/dL) and thus were considered artifacts). The higher values (ranging from 88-123 mg/dL) may reflect stress responses caused by handling, dietary considerations (including proximity to a meal), or real differences in glucose metabolism. In order to determine whether the hyperglycemia was transient or sustained, urinalysis results were checked for these animals. The renal threshold (the blood level at which glucose spills over into the urine) in cattle is approximately 100 mg/dl. As none of the urinalyses tested positive for glucose, it is unlikely that the hyperglycemia observed in the blood had been sustained long enough to allow spillover from the blood into the urine. Further, no mention of increased water intake or urination was noted on any of the veterinary records of these clones, which are clinical signs of sustained hyperglycemia. Therefore, the most physiologically plausible interpretation of these elevated levels is transient hyperglycemia as a short-lived response to stress.

Total Protein (TP) is an analyte that contains globulin and albumin. Two animals had TP values that were outside the comparator range (Clone #72 and clone #73 were low). Three animals (Clones #94, 102, 128) had decreased levels of SDH relative to the comparators; three others (Clones # 56, 73, 116) exhibited lower GGT levels than the comparators. Although elevated blood levels of SDH and GGT may be indicative of liver disease, the relevance of decreased levels is unclear.

Evaluation of hemograms (Chart E211) indicated that there were no anemic animals. The one value out of range of the comparator group was an elevation, and not a decrease, in RBCs. Although other red cell indices such as mean cell hemoglobin (MCHC) concentration (4/44 clones had elevated levels relative to comparators) and red cell distribution width (RDW) (3/44 clones had lower levels than comparators) were out of comparator range, they are secondary indicators of red cell status. Hemoglobin (Hb) and hematocrit are the primary indicators of red cell status, and were effectively no different from comparators. No variables were consistently out of range for white blood cell evaluations. Four of the 44 clones had elevated basophil counts; the significance of these measurements is unclear. Twelve of the 41 clones had mean platelet volume (MPV) values that were elevated. See clone #102 below for a discussion of MPV.

(h) Animal Evaluation (1 to 6 months Age Group)

Clone #41, a 141 day old Holstein heifer, exhibited no clinical chemistry and one minor hematology value out of range of the comparators. Her health records, however, stated that she had umbilical surgery and harsh lung sounds. No further information is available on this animal.

Clone #58, a 161 day old Angus heifer had a normal veterinary exam, with indication of umbilical surgery. Four clinical chemistry values were out of the comparator range: creatinine, albumin, bile acids, and the A/G ratio were all low. Potential causes of low albumin levels include decreased production from chronic liver failure, or increased loss due to nephropathy (a kidney disease in which proteins are excreted), intestinal disease (enteropathy), or loss into a body cavity such as the thorax or abdomen. Chronic liver failure is accompanied by elevated bile acid levels; clone #58's bile acids were low. There were no other analyte or health measures indicating enteropathy or nephropathy. The low albumin level was also accompanied by a high globulin level, which could well be attributed to the umbilical abscess that was surgically removed, as the globulin response was appropriate to antigenic stimulation. The relevance of low creatinine is unknown. This calf also had four hemogram variables that differed from comparators. Hemoglobin (Hb), TP, and RDW were high, and the lymphocyte count was relatively low. The elevated Hb is consistent with the high hemolysis index of the sample, and is therefore probably not a reflection of the biology of the animal. The elevated TP was likely caused by the elevated globulin in response to antigenic stimulation discussed in the above.

Three Holstein calves (Clones #71, 72, and 73) were derived from the same cell line and were the same age at blood draw (48 days). All three presented with normal veterinary exams, elevated alkaline phosphatase levels, glucose, and phosphate levels. Clone #71 did not exhibit any other clinical values outside the comparator range. The elevated glucose measurements, as discussed above, appear to be transient and likely stress related. Clone #72, also presented with a normal veterinary exam. Her clinical chemistry measurements indicated low anion gap, TP, and globulin levels, and elevated phosphate, A/G ratio, glucose, and lipemia levels. Low anion gap is rare, and can be related to low albumin, which this animal did not exhibit. Its importance is unknown, and in this case, may simply be an anomaly. The low TP and globulin levels are likely age-related, as at this age calves are transitioning from maternal antibody to endogenous production, and there is often a lag in globulin concentration during this age. Plausible explanations for elevated phosphate, A/G, and glucose levels have been discussed previously. The lipemia level for clone #72 was 30U compared to 25U. As the lipemia index is relatively arbitrary, in the absence of corroborative health evidence, it is likely that this value has no real clinical significance. Clone #73 had low anion gap, TP, globulin, and GGT levels in addition to the elevated phosphorus, A/G, and glucose values. As previously discussed,

particularly considering the very young age and high genetic merit of these animals, the elevated phosphorus and alkaline phosphatase levels are not surprising. The influence of colostrum on GGT levels has been discussed previously, although its significance in this age group is not clear. Comparison of clone #41 to clone #73 shows that the former exhibited no laboratory values out of the range of the comparators, but did have health problems, while #73 exhibited many laboratory values outside the comparator range, but no health problems.

Clone #79 was a 65-day-old Holstein heifer at the time of the blood sampling. She was culled for poor posture and gait. Clinical chemistry indicated elevated creatinine, A/G, alkaline phosphatase, cholesterol, and reduced globulin levels.

Clones #87, 88, and 89 were all derived from the same Jersey cell line, and were, respectively, 141, 140, and 131 day old heifers at the time of blood draw. Another animal derived from the same cell line died at birth from LOS-related complications. All three had umbilical surgery and were dehorned. The differences in body weight in these animals illustrate the variability seen among clones derived from the same cell line. Clone #87, the oldest at 141 days, weighed 282 lbs; clone #88 (140 days) weighed 197 lbs; and the youngest (Clone #89 at 131 days) weighed 215 lbs.

Clone #100, a 174 day old Holstein bull, had an elevated WBC (26,500 cells), along with a history of umbilical abscess that was treated surgically. The elevated WBC is an appropriate response to an umbilical abscess.

Clone #102, a 135 day old Holstein heifer also had umbilical surgery, reduced platelets (241×10^3), and an elevated MPV and MCHC relative to the comparators. By itself, the latter measurement has little relevance unless anemia is present. Based on the RBC counts, this calf did not have anemia. The relatively low platelet count also does not appear to be significant; for reference, the low end of the Cornell Reference Range is 232×10^3 , or functionally the same number. Also, automated platelet counters may erroneously count platelets, as they tend to aggregate (clump together). The platelet smear listed platelets as adequate, corroborating that the platelet numbers were likely physiologically appropriate.

(i) Conclusion for 1 to 6 month old group: Animal Safety

The clones in this age cohort were mostly normal. Only one calf was culled for reasons of performance (poor conformation) and not animal health. Such calves are not selected for future breeding and their appearance (and subsequent culling) in a herd is not unique to clones. Culling occurs routinely in conventional breeding programs. The observation of poor conformation in a clone is interesting in that the animal providing the donor cell would likely have exhibited

acceptable conformation, raising the question of whether conformation of this animal is a function of its uterine environment or changes in gene expression. Clones from the same cell line showed considerable variation in their phenotype (see clones #87, 88, and 89 above with respect to weight).

Some of the clones had overt health problems. These included the increased incidence of umbilical problems (*e.g.*, enlargements, excessive bleeding, oomphalitis (navel infection)) tendon contracture, and cryptorchidism. Clones had umbilical extirpation (surgical removal of tissue) at a much higher rate than comparators. This increase represents a real risk to clones related to surgery. Surgical risks include complications that may arise from anesthesia and recovery from surgery, sepsis from manipulating an infected umbilicus, dehiscence (suture line not holding or infection of the suture line), and aspiration of stomach contents into the lungs. Contracted tendons also seemed to occur at a higher frequency than in conventional calves. (Tendon contracture can generally be treated successfully.) Three cases of cryptorchidism were identified. This condition is thought to be heritable, and is relatively uncommon in calves. The risk to the animal is that retained abdominal testicles can develop neoplasia (testicular cancer) in later age. The life cycle of food animals is such that bulls rarely live long enough for neoplasia to develop. In fact, the only food animals that would likely survive to develop such a condition would be breeding bulls. Given that a cryptorchid bull would fail its breeding soundness exam and would not be used for breeding stock (*i.e.*, would be castrated and sent to slaughter when the steer reached the appropriate weight), this eventuality is not likely to occur.

(j) Conclusion for 1 to 6 month old group: Food Safety

It is not likely that clones of this age group would be consumed for food, although there may be some circumstances in which culled clones might be sent into the food supply. When the results of the laboratory analyses are considered in the context of the Cyagra clones' clinical presentation, there were no consistent analyte or physical observations indicating a food safety concern. For example, although some calcium, phosphorus, alkaline phosphatase, and glucose levels fell above the comparator range, all of the elevations can be explained by the clones' stage of life or stress level, and the increased levels observed do not represent a food consumption risk. Further, the laboratory work is consistent with clinical presentation: Calf #100 presented with both umbilical abscess and a high WBC count. In the unlikely event that this animal was sent to slaughter with a large abscess, it would be detected on inspection. The carcass would be condemned if there was evidence of systemic involvement. The abscess would otherwise be cut out and the carcass processed normally. Healthy clones of this age group do not appear to be materially different from the comparators, and would not likely pose a food consumption risk different from conventional animals.

viii. Age Range: 6-18 Months (Charts E301 and E311)

There were 18 clones and 21 comparators in this age cohort.

(k) Analyte Analysis

Review of Chart 301 (Clone: Comparator Population Clinical Chemistry) indicates that there were very few differences between these two cohorts: 33 of the 33 analytes showed no or one value out of the range defined by the comparator population. Two analytes, on first impression appeared to exceed that range: estradiol-17 β (E2) and insulin-like growth factor-1 (IGF-I). On further scrutiny, these values were judged to be of no clinical relevance. Because hormones are important from a physiological and food safety standpoint, their lack of clinical significance is discussed below.

IGF-I is a hormone produced by all mammals, whose presence is necessary for growth and development. Circulating levels of IGF-I have been linked to weight gain and growth rate, and higher levels have been used as a physiological marker for superior genetics in cattle, swine, sheep, and chickens (Davis and Simmen 2000). In this study, IGF-I levels tended to be higher in male clones than in females, and in three bull calves (Clone #24, 33, and 35) were slightly increased (less than 10 percent) relative to the comparator group (respective IGF-I levels of 924, 916, and 938 ng/mL relative to the comparator range of 33-875 ng/mL).

Basal circulating levels of IGF-I vary with a range of factors, and fluctuate dramatically among individual bovines in herds (Vega et al. 1991). In an analysis of 603 conventional Angus cattle conducted 42 days after weaning, the serum concentrations of IGF-I ranged from 17 to 883 ng/mL (Davis and Simmen 2000). Basal IGF-I levels also vary between males and females, with 12 month old bulls exhibiting higher concentrations of IGF-I than steers, heifers, or ovariectomized (animals whose ovaries have been surgically removed) heifers (Plouzek and Trenkle 1991 a,b). Plasma concentrations of IGF-I are also influenced by diet composition and intake, with basal IGF-I levels significantly lower in cattle during feed restriction compared to cattle that are fed to meet maximum growth or production potential. The primary nutritional determinants of basal IGF-I levels appear to be crude protein and total metabolizable energy¹²⁵ (Elsasser et al. 1989). Given that most non-transgenic clones are derived from animals of superior genetic merit for traits such as growth and development, the likelihood that their diets would be highly controlled, and the wide variability in normal IGF-I levels, the observed 10 percent elevations in IGF-I levels are of no clinical significance.

¹²⁵ Metabolizable energy is an estimate of the number of calories absorbed by the animal after digestion.

Of the five clones (# 24, 33, 35, 36, and 69) in the 6 to 18 month dataset that were identified as having plasma E2 levels above the comparators, all were bulls. These differences in E2 levels prompted closer scrutiny. The range in concentrations of these five bulls was 14.16 to 24.33 pg/mL. The range for all 18 clones in this age group was 4.28 to 24.33 pg/mL. The laboratory reference range is 10 to 40 pg/mL, while the comparator range was 4.1 to 11.41 pg/mL. As the laboratory reference range most likely included cycling females, we sought more specific information on E2 concentrations in bull plasma. We then compared the values to the Cornell Reference Range, derived from adult animals, and found that none were outside that range.

Although male mammals produce E2, little research effort has been devoted to studying the role, normal concentration, and fluctuation of endogenous estrogens in the bull. Estrogens are produced in the Sertoli cells of the testis, as well as in adipose tissue and the brain (Henney et al. 1990). Estradiol-17 β (E2) is produced when testosterone binds to cells in the hypothalamus and is converted to E2 by the aromatase enzyme. Receptors for E2 have been identified in the urogenital tracts of growing and adult male mammals of several species, and may be necessary for normal structural and functional development of the male reproductive system (Nilsson et al. 2001). The ratio of E2 to testosterone may be an important factor in male sexual behavior and libido (Henney et al. 1990).

Henney et al. (1990) attempted to relate various hormone concentrations in plasma to libido in 18 Holstein bulls aged 4 to 5 years. Mean concentration of E2 in plasma of these bulls was 10.2 pg/mL, but ranged from 2.8 to 21.7 pg/mL. A more recent study by Sauerwein et al. (2000) measured fluctuations in plasma E2 in Simmental breeding bulls with an average age of 8.4 years with and without recombinant bovine somatotropin (rbST) treatment. Untreated controls (injected with vehicle only) and treated bulls exhibited fluctuations in E2 concentrations over the 25 week study. Concentrations of E2 in untreated bulls ranged from approximately 5 pg/mL to approximately 23 pg/mL, based on Figure 2 in this paper, with a mean pre-injection concentration of 12.0 ± 1.5 pg/mL. No papers were identified which discussed E2 concentration in young, growing bulls; however, given its possible role in normal development, increased levels of E2 in growing bulls of high genetic merit may be expected to have slightly elevated levels. Based on these ranges and those established by the reference laboratory, E2 concentrations of 24 pg/mL are not a concern, and should be considered within normal fluctuations for bulls.

Analysis of the hematological parameters for all of the clones and comparators (Chart E311) showed no remarkable findings. Sixteen of the 17 analytes measured for clones were within the range of the comparators, or had only one difference. No problems were identified with red blood cell measurements (*e.g.*, anemia or polycythemia (increase in the total mass of red blood

cells in the body)), or white blood cell problems (*e.g.* leukocytosis (increase in WBC count) or leukopenia (low WBC count)) were seen. Two animals (Clones #99 and #108) presented with a MCV below that of the comparators, but given the lack of corresponding evidence of anemia in these animals, these values have no clinical significance. Clone #108 was culled due to dwarfism.

(l) Animal Analysis

Unless specifically mentioned, no differences were observed between an individual clone's clinical chemistry or hematology values and the range presented by the comparators.

Clone #103 (Red Angus, 6.5 month old heifer) exhibited elevated potassium (K^+), asparagine transferase (AST), and creatine kinase (CK) levels relative to the comparators. None of the values in the hemogram exceed the range defined by the comparators. A physical exam conducted on the same day as the blood draws showed no abnormalities. The sample drawn from this calf had the highest hemolysis index (353) of all of the samples, indicating poor sample handling. Potassium is an electrolyte found mostly within the cell, and its elevation could be caused by sample hemolysis. Asparagine transferase (AST) is an enzyme normally found in liver or muscle tissue that can be released when liver or muscle is damaged. Creatine kinase is a muscle specific enzyme that is released when there is muscle damage. Although these values are elevated, they are low enough to imply only minor muscle damage, similar to that observed when an animal is recumbent for an extended period of time, struggling in a squeeze chute, or may be artifactual due to blood sample handling leading to hemolysis. Given that this animal exhibited no clinical abnormalities, even if these analyte levels are not due to artifact or poor sample handling, these changes would likely have no clinical significance.

Clone #108 was an Angus heifer that is characterized as a dwarf with frequent bloat. She was reported to have been severely deformed with abnormal gastro-intestinal (GI) motility. Interestingly, clinical chemistry results show only a decrease in TIBC compared to the comparator group. The hemogram indicated high RBC and slightly lower MCV relative to comparators. The hematocrit was within the range of the comparators. Despite the animal's obvious physical abnormalities, laboratory values were not significantly out of range.

(m) Conclusions for 6-18 Month Group: Animal Safety

Clones in this age group exhibited no differences from comparators with respect to their overall health, and were indistinguishable from the comparator group on the basis of clinical and laboratory tests. No residual health problems were noted in this group of animals, with the exception of the dwarf calf that was identified visually and culled. Based on these observations, there are no apparent health risks in this age group of animals.

(n) Conclusions for 6-18 Month Group: Food Safety

No material differences were observed between the six to eighteen month old clone cohort and their approximately age-matched, similarly reared comparators. In fact, the clones are indistinguishable from the comparators, and thus would be unlikely to pose any risk for consumption as human food or in animal feed above conventional animals now in the food supply.

5. Charts and Tables

The following Charts summarize the results of laboratory data derived from the Cyagra clones and corresponding non-clone comparators. In Charts 100, 102, 110, 112, 200, 202, 212, 300, 302, 310, and 312, the clones' laboratory data were compared to the Cornell veterinary laboratory data and to the approximately age- and breed- matched comparators' data. A laboratory value from a clone that exceeded the range of the Cornell Reference Range or comparators was initially flagged regardless of how much it was out of range. After all of the comparisons had been made (*i.e.*, clones to comparators, clones to reference range, and comparators to reference range), it became apparent that the most appropriate comparison for clinical relevance was clones to comparators. We then determined the clinical relevance of the out of range values. Clinical relevance was defined as laboratory value observed in the clones that was more than 10 percent out of the comparator range, or, one that based on veterinary clinical judgment, was likely to cause concern. The published literature was consulted for non-standard clinical chemistry endpoints such as IGF-I and estrogen. Laboratory values determined to be not clinically relevant are represented in the Charts as gray boxes. Clinical relevance is presented in charts 101, 111, 201, 211, 301, and 311.

Cyagra Data Chart Interpretations

Table 100 Summary of Clinical Chemistry Data for Cyagra Clones and Cornell Reference Range: Neonates

Animal	43	71	72	73	75	78	79	80	119	132	Summary ¹
Sodium	↑	■	■	■	■	■	■	↑	■	■	2/10
Potassium	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	10/10
Chloride	■	■	■	■	■	■	■	■	↓	■	1/10
Bicarbonate	■	■	■	■	↓	↓	■	↓	■	■	3/10
Anion Gap	■	■	■	■	↑	↑	↑	↑	↑	■	5/10
Urea	■	■	■	■	■	■	■	■	■	■	0/10
Creatinine	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	10/10
Calcium	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	10/10
Phosphate	↑	■	■	↑	↑	↑	↑	↑	■	■	6/10
Mg - XB	■	■	■	■	■	■	■	■	↑	■	1/10
Total Protein	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	10/10
Albumin-bulk	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	10/10
Globulin	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	10/10
A/G	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	10/10
Glucose	↑	↓	↓	↓	↓	↓	↓	↑	↓	■	9/10
AST/P5P	↓	↓	↓	↓	↓	↓	↓	↓	■	↓	9/10
SDH	↓	↓	↓	↓	↓	↓	↓	↓	■	↓	9/10
Alk Phos	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	10/10
GGT	↓	↓	↓	↓	↓	↓	↓	↓	↑	↓	10/10
Σ Bilirubin	↑	↑	↑	↑	↑	↑	↑	■	↑	↑	9/10
Bilirubin-d	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	10/10
Bilirubin-i	■	↑	↑	↑	■	↑	■	■	↑	↑	6/10
Amylase											
Cholesterol	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	10/10
CK	■	■	↓	■	■	■	■	■	↑	■	2/10
Iron	↓	↓	↓	↓	↓	↓	↓	■	↓	■	8/10
TIBC	■	↓	↓	↓	↓	↓	↓	↓	■	■	7/10
% Sat	↓	↓	↓	↓	↓	■	↓	■	↓	■	7/10
hBA	*	↓	↓	↓	↓	↓	↓	■	■	*	6/9
Lipemia											
Icterus											
IGF-1	■	■	*	■	■	■	■	■	■	*	0/8
Estradiol	↑	↑	↑	↑	■	■	■	■	*	■	4/9
Summary ²	20/30	21/30	22/29	22/30	22/30	23/30	21/30	19/30	20/29	15/28	

- In Comparison Range
- Outside the comparison range but not clinically relevant
- ↑/↓ Animal is above/below the clinically relevant range
- * No numerical value was provided
- X = Presumed artifact

^b = Animal culled

¹ = # of animals with outcomes outside comparator range/total animals

² = # of outcomes outside range/total outcomes per animal

For description of analytes and their relevance to assess animal health consult Appendix J

Table 101 Cyagra Clones and Comparator Population: neonates

Animal	43	71	72	73	75	78	79	80	119	132	Summary ¹
Sodium	■	■	■	■	■	■	■	↑	■	■	1/10
Potassium	■	■	■	■	■	■	■	■	■	■	0/10
Chloride	■	■	■	■	■	■	■	■	■	■	0/10
Bicarbonate	■	■	■	■	■	■	■	■	■	■	0/10
Anion Gap	■	■	■	■	■	■	■	■	■	■	0/10
Urea	■	■	■	■	■	■	■	■	■	■	0/10
Creatinine	■	■	■	■	■	■	■	■	■	■	0/10
Calcium	■	■	■	■	■	■	■	■	■	■	0/10
Phosphate	■	■	■	■	■	■	■	■	■	■	0/10
Magnesium- XB	■	■	■	■	■	■	■	■	■	■	0/10
Total Protein	■	■	■	■	■	■	■	■	■	■	0/10
Albumin- bulk	■	■	■	■	■	■	■	■	■	■	0/10
Globulin	■	■	■	■	■	■	■	■	■	■	0/10
A/G	■	■	■	■	■	■	■	■	■	■	0/10
Glucose	■	■	■	■	■	■	■	■	■	■	0/10
AST/P5P	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	9/10
SDH	■	■	■	■	■	■	■	■	■	■	0/10
Alkaline Phos	■	■	■	■	■	■	■	■	■	■	0/10
GGT	↓	↓	↓	↓	↓	↓	■	■	■	■	1/5
Total Bilirubin	■	■	■	■	■	■	■	■	■	■	0/10
Direct Bilirubin	■	■	■	■	■	■	■	■	■	■	0/10
Indirect Bilirubin	■	■	■	■	■	■	■	■	■	■	0/10
Amylase	■	■	■	■	■	■	*	*	■	■	0/8
Cholesterol	↓	■	■	↓	↓	■	■	■	■	↓	4/10
Creatine Kinase	■	■	↓	■	■	■	■	■	■	■	1/10
Iron	■	■	■	■	■	■	■	↑	■	↓	2/10
TIBC	■	■	■	↓	■	■	■	■	■	■	1/10
% Saturation	■	■	■	■	■	■	■	↑	■	■	1/10
hBA- Random	↓	↓	↓	↓	↓	■	↓	■	■	*	6/9
Lipemia	■	■	■	■	■	■	■	■	■	■	0/10
Icterus	■	■	■	■	■	■	■	■	■	■	0/10
IGF-1	■	■	*	■	■	■	■	■	■	*	0/8
Estradiol	■	■	■	■	■	■	■	■	*	■	0/9
Summary ²	4/33	3/32	4/32	5/33	4/33	2/33	2/32	4/32	0/32	3/31	

- In Comparison Range
- Outside the comparison range but not clinically relevant
- ↑/↓ Animal is above/below the clinically relevant range
- * No numerical value was provided
- X = Presumed artifact

^b = Animal culled

¹ = # of animals with outcomes outside comparator range/total animals

² = # of outcomes outside range/total outcomes per animal

For description of analytes and their relevance to assess animal health consult Appendix J

Table 102 Summary of Clinical Chemistry Data for Cyagra Comparison Population and Cornell Reference Range: neonates

Animal	140	141	135	136	137	138	139	186	187	188	189	190	191	192	193	194	195	Summary ¹
Sodium	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1/17
Potassium	■	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	16/17
Chloride	■	↓	↓	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	3/17
Bicarbonate	↓	■	↓	↑	↓	■	■	■	■	↓	■	■	■	■	■	■	■	5/17
Anion Gap	↑	↑	↑	■	↑	■	■	■	■	↑	■	■	↑	↑	■	■	↑	8/17
Urea	↓	↓	↑	■	■	■	■	↓	■	■	↓	↑	■	↓	■	■	■	7/17
Creatinine	↑	↑	↑	■	↑	↑	↑	↑	↑	↑	■	↑	■	■	↑	↑	↑	13/17
Calcium	↑	↑	↑	■	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	16/17
Phosphate	■	■	↑	■	↑	■	■	■	↑	↑	■	↑	↑	↑	↑	■	■	9/17
Mg - XB	■	■	↑	■	↑	■	■	↑	↑	■	↓	↑	■	↓	■	■	■	7/17
Total Protein	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	17/17
Albumin-bulk	↓	↓	↓	↓	■	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	16/17
Globulin	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	■	■	↓	↓	↓	↓	↓	15/17
A/G	↑	↑	■	■	↑	■	↑	■	■	↑	↓	■	■	↑	■	■	↑	8/17
Glucose	■	■	↓	↑	↑	■	■	■	■	↑	■	■	↑	↑	■	■	↓	7/17
AST/P5P	↓	■	↑	■	↑	■	■	■	■	■	■	■	■	■	↑	↑	↓	6/17
SDH	↓	↓	↑	↓	↑	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	15/17
Alk Phos	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	17/17
GGT	■	■	↑	↑	■	↑	↓	↑	↑	■	↑	↑	↑	↑	↑	↑	↓	13/17
∑ Bilirubin	↑	↑	↑	↑	■	↑	↑	↑	↑	■	↑	↑	↑	↑	↑	↑	↑	15/17
Bilirubin-d	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	17/17
Bilirubin-i	↑	↑	↑	↑	■	↑	↑	↑	↑	■	↑	↑	↑	↑	↑	↑	↑	15/17
Amylase																		
Cholesterol	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	17/17
CK	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	■	↓	↑	■	↑	↑	↑	15/17
Iron	■	■	↓	↓	■	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	14/17
TIBC	■	■	■	■	■	■	■	■	■	■	■	■	↑	↓	↓	↓	■	4/17
% Sat	■	■	↓	↓	■	↓	■	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	13/17
hBA	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	1/17
Lipemia																		
Icterus																		
IGF-1	■	■	↓	*	■	■	■	■	■	↓	↓	↓	■	↓	■	↓	↓	7/16
Estradiol	■	↑	↑	■	↑	■	■	■	■	■	↓	↓	■	↓	■	↑	↑	8/17
Summary ²	17/30	18/30	26/30	17/29	18/30	18/30	16/30	19/30	18/30	19/30	18/30	20/30	20/30	22/30	19/30	20/30	21/30	

■ In Comparison Range
 □ Outside the comparison range but not clinically relevant
 No numerical value was provided
 ↑ ↓ Animal is above/below the clinically relevant range
 X = Presumed artifact

^b = Animal culled
¹ = # of animals with outcomes outside comparator range/total animals
² = # of outcomes outside range/total outcomes per animal
 For description of analytes and their relevance to assess animal health consult Appendix J

Table 111 Summary of Hemogram Data for Cyagra Clones and Comparator Population: neonates

Animal	43	71	72	73	75	78	79	80	119	132	Summary ¹
Hematocrit	↓	■	*	■	■	■	*	■	■	*	1/7
Hemoglobin	■	■	*	■	■	■	*	■	■	*	0/7
RBC	↓	■	*	■	■	■	*	■	■	*	1/7
MCV	■	■	*	■	■	■	*	■	■	*	0/7
MCH	↓	■	*	■	■	■	*	■	■	*	1/7
MCHC	↓	■	*	■	■	■	*	■	■	*	1/7
RDW	↑	■	*	■	■	■	*	■	■	*	1/7
WBC	■	■	*	■	■	■	*	■	■	*	0/7
Seg. Neutr	■	■	*	■	■	■	*	■	■	*	0/7
Banded Neutr	↑	■	*	■	■	■	*	■	■	*	1/7
Lymphocytes	↓	■	*	■	■	↓	*	↓	■	*	3/7
Monocytes	■	■	*	■	■	■	*	■	■	*	0/7
Eosinophils	■	■	*	■	■	■	*	■	■	*	0/7
Basophils	■	■	*	■	■	■	*	■	■	*	0/7
Platelets	↑	■	*	■	■	↓	*	■	■	*	2/7
MPV	↑	■	*	■	■	■	*	■	■	*	1/7
TP- Ref	■	■	*	■	■	■	*	■	■	*	0/7
Summary ²	9/17	0/17		0/17	0/17	2/17		1/17	0/17		

■ In Comparison Range

■ Outside the comparison range but not clinically relevant

↑/↓ Animal is above/below the clinically relevant range

* No numerical value was provided

X = Presumed artifact

^b = Animal culled

¹ = # of animals with outcomes outside comparator range/total animals

² = # of outcomes outside range/total outcomes per animal

For description of analytes and their relevance to assess animal health co

Table 112 Summary of Hemogram Data for Cyagra Comparison Population and Cornell Reference Range: neonates

Animal	140	141	135	136	137	138	139	186	187	188	189	190	191	192	193	194	195	Summary ¹
Hematocrit	⬆	⬆	⬇	■	*	⬆	⬆	⬆	⬆	⬆	⬇	*	*	⬆	*	⬆	■	11/13
Hemoglobin	■	⬆	⬇	⬇	*	■	■	■	■	■	⬇	*	*	■	*	■	⬇	5/13
RBC	■	⬆	⬇	■	*	⬆	⬆	■	■	■	⬇	*	*	■	*	■	■	5/13
MCV	■	■	■	■	*	■	■	■	■	■	■	*	*	■	*	■	■	0/13
MCH	■	⬇	■	⬇	*	■	■	■	⬇	⬇	⬇	*	*	■	*	⬇	⬇	7/13
MCHC	⬇	⬇	⬆	⬇	*	⬇	⬇	⬇	⬇	⬇	⬇	*	*	⬇	*	⬇	⬇	13/13
RDW	■	■	⬆	⬆	*	■	■	■	⬆	■	⬆	*	*	⬆	*	■	■	5/13
WBC	■	■	⬇	■	*	■	■	⬆	■	■	⬇	*	*	⬇	*	⬆	■	5/13
Seg. Neutr	⬆	⬆	■	■	*	⬆	■	⬆	⬆	■	⬇	*	*	■	*	⬆	■	7/13
Banded Neutr	■	■	■	■	*	■	■	■	■	■	■	*	*	■	*	■	■	0/13
Lymphocytes	■	⬇	■	⬇	*	⬇	⬇	■	■	■	■	*	*	⬇	*	⬇	■	6/13
Monocytes	■	■	■	■	*	■	■	■	■	■	■	*	*	■	*	■	■	0/13
Eosinophils	■	■	■	■	*	■	■	■	■	■	■	*	*	■	*	■	■	0/13
Basophils	■	■	■	■	*	■	■	■	■	■	■	*	*	■	*	■	■	0/13
Platlets	■	⬆	■	■	*	*	⬆	⬆	■	*	■	*	*	■	*	■	■	3/13
MPV	⬆	■	⬆	⬆	*	*	■	⬆	⬆	*	■	*	*	■	*	⬆	■	6/13
TP- Ref	⬇	⬇	■	■	*	⬇	⬇	⬇	■	⬇	■	*	*	⬇	*	⬇	⬇	9/13
Summary ²	5/17	9/17	7/17	6/17		6/17	6/17	7/17	6/17	4/15	8/17			6/17		8/17	4/17	

■ In Comparison Range
 ■ Outside the comparison range but not clinically relevant
 ⬆ ⬇ Animal is above/below the clinically relevant range
 * No numerical value was provided
 X = Presumed artifact

^b = Animal culled
¹ = # of animals with outcomes outside comparator range/total animals
² = # of outcomes outside range/total outcomes per animal
 For description of analytes and their relevance to assess animal health cons

Table 210 Summary of Hemogram Data for Cyagra Clones and Cornell Reference Range: 1-6 month cohort

Animal	39	40	41	42	56	58	71	72	73	74	76	78	79	81	82	83	85	87	88	89	90	93	94	96	100	101	102	106	110	111	114	115	116	117	118	119	120	121	124	126	127	128	129	130	131	132	Summary ¹													
Hematocrit	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	*	☞	☞	☞	☞	☞	☞	*	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	43/44									
Hemoglobin	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	☞	*	■	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2/44				
RBC	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	*	☞	☞	☞	☞	☞	☞	*	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	41/44					
MCV	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	*	☞	☞	☞	☞	☞	☞	*	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	43/44				
MCH	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	*	☞	☞	☞	☞	☞	☞	*	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	44/44			
MCHC	■	☞	■	■	■	■	☞	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	6/44				
RDW	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	*	☞	☞	☞	☞	☞	☞	*	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	41/44			
WBC	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	☞	*	■	■	■	■	■	■	*	☞	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	5/44			
Seg. Neut.	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	*	■	■	☞	■	■	☞	*	■	■	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	15/44		
Banded Neut.	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/44				
Lymphocytes	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	*	■	■	☞	■	■	☞	*	☞	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	6/44			
Monocytes	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	☞	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2/44		
Eosinophils	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/44			
Basophils	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/44			
Platelets	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	*	☞	☞	☞	☞	☞	*	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	18/41
MPV	■	☞	☞	*	■	■	☞	☞	☞	■	■	☞	☞	☞	☞	☞	*	☞	☞	*	☞	☞	*	■	■	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	☞	14/41
TP- Ref	■	■	■	■	■	☞	☞	☞	☞	■	■	■	■	☞	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	5/44	

■ In Comparison Range
 ☞ Animal is above/below the clinically relevant range
 ☞ Outside the comparison range but not clinically relevant
 No numerical value was provided
 X = Presumed artifact
^b = Animal culled
¹ = # of animals with outcomes outside comparator range/total animals
² = # of outcomes outside range/total outcomes per animal
 For description of analytes and their relevance to assess animal health consult Appendix J

Table 211 Summary of Hemogram Data for Cyagra Clones and Comparator Population: 1-6 month cohort

Animal	39	40	41	42	56	58	71	72	73	74	76	78	79	81	82	83	85	87	88	89	90	93	94	96	100	101	102	106	110	111	114	115	116	117	118	119	120	121	124	126	127	128	129	130	131	132	Summary ¹						
Hematocrit	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/44		
Hemoglobin	■	■	■	■	■	⬆	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/44	
RBC	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/44
MCV	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/44
MCH	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/44
MCHC	■	■	⬆	■	■	■	■	■	■	■	■	■	■	⬆	■	■	*	■	■	■	■	■	■	*	■	⬆	⬆	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	4/44	
RDW	■	■	■	■	■	■	■	■	■	■	■	■	■	⬇	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	⬇	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	⬇	■	■	3/44	
WBC	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	⬆	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/44	
Seg. Neut.	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/44	
Banded Neut.	■	■	■	■	■	■	■	■	■	■	■	■	⬆	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/44	
Lymphocytes	■	■	■	■	■	⬇	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/44	
Monocytes	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/44	
Eosinophils	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	⬆	*	■	■	⬆	■	■	*	■	⬆	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2/44	
Basophils	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	⬆	■	■	■	⬆	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	4/44	
Platelets	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	*	■	■	*	■	■	⬇	■	■	■	⬇	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3/41		
MPV	■	⬆	■	*	■	■	■	■	■	■	■	■	⬆	⬆	■	■	*	■	⬆	■	*	■	■	*	■	■	⬆	■	■	⬆	⬆	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	⬆	■	9/41		
TP- Ref	■	■	■	■	■	⬆	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2/44		
Summary ²	0/16	1/16	1/16	0/14	0/16	3/16	0/16	0/16	0/16	0/16	0/16	1/16	3/16	2/16	0/16	1/16		0/16	1/16	1/16	0/14	0/16	0/16		2/16	1/16	3/16	0/16	1/16	2/16	2/16	0/16	2/16	0/16	0/16	0/14	0/16	0/16	0/16	0/16	0/16	0/16	0/16	0/16	0/16	2/16	0/16						

■ In Comparison Range
 ■ In Comparison Range
 ⬆ Animal is above/below the clinically relevant range
 ⬆ Outside the comparison range but not clinically relevant
 * No numerical value was provided
 X = Presumed artifact
^b = Animal culled
¹ = # of animals with outcomes outside comparator range/total animals
² = # of outcomes outside range/total outcomes per animal
 For description of analytes and their relevance to assess animal health consult Appendix J

Table 212 Summary of Hemogram Data for Cyagra Comparison Pop and Cornell Reference Range: 1-6 month cohort

Animal	142	143	144	145	146	147	148	149	150	151	152	153	155	156	157	158	159	162	164	166	167	168	170	171	172	173	174	175	176	177	178	179	180	181	182	183	184	185	196	197	198	199	200	201	202	203	204	213	214	Summary ¹								
Hematocrit	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	42/45							
Hemoglobin	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	3/45					
RBC	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	42/45				
MCV	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	28/45				
MCH	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	45/45			
MCHC	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	36/45			
RDW	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	44/45			
WBC	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	7/45				
Seg. Neut.	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	5/45			
Banded Neut.	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	0/45			
Lymphocytes	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	12/45			
Monocytes	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	4/45			
Eosinophils	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	0/45		
Basophils	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	0/45		
Platelets	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	17/45		
MPV	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	9/45		
TP- Ref	█	█	*	█	█	█	█	█	█	█	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	*	*	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	█	1/45
Summary ²	7/17	6/17	10/17		6/17	4/17	7/17	5/15	6/17	7/17	8/17		7/17	7/17	6/17	8/17	7/17	5/17	5/17	5/17	7/17	7/17	4/17	5/17	6/17	7/17	6/17	8/17	5/17	4/17			6/17	8/17	7/17	5/17	7/17	5/17	5/17	5/17	8/17	6/17	9/17	8/17	8/17	8/17	7/17	7/17	6/17									

█ In Comparison Range
 █ Animal is above/below the clinically relevant range
 █ Outside the comparison range but not clinically relevant
 █ No numerical value was provided
 X = Presumed artifact
^b = Animal culled
¹ = # of animals with outcomes outside comparator range/total animals
² = # of outcomes outside range/total outcomes per animal

For description of analytes and their relevance to assess animal health consult Appendix J

Table 300 Summary of Clinical Chemistry Data for Cyagra Clones and Cornell Reference Range: 6-18 month cohort

Animal	24	25	26	27	33	35	36	38	37	55	67	69	70	98	99	103 ^b	104	108 ^b	Summary ¹	
Sodium	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	1/18
Potassium	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	↑	↑	4/18
Chloride	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Bicarbonate	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Anion Gap	↑	■	■	↑	↑	■	■	■	■	■	↑	↑	↑	■	■	↑	■	■	■	7/18
Urea	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	1/18
Creatinine	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	↓	■	↑	■	3/18
Calcium	■	■	■	↑	■	■	■	■	■	↑	↑	■	↑	■	■	↑	↑	↑	■	7/18
Phosphate	↑	↑	↑	↑	↑	↑	■	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	17/18
Mg - XB	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Total Protein	■	↓	↓	■	■	■	■	↓	↓	↓	↓	■	■	↓	■	↓	↓	↓	■	10/18
Albumin-bulk	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	1/18
Globulin	■	↓	■	■	■	■	■	■	↓	↓	↓	■	■	↓	■	↓	↓	↓	■	8/18
A/G	■	■	■	■	■	■	■	■	↑	↑	■	■	■	■	■	■	■	↑	■	3/18
Glucose	↑	■	↑	↑	■	■	↑	↑	↑	■	■	■	↑	↑	↑	↓	■	↓	■	11/18
AST/P5P	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
SDH	■	■	■	■	■	■	■	■	↓	↓	■	■	■	■	■	↓	↓	■	■	4/18
Alk Phos	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	18/18
GGT	■	■	■	■	■	■	■	■	↓	↓	■	■	■	■	■	↓	↓	■	■	4/18
Σ Bilirubin	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Bilirubin-d	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Bilirubin-i	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Amylase																				
Cholesterol	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
CK	■	↑	■	■	■	■	■	■	↑	■	↑	■	↑	■	■	↑	↑	■	■	6/18
Iron	■	■	■	■	■	■	■	↓	■	↑	■	■	■	■	■	■	↑	■	■	3/18
TIBC	■	■	↓	■	■	■	■	↓	■	■	■	■	■	■	↓	■	■	↓	■	4/18
% Sat	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	↑	■	3/18
hBA	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	↓	■	■	2/18
Lipemia																				
Icterus																				
IGF-1	↑	↑	↑	↑	↑	↑	↑	↑	*	■	↑	■	↑	■	■	■	■	↓	■	11/17
Estradiol	■	■	■	■	■	■	■	↓	*	↓	■	■	■	↓	↓	↓	↓	↓	■	7/17
Summary ²	5/30	6/30	6/30	6/30	5/30	3/30	3/30	8/30	10/28	12/30	8/30	4/30	8/30	6/30	5/30	14/30	13/30	13/30		

■ In Comparison Range
 □ Outside the comparison range but not clinically relevant
 ↑ ↓ Animal is above/below the clinically relevant range
 * No numerical value was provided
 X = Presumed artifact

^b = Animal culled
¹ = # of animals with outcomes outside comparator range/total animals
² = # of outcomes outside range/total outcomes per animal

For description of analytes and their relevance to assess animal health consult Appendix J

Table 301 Summary of Clinical Chemistry Data for Cygra Clones and Comparator Population: 6-18 month cohort

Animal	24	25	26	27	33	35	36	38	37	55	67	69	70	98	99	103	104	108	Summary ¹	
Sodium	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Potassium	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	1/18
Chloride	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Bicarbonate	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Anion Gap	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Urea	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Creatinine	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Calcium	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Phosphate	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Mg - XB	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Total Protein	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Albumin-bulk	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Globulin	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
A/G	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Glucose	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
AST/P5P	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/18
SDH	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/18
Alk Phos	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
GGT	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Σ Bilirubin	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Bilirubin-d	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Bilirubin-i	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Amylase	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Cholesterol	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
CK	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/18
Iron	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/18
TIBC	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/18
% Sat	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
hBA	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Lipemia	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Icterus	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
IGF-1	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Estradiol	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Summary ²	0/33	0/33	0/33	0/33	0/33	0/33	0/33	1/33	1/31	0/33	0/33	0/33	0/33	0/33	0/33	3/33	0/33	1/33		

■ In Comparison Range
 ■ Outside the comparison range but not clinically relevant
 * No numerical value was provided
 X = Presumed artifact

^b = Animal culled
¹ = # of animals with outcomes outside comparator range/total animals
² = # of outcomes outside range/total outcomes per animal

For description of analytes and their relevance to assess animal health consult Appendix J

Table 302 Summary of Clinical Chemistry Data for Cygra Comparison Pop and Cornell Reference Range: 6-18 month cohort

Animal	135	154	206	160	161	162	163	165	169	205	207	208	209	210	211	212	215	216	217	218	219	Summary ¹	
Sodium	■	■	■	↑	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2/21
Potassium	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	2/21
Chloride	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	1/21
Bicarbonate	↓	■	■	■	■	■	↓	↓	■	■	■	■	■	■	■	↓	↓	↓	↓	↓	↓	↓	8/21
Anion Gap	■	■	■	↑	↑	■	↑	↑	↑	■	■	■	■	■	■	■	↑	↑	↑	↑	↑	↑	10/21
Urea	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/21
Creatinine	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/21
Calcium	■	↑	↑	■	↑	■	↑	↑	↑	↑	■	■	■	■	■	↑	↑	■	■	■	■	■	9/21
Phosphate	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	■	↑	↑	■	↑	↑	↑	↑	↑	↑	↑	↑	19/21
Mg - XB	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/21
Total Protein	↓	↓	↓	■	↓	↓	■	↓	■	■	↓	↓	↓	↓	↓	■	↓	↓	↓	↓	↓	↓	16/21
Albumin-bulk	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/21
Globulin	↓	↓	■	↓	↓	↓	■	↓	■	■	↓	↓	■	■	■	■	↓	↓	↓	↓	↓	↓	12/21
A/G	■	↑	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	↑	↑	↑	↑	5/21
Glucose	■	↑	↑	■	■	↓	■	↓	■	↑	↑	↑	↑	↑	↑	↑	■	↑	■	■	↑	↑	13/21
AST/P5P	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/21
SDH	↓	■	■	■	■	■	■	■	■	■	↓	↓	■	■	■	■	↓	■	■	■	■	■	4/21
Alk Phos	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	21/21
GGT	↓	■	■	■	■	↓	■	■	↓	■	■	↓	■	■	■	■	↓	↓	↓	↓	↓	↓	9/21
Σ Bilirubin	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/21
Bilirubin-d	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/21
Bilirubin-i	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/21
Amylase																							0
Cholesterol	■	■	■	■	■	↓	■	■	■	■	■	↓	↓	■	↓	■	■	■	■	■	■	■	4/21
CK	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	↑	↑	↑	■	↑	5/21
Iron	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/21
TIBC	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/21
% Sat	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/21
hBA	↓	■	■	■	■	■	■	■	■	■	■	↓	■	■	↓	■	■	■	■	■	■	■	3/21
Lipemia																							0
Icterus																							0
IGF-1	*	■	■	■	■	■	■	■	■	↑	↑	↑	↑	↑	↑	↑	■	■	■	■	■	■	7/20
Estradiol	*	↓	↓	■	↓	↓	↓	↓	↓	↓	■	■	■	■	↓	■	↓	↓	↓	↓	↓	↓	14/20
Summary ²	8/29	10/30	7/30	6/30	7/30	11/30	5/30	12/30	6/30	6/30	6/30	9/30	6/30	5/30	9/30	6/30	11/30	10/30	10/30	9/30	10/30		

■ In Comparison Range
 ■ Animal is above/below the clinically relevant range
 □ Outside the comparison range but not clinically relevant
 □ No numerical value was provided
 X = Presumed artifact

^b = Animal culled
¹ = # of animals with outcomes outside comparator range/total animals
² = # of outcomes outside range/total outcomes per animal

For description of analytes and their relevance to assess animal health consult Appendix J

Table 310 Summary of Hemogram Data for Cyagra Clones and Cornell Reference Range: 6-18 month cohort

Animal	24	25	26	27	33	35	36	38	37	55	67	69	70	98	99	103 ^b	104	108 ^b	Summary ¹	
Hematocrit	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	18/18
Hemoglobin	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	1/18
RBC	■	■	■	■	■	■	■	■	↑	↑	↑	↑	↑	↑	↑	■	■	↑	■	8/18
MCV	■	■	■	■	■	■	■	■	■	↓	■	↓	↓	↓	↓	■	■	↓	■	6/18
MCH	■	■	■	■	■	■	■	■	■	↓	↓	↓	↓	↓	↓	■	■	↓	■	7/18
MCHC	↓	↓	↓	↓	↓	↓	↓	↓	■	■	↓	↓	↓	■	■	■	■	■	■	11/18
RDW	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	■	17/18
WBC	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1/18
Seg. Neut.	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Banded Neut.	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Lymphocytes	■	■	■	■	■	↑	■	■	■	↑	■	■	■	■	■	■	■	■	■	2/18
Monocytes	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Eosinophils	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Basophils	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Platelets	■	■	*	*	■	*	■	*	■	*	*	↑	↑	■	↑	■	↑	■	■	4/12
MPV	■	■	*	*	■	*	■	*	↑	*	*	■	■	■	↓	↑	■	↑	■	4/12
TP- Ref	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Summary ²	3/17	4/15	3/15	3/15	3/17	4/15	3/17	3/15	4/17	6/15	5/15	7/17	7/17	5/17	7/17	3/17	3/17	6/17		

■ In Comparison Range
 □ Outside the comparison range but not clinically relevant
 ↑ ↓ Animal is above/below the clinically relevant range
 * No numerical value was provided
 X = Presumed artifact

^b = Animal culled
¹ = # of animals with outcomes outside comparator range/total animals
² = # of outcomes outside range/total outcomes per animal

For description of analytes and their relevance to assess animal health consult Appendix J

Table 311 Summary of Hemogram Data for Cyagra Clones and Comparator Population: 6-18 month cohort

Animal	24	25	26	27	33	35	36	38	37	55	67	69	70	98	99	103	104	108	Summary ¹	
Hematocrit	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Hemoglobin	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
RBC	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	1/18
MCV	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	2/18
MCH	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
MCHC	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
RDW	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
WBC	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Seg. Neut.	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Banded Neut.	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Lymphocytes	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Monocytes	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Eosinophils	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Basophils	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Platelets	■	■	*	*	■	*	■	*	■	*	*	■	■	■	■	■	■	■	■	0/12
MPV	■	■	*	*	■	*	■	*	■	*	*	■	■	■	■	■	■	■	■	0/12
TP- Ref	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0/18
Summary ²	0/17	0/17	0/15	0/15	0/15	0/15	0/17	0/15	0/17	0/15	0/15	0/17	0/17	0/17	1/17	0/17	0/17	2/17		

■ In Comparison Range
 ■ In Comparison Range
 ↑ ↓ Animal is above/below the clinically relevant range
 * Outside the comparison range but not clinically relevant
 * No numerical value was provided
 X = Presumed artifact
^b = Animal culled
¹ = # of animals with outcomes outside comparator range/total animals
² = # of outcomes outside range/total outcomes per animal
 For description of analytes and their relevance to assess animal health consult Appendix J

Table 312 Summary of Hemogram Data for Cyagra Comparison Population and Cornell Reference Range: 6-18 month cohort

Animal	135	154	206	160	161	162	163	165	169	205	207	208	209	210	211	212	215	216	217	218	219	Summary ¹	Animal
Hematocrit	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	↑	*	↑	↑	↑	↑	↑	↑	↑	↑	↑	20/20	Hematocrit
Hemoglobin	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	↑	■	■	↑	■	2/20	Hemoglobin
RBC	■	↑	↑	■	↑	↑	↑	↑	↑	■	■	*	↑	↑	■	↑	↑	↑	■	↑	↑	14/20	RBC
MCV	■	↓	↓	■	■	■	■	↓	↓	■	■	*	■	■	■	■	■	■	■	■	↓	5/20	MCV
MCH	■	↓	↓	■	↓	↓	↓	↓	↓	■	■	*	↓	↓	■	↓	■	■	■	■	■	10/20	MCH
MCHC	↓	■	■	■	↓	↓	↓	↓	↓	■	■	*	↓	↓	■	↓	↓	■	↓	↓	■	12/20	MCHC
RDW	■	↑	↑	↑	↑	↑	↑	↑	↑	■	■	*	↑	↑	↑	↑	↑	↑	↑	↑	↑	17/20	RDW
WBC	■	■	■	■	■	↑	↑	↑	■	■	■	*	■	■	■	■	■	■	■	■	■	3/20	WBC
Seg. Neut.	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	0/20	Seg. Neut.
Banded Neut.	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	0/20	Banded Neut.
Lymphocytes	↓	■	↑	■	↑	↑	↑	↑	■	↑	■	*	■	■	■	■	■	■	■	■	■	7/20	Lymphocytes
Monocytes	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	0/20	Monocytes
Eosinophils	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	0/20	Eosinophils
Basophils	■	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	0/20	Basophils
Platelets	↑	■	↑	■	■	■	■	■	■	■	*	*	*	↓	■	■	■	↑	■	■	■	4/18	Platelets
MPV	↑	■	↓	■	■	■	■	■	■	↑	*	*	*	■	■	■	■	■	■	■	■	3/18	MPV
TP- Ref	↑	■	■	■	■	■	■	■	■	■	■	*	■	■	■	■	■	■	■	■	■	1/20	TP- Ref
Summary ²	6/17	5/17	8/17	2/17	6/17	7/17	7/17	8/17	6/17	3/17	1/17	/17	5/17	6/17	2/17	5/17	5/17	4/17	3/17	5/17	4/17		

■ In Comparison Range
 ■ Animal is above/below the clinically relevant range
 * Outside the comparison range but not clinically relevant
 No numerical value was provided

^b = Animal culled
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X = Presumed artifact

For description of analytes and their relevance to assess animal health consult Appendix J

Cyagra Raw Data

Cyagra Inc.
Table E-5: Cloned Birth Outcome Summary

Calf #	Sex	Breed	Birth Date	Birth Wt (kg)	Birth	Current Status	Age 03/24/03 (mos)	Age at Death (d)	Cause of Death	Glucose Req'd	Thermoreg. Prob.	Calf #	Oxygen Req'd	Umbilicus (Probs.)	Umbilicus Hernia/ Infection	Umbilical Surgery	Noted Minor Problems	Major Problems	
1	F	NP	4-Aug-01	45.5	NP	alive	19.9	NA	NA	NP	NP	1	NP	NP	NP	NP	NP	NP	
2	F	NP	5-Aug-01	54.5	NP	alive	19.9	NA	NA	NP	NP	2	NP	NP	NP	NP	NP	NP	
3	F	NP	29-Sep-01	NP	NP	dead	NA	0	stillborn	NP	NP	3	NP	NP	NP	NP	NP	abn dev	
4	F	NP	28-Sep-01	NP	NP	alive	18.1	NA	NA	NP	NP	4	NP	NP	NP	NP	not clone of donor	NP	
5	M	NP	7-Nov-01	NP	NP	alive	16.7	NA	NA	NP	NP	5	NP	NP	NP	NP	NP	NP	
6	M	NP	7-Nov-01	NP	NP	dead	NA	16	accident	NP	NP	6	NP	NP	NP	NP	NP	calf hung in stall	
7	M	NP	8-Nov-01	NP	NP	alive	16.7	NA	NA	NP	NP	7	NP	NP	NP	NP	NP	NP	
8	M	NP	7-Nov-01	NP	NP	alive	16.7	NA	NA	NP	NP	8	NP	NP	NP	NP	yes	scouring	
9	M	NP	7-Nov-01	NP	NP	alive	16.7	NA	NA	NP	NP	9	NP	NP	NP	NP	NP	fever	
10	M	NP	7-Nov-01	NP	NP	alive	16.7	NA	NA	NP	NP	10	NP	NP	NP	NP	NP	NP	
11	M	NP	7-Nov-01	NP	NP	dead	NA	0	euthanized	NP	NP	11	NP	NP	NP	NP	NP	abn dev	
12	M	NP	17-Nov-01	NP	NP	dead	NA	0	stillborn	NP	NP	12	NP	NP	NP	NP	NP	recip ruptured uterus	
13	M	NP	19-Nov-01	NP	NP	dead	NA	0	unknown	NP	NP	13	NP	NP	NP	NP	NP	appeared normal	
14	NP	AN	12-Jan-02	33.2	C-section	dead	NA	13	septicemia/umbilicus	no	no	14	yes	no	no	no	infection, oxygenation	contracture	
15	NP	AN	12-Jan-02	37.3	C-section	alive	14.5	NA	NA	no	no	15	no	no	no	no	infection, heart murmur	NP	
16	NP	AN	15-Jan-02	50	C-section	dead	NA	2	failure to trans to neonat circ	no	no	16	yes	yes	no	no	slack abdomen	breathing	
17	NP	AN	16-Feb-02	44.5	Natural	alive	13.4	NA	NA	no	no	17	no	no	no	no	no	NP	
18	NP	AN	28-Feb-02	88.2	Natural	dead	NA	0	stillborn	NP	NP	18	NP	NP	NP	NP	NP	polycystic kidneys	
19	NP	AN	28-Feb-02	69.1	Natural	dead	NA	0	stillborn	NP	NP	19	NP	yes	NP	NP	NP	flaccid abdomen	
20	M	HO	25-Nov-01	NP	C-section	dead	NA	0	euthanized	NP	NP	20	NP	NP	NP	NP	NP	abn dev	
21	M	HO	28-Nov-01	41.8	C-section	alive	16	NA	NA	no	no	21	yes	no	no	no	no	NP	
22	M	HO	28-Nov-01	NP	C-section	alive	16	NA	NA	no	no	22	yes	no	no	no	low PCV	NP	
23	M	HO	30-Nov-01	45.5	C-section	dead	NA	0	euthanized	yes	NP	23	yes	yes	NP	NP	NP	abn dev, int. bleeding	
24	M	HO	30-Nov-01	31.8	C-section	alive	16	NA	NA	yes	no	24	yes	no	no	no	NP	NP	
25	M	HO	12-Dec-01	54.5	C-section	alive	15.6	NA	NA	yes	NP	25	NP	NP	NP	no	NP	NP	
26	M	HO	18-Dec-01	NP	C-section	alive	15.4	NA	NA	no	NP	26	NP	NP	NP	no	NP	NP	
27	M	HO	22-Dec-01	NP	C-section	alive	15.2	NA	NA	no	NP	27	NP	NP	NP	no	NP	NP	
28	M	HO	25-Nov-01	NP	C-section	dead	NA	0	stillborn	NP	NP	28	NP	NP	NP	NP	NP	NP	
29	M	HO	27-Nov-01	NP	C-section	dead	NA	0	euthanized	NP	NP	29	NP	yes	NP	NP	NP	abn dev	
30	M	HO	4-Dec-01	72.3	C-section	alive	15.8	NA	NA	NP	yes	30	yes	no	no	no	hyperthermia, mild contracture	NP	
31	M	HO	5-Dec-01	76.8	C-section	dead	NA	0	euthanized	NP	NP	31	NP	no	NP	NP	NP	abn (renal) dev	
32	M	HO	11-Dec-01	54.5	C-section	alive	15.6	NA	NA	no	no	32	yes	no	no	no	mild contracture	NP	
33	M	HO	17-Dec-01	57.7	C-section	alive	15.4	NA	NA	yes	no	33	yes	no	no	no	NP	NP	
34	M	HO	27-Nov-01	NP	C-section	dead	NA	0	stillborn	NP	NP	34	NP	NP	NP	NP	NP	NP	
35	M	HO	7-Feb-02	41.8	C-section	alive	13.7	NA	NA	yes	no	35	yes	no	no	no	immature lungs	NP	
36	M	HO	8-Feb-02	52.7	C-section	alive	13.6	NA	NA	no	no	36	no	no	no	no	NP	NP	
37	M	HO	NP	NP	NP	alive	NA	NA	NA	NP	NP	37	NP	NP	NP	NP	NP	NP	
38	M	HO	19-Feb-02	67.3	C-section	alive	13.3	NA	NA	NP	NP	38	NP	no	no	no	anemia, slight forelimb contracture	NP	
39	F	HO	19-Feb-03	NP	NP	NP	NP	NP	NP	NP	NP	39	NP	NP	NP	yes	NP	NP	
40	F	HO	24-Feb-03	NP	NP	NP	NP	NP	NP	NP	NP	40	NP	NP	NP	yes	NP	NP	
41	F	HO	3-Dec-02	NP	C-section	alive	3.7	NA	NA	yes	no	41	yes	yes	yes	yes	yes	contracture/splinting	NP
42	F	HO	10-Nov-02	NP	C-section	alive	4.5	NA	NA	yes	no	42	yes	large	yes	NP	NP	NP	
43	F	HO	10-Mar-02	NP	NP	dead	NA	1	rotavirus	NP	NP	43	NP	NP	NP	NP	NP	diarrhea	rotavirus
44	F	HO	29-Jan-02	45.5	C-section	alive	14	NA	NA	no	no	44	yes	yes	no	no	slight int. bleeding	NP	
45	F	HO	29-Jan-02	NP	C-section	alive	14	NA	NA	no	no	45	no	yes	no	no	slight int. bleeding	NP	
46	F	HO	24-Feb-02	51.4	C-section	alive	13.1	NA	NA	no	no	46	yes	no	no	no	NP	NP	
47	F	HO	31-Jan-02	NP	C-section	dead	NA	0	stillborn	NP	NP	47	NP	NP	NP	NP	NP	NP	
48	F	HO	1-Feb-02	54.5	C-section	dead	NA	0	stillborn	NP	NP	48	NP	NP	NP	NP	NP	NP	
49	F	HO	1-Feb-02	NP	C-section	dead	NA	0	stillborn	NP	NP	49	NP	NP	NP	NP	NP	NP	
50	F	HO	8-Mar-02	59.5	C-section	alive	12.7	NA	NA	no	no	50	no	yes	no	yes	NP	flaccid abdomen	
51	F	HO	15-Mar-02	NP	C-section	dead	NA	0	stillborn	NP	NP	51	NP	yes	NP	NP	NP	flaccid abdomen, bulldog	
52	F	HO	17-Mar-02	NP	Fetotomy	dead	NA	0	stillborn	NP	NP	52	NP	NP	NP	NP	NP	NP	
53	F	HO	2-Apr-02	56.8	C-section	alive	11.9	NA	NA	no	no	53	no	yes	NP	NP	yes	NP	NP
54	F	HO	4-Jun-02	59.1	C-section	dead	NA	0	euthanized	NP	NP	54	NP	NP	NP	NP	NP	reverted to fetal circulation	cardian, neurological problems
55	F	HO	6-Jun-02	54.5	C-section	alive	9.7	NA	NA	no	no	55	yes	yes	yes	yes	large abdomen	NP	
56	F	AN	11-Nov-02	NP	C-section	alive	4.4	NA	NA	yes	no	56	yes	no	no	no	NP	NP	
57	F	AN	12-Nov-02	NP	induced/vaginal	dead	NA	23	ruptured abomasum	no	no	57	yes	no	no	no	NP	NP	
58	F	AN	13-Nov-02	NP	C-section	alive	4.4	NA	NA	no	no	58	yes	yes	no	yes	NP	lost blood	
59	F	Longhorn	21-May-02	27.3	C-section	alive	10.2	NA	NA	no	no	59	no	no	no	no	NP	NP	

Cal#	Sex	Breed	Birth Date	Birth Wt (kg)	Birth	Current Status	Age 03/24/03 (mos)	Age at Death (d)	Cause of Death	Glucose Req'd	Thermoreg. Prob.	Cal#	Oxygen Req'd	Umbilicus (Probs.)	Umbilicus Hernia/ Infection	Umbilical Surgery	Noted Minor Problems	Major Problems
121	M	HO	2-Jan-03	NP	C-section	alive	2.7	NA	NA	yes	no	121	yes	no	NP	NP		NP
122	M	HO	6-Jan-03	NP	C-section	alive	2.6	NA	NA	yes	no	122	yes	proud flesh, moist	NP	NP	contracture, req'd phys therapy	transfusion needed
123	M	HO	28-Jan-03	NP	C-section	dead	NA	9	pyelonephritis/umbilicus	yes	no	123	yes	NP	NP	NP	contracted front fetlocks	NP
124	M	HO	28-Jan-03	NP	C-section	alive	1.8	NA	NA	yes	no	124	yes	large and moist	no	NP		NP
125	M	HO	6-Feb-03	NP	C-section	dead	NA	0	euthanized	NP	NP	125	NP	NP	NP	NP		NP
126	M	HO	10-Feb-03	NP	C-section	alive	1.4	NA	NA	yes	no	126	yes	ripped vessels	no	NP	large cotyledons, normal	NP
127	M	HO	10-Feb-03	NP	C-section	alive	1.4	NA	NA	yes	no	127	yes	NP	NP	NP		NP
128	M	HO	12-Feb-03	NP	NP	NP	NP	NP	NP	NP	NP	128	NP	NP	NP	yes	cryptorchid	NP
129	M	HO	13-Feb-03	NP	NP	NP	NP	NP	NP	NP	NP	129	NP	NP	NP	yes		NP
130	M	HO	16-Feb-03	NP	NP	NP	NP	NP	NP	NP	NP	130	NP	NP	NP	NP	right testicle not descended	NP
131	M	HO	21-Feb-03	NP	NP	NP	NP	NP	NP	NP	NP	131	NP	NP	NP	yes	left testicle not descended	NP
132	M	HO	24-Feb-03	NP	NP	NP	NP	NP	NP	NP	NP	132	NP	NP	NP	NP		NP
133	M	HO	26-Apr-03	NP	NP	NP	NP	NP	NP	NP	NP	133	NP	NP	NP	NP		NP
134	M	HO	26-Apr-03	NP	NP	NP	NP	NP	NP	NP	NP	134	NP	NP	NP	NP		NP
KEY																		
NP= Not Provided																		
2WP= Two Weeks Premature																		

Table E-6: Assay Dates (6-18 month calves)

	Calf #	Breed	Sex	DOB	Assay Date	Age (Days)	
Clones	10	Holstein	F	6-Jun-02	15-Apr-03	313	
	98	Holstein	F	2-Sep-02	30-Apr-03	240	
	99	Holstein	F	2-Sep-02	30-Apr-03	240	
	103	Red Angus	F	17-Oct-02	7-May-03	202	
	104	Red Angus	F	18-Oct-02	23-Apr-03	187	
	108	Angus	F	31-Oct-02	7-May-03	188	
	60	Texas Long	F	22-May-02	23-Apr-03	336	
	24	Holstein	M	30-Nov-01	10-Jun-03	557	
	25	Holstein	M	12-Dec-01	10-Jun-03	545	
	26	Holstein	M	18-Dec-01	10-Jun-03	539	
	27	Holstein	M	21-Dec-01	10-Jun-03	536	
	33	Holstein	M	17-Dec-01	10-Jun-03	540	
	35	Holstein	M	7-Feb-02	10-Jun-03	488	
	36	Holstein	M	8-Feb-02	10-Jun-03	487	
	38	Holstein	M	20-Feb-02	10-Jun-03	475	
	67	Holstein	M	26-Jul-02	10-Jun-03	319	
	69	Holstein	M	15-Aug-02	10-Jun-03	299	
	70	Holstein	M	16-Aug-02	10-Jun-03	298	

Table E-100a: Individual Animal Clinical Chemistry at Birth

		Individual animal clinical chemistry data at birth																		
Birth	Blood Chem	Sodium	Potassium	Chloride	Bicarbonate	Anion Gap	Urea	Creatine-rb	Calcium	Phosphate	Magnesium-xb	Total Protein	Albumin-bulk	Globulin	A/G	Glucose	AST/P5P	SDH	Alk. Phos.	
	Ref Low	134	3.9	94	22	17	10	0.4	8.3	4.2	1.7	7.2	3.2	3.5	0.6	31	53	10	23	
	Ref High	145	5.3	105	30	24	25	1	10.4	7.7	2.2	9.4	4.2	5.8	1.2	77	162	50	78	
	Units	mEq/L	mEq/L	mEq/L	mEq/L	mEq/L	mg/dL	mg/dL	mg/dL	mg/dL	mEq/L	g/dL	g/dL	g/dL		mg/dL	U/L	U/L	U/L	
Controls	140	145	5.3	100	21	29	8	2.4	12.4	6.7	2.1	4.2	2.7	1.5	1.8	60	47	4	348	
	141	141	5.8	93	27	27	8	2.6	11.8	5.9	2	4.1	2.7	1.4	1.93	58	54	4	418	
	135	142	7.8	86	16	48	43	5	15.1	12.3	4.6	4.5	2.4	2.1	1.14	12	386	145	882	
	136	134	7.3	93	31	17	11	0.9	10.4	6.7	1.7	5.8	2.5	3.3	0.76	87	162	<2	229	
	137	141	8.5	99	15	36	25	2.5	13.9	13.3	3	4.6	3.2	1.4	2.29	182	182	211	379	
	138	140	6.2	96	28	22	10	1.4	11.5	6.9	2	4.7	2.5	2.2	1.14	75	109	<2	840	
	139	142	7.2	101	24	24	13	2.2	11.5	7.3	1.8	4	2.7	1.3	2.08	66	63	<2	126	
	186	138	9.6	97	28	23	8	1.5	11.6	8.4	2.3	5.4	2.6	2.8	0.93	73	123	4	628	
	187	138	6.5	94	28	23	10	1.7	11.2	7.8	2.3	5.7	2.4	3.3	0.73	69	137	4	1415	
	188	140	8.3	95	19	34	10	2.6	12.9	9.9	2.2	4.3	2.8	1.5	1.87	248	76	6	564	
	189	141	5.8	100	24	23	9	0.6	11.2	5.8	1.5	6.3	2.3	4	0.58	60	66	5	136	
	190	144	6.2	101	25	24	30	1.2	11.8	9.1	2.4	6.3	2.8	3.5	0.8	65	53	0	143	
	191	146	6.4	103	24	25	12	0.8	11.1	8.3	1.8	5.2	2.8	2.4	1.17	84	79	3	343	
	192	142	6.7	98	26	25	5	0.9	11.4	8.1	1.5	4.5	2.6	1.9	1.37	81	72	10	225	
	193	138	7.2	97	29	19	15	1.8	10.6	7.8	2	5.3	2.5	2.8	0.89	44	166	6	707	
	194	140	6.5	99	27	21	10	1.7	11.3	7.4	1.9	5	2.4	2.6	0.92	60	170	2	487	
	195	140	5.6	98	22	26	25	1.5	12.6	7.2	1.8	4.3	2.8	1.5	1.87	17	51	21	103	
Clones	43	146	7.1	104	27	22	16	1.4	13	7.9	1.9	4.5	2.8	1.7	1.65	98	46	4	154	
	71	140	6	99	25	22	17	2.1	12.8	7.2	1.9	4.2	2.5	1.7	1.47	7	18	3	215	
	72	142	5.8	100	26	22	17	1.9	12.8	6.8	1.9	4.2	2.5	1.7	1.47	25	15	<2	214	
	73	142	5.7	101	24	23	20	3.1	12.5	8.2	2	4.1	2.6	1.5	1.73	12	22	3	241	
	75	143	5.5	97	21	31	20	2.2	13.3	8	2	4.4	2.6	1.8	1.44	5	15	4	204	
	78	143	5.4	100	19	29	13	2.4	13.3	7.9	2	4.4	2.6	1.8	1.44	10	15	<2	355	
	79	141	5.5	98	22	27	17	3.3	13.2	7.8	2.1	4.5	2.8	1.7	1.65	17	40	5	323	
	80	149	5.7	101	19	35	13	2.5	13.5	8.5	2	4.5	2.9	1.6	1.81	69	22	6	372	
	119	138	5.9	90	25	29	15	3.5	12.8	6	2.6	5.1	3	2.1	1.43	30	150	13	798	
	132	139	5.4	94	27	23	17	1.7	12.6	7.3	2	4.4	2.9	1.5	1.93	42	17	3	378	

Table E-100a: Individual Animal Clinical Chemistry at Birth

Birth	Blood Chem	GGT	Total Bilirubin	Dir. Bilirubin	Ind. Bilirubin	Amylase	Cholesterol	CK	Iron	TIBC	% Saturation	hBA-random	Lipemia	Hemolysis	Icterus	IGF-1	Estradiol
	Ref Low	11	0	0	0		73	77	113	362	28	9				34.7	10
	Ref High	39	0.1	0	0.1		280	265	226	533	48	455				472.27	40
	Units	U/L	mg/dL	mg/dL	mg/dL	U/L	mg/dL	U/L	ug/dL	ug/dL	%	umol/L				ng/ml	pg/ml
Controls	140	21	0.3	0.1	0.2	53	22	331	136	421	32		16	39	0	203.57	29.3
	141	21	0.6	0.1	0.5	48	26	551	156	375	42	12	15	43	1	137.4	52.73
	135	691	0.8	0.2	0.6	40	20	11033	54	373	14	54.3	9	20	1	0.065	59.44
	136	1217	1.7	0.1	1.6	31	37	731	70	450	16	18.1	22	53	2	nes	27.66
	137	12	0.1	0.1	0	68	15	559	148	473	31	58.5	17	44	0	113.51	103.76
	138	1413	1	0.1	0.9	29	34	501	88	365	24	10.5	21	117	1	141.01	17.33
	139	8	1	0.1	0.9	19	28	356	104	367	28	13.7	21	75	1	87.77	25.5
	186	1012	0.3	0.1	0.2	24	24	379	53	397	13	9.5	25	55	0	145.61	28.53
	187	1829	0.9	0.1	0.8	39	27	768	82	392	21	12.7	5	66	1	57.12	33.99
	188	19	0.1	0.1	0	14	25	1209	57	411	14	16.2	0	114	1	3.07	37.99
	189	893	1.5	0.1	1.4	62	59	111	24	378	6	26.3	15	22	2	6.75	6.14
	190	391	0.8	0.1	0.7	60	37	65	29	411	7	7.6	16	35	1	11.79	9.81
	191	268	1.7	0.1	1.6	62	47	462	60	536	11	10	0	162	3	41.82	20.58
	192	383	0.9	0.1	0.8	33	45	87	44	348	13	21.7	2	15	1	16.69	6.18
	193	3213	1	0.2	0.8	48	44	513	47	354	13	15.2	21	251	1	127.94	18.79
	194	1623	1.2	0.1	1.1	42	29	929	48	337	14	16.6	26	33	2	29.67	42.88
	195	5	0.9	0.1	0.8	49	27	637	79	376	21	28.2	7	20	1	6.89	50.45
Clones	43	<3	0.2	0.1	0.1	33	10	125	48	532	9	0	0	293	0	92.06	46.04
	71	3	0.9	0.1	0.8	20	14	209	66	314	21	5	13	24	1	106.85	61.51
	72	3	0.7	0.1	0.6	20	14	61	65	318	20	2.4	13	29	1		44.54
	73	<3	0.4	0.1	0.3	14	11	106	52	264	20	2	10	122	0	128.66	48.06
	75	<3	0.2	0.1	0.1	15	12	118	33	342	10	1.1	25	7	0	71.67	23.34
	78	<3	0.3	0.1	0.2	28	16	179	112	331	34	8.2	20	21	0	97.68	35.42
	79	5	0.3	0.2	0.1		24	201	38	312	12	3.3	8	19	0	82.2	17.16
	80	6	0.1	0.1	0		18	186	164	343	48	12.3	0	13	0	149.44	27.38
	119	845	0.4	0.2	0.2	59	18	711	93	509	18	15.9	14	5	0	187.12	
	132	5	0.3	0.1	0.2	35	12	91	163	383	43		22	28	0		31.03

Table E-100b: Individual Animal Hematology at Birth

		Individual animal hematology data at adulthood														
Hemogram		Hematocrit (HCT)	Hemoglobin (HB)	RBC	MCV	MCH	MCHC	RDW	WBC	Seg. Neut.	Band. Neut.	Lymphocytes	Monocytes	Eosinophils	Basophils	
Ref Low	23	8.6	5.4	36	14	36	16.2	5.6	1.8	0	1.9	0	0	0		
Ref High	25	13.2	8.2	49	19	40	19.7	13.7	7.7	0	7.4	1.2	1.4	0.3		
Units	%	g /dL	mill/uL	fL	pg	g /dL	%	thou/uL	thou/uL	thou/uL	thou/uL	thou/uL	thou/uL	thou/uL		
Controls	135	33	11.8	7.5	44	16	35	18.4	6	3.7	0	1.7	0.1	0.5	0	
	154	28	11	8.4	33	13	40	20.7	8.5	2.4	0	6.1	0	0	0	
	206	28	10.8	8.9	32	12	39	23.3	13.2	5	0	7.5	0.7	0	0	
	160	30	11.8	7.9	38	15	39	19.8	12.4	6	0	6.2	0.2	0	0	
	161	34	11.4	8.6	39	13	34	22.2	13.4	3.1	0	10.1	0.3	0	0	
	162	35	10.3	8.4	42	12	29	31.4	14.4	2.7	0	10.8	0.7	0	0.1	
	163	34	10.3	9.4	37	11	30	28.3	14.6	4.5	0	9.9	0.1	0	0	
	165	34	11.6	9.6	35	12	35	31.1	24.9	5.2	0	19.2	0.5	0	0	
	169	33	11.5	9.7	34	12	35	21.1	12.2	4.5	0	7	0.2	0.5	0	
	205	30	11.4	6.8	44	17	38	17.4	12.7	3.9	0	8.3	0.1	0.3	0.1	
	207	29	11	7.3	39	15	38	18.4	8.6	2.6	0	5.8	0.1	0.1	0.1	
	208															
	209	32	10.5	8.7	37	12	33	33.1	11.3	4.4	0	6.7	0.2	0	0	
	210	34	10.6	9	38	12	31	32.1	10	4.4	0	5.1	0.4	0.1	0	
	211	28	10.7	7.8	36	14	38	22.7	9.8	3.7	0	5.8	0.1	0.2	0	
	212	34	10.5	8.6	39	12	31	30.2	12.5	5.1	0	6.8	0.6	0	0	
	215	46	14.8	10.1	46	15	32	34.6	9.6	4.1	0	5.4	0	0.1	0	
	216	32	12.7	9	36	14	40	24.1	8.4	2.8	0	5.5	0.1	0	0	
	217	38	12.7	8	47	16	34	23.1	13.6	6.9	0	6.4	0.1	0.1	0	
	218	42	13.8	10.2	42	14	33	36	9.9	2.6	0	6.8	0.4	0	0.1	
219	31	12.2	9	34	14	40	22.3	12.4	6.9	0	4.8	0.5	0	0.1		
Clones	24	33	11	6.9	48	16	33	20.2	11.6	6.4	0	4.8	0.5	0	0	
	25	32	10.7	6.9	46	16	34	23.8	13.9	6.1	0	7.2	0.6	0	0	
	26	32	10.6	6.8	47	16	34	21.4	9.5	2.4	0	6.7	0.2	0.2	0	
	27	32	10.5	6.7	47	16	33	20.5	7.8	2.5	0	4.8	0.3	0.2	0	
	33	33	10.7	7.7	43	14	32	33	10.2	2.1	0	7	0.5	0.4	0.1	
	35	34	11.1	7.7	44	15	33	33.9	10.9	2.7	0	7.7	0.2	0.1	0.1	
	36	30	9.9	6.9	44	14	33	32.2	9.2	3	0	5.9	0.1	0.1	0.1	
	38	31	9.7	6.6	46	15	32	28.3	10.5	4.7	0	5.4	0	0.4	0	
	37	35	12.2	8.7	40	14	35	20.1	8.1	2.3	0	5.2	0.6	0.1	0	
	55	30	11.1	8.6	35	13	37	21.7	10.5	2.1	0	7.6	0.5	0.3	0	
	67	34	11.1	9.5	36	12	32	32.4	9.5	4.8	0	3.8	0.3	0.6	0	
	69	31	10.1	9.5	33	11	33	29.5	8.2	2.5	0	5.4	0.1	0.2	0	
	70	33	10.8	9.9	34	11	32	31.7	8.2	2.5	0	5.2	0.1	0.3	0	
	98	28	10.8	8.9	32	12	38	22.6	8.1	2.1	0	5.4	0.2	0.1	0.3	
	99	28	10.6	9.3	30	12	38	20.4	8.5	2.4	0	5.7	0.4	0	0	
	103	28	10.5	7.2	40	15	37	21.6	8.6	3.5	0	4.9	0.2	0	0	
	104	28	10.9	7.4	38	15	39	19.9	8.8	5.5	0	3	0.2	0.2	0	
	108	37	13.8	11.9	31	12	37	19.4	8.2	3.5	0	3.9	0.3	0.2	0.2	

Notes:

- NSA No Significant Abnormalities
- NS None Seen
- N Normal
- SCM Smudged Cells-Moderate

Table E-100b: Individual Animal Hematology at Birth

Hemogram		Platelet Smear	Platelets	MPV	TP-Ref	RBC Morphology	Acanthocytes	Anisocytosis	Echinocytes	Poikilocytes	Schistocytes	Parasites	WBC Exam	Plasma Appearance
Ref Low			232	5.6	5.9									
Ref High			596	8.2	8.1									hemolysis
Units			thou/uL	fL	g /dL									
Controls	135	Incr.	812	16.8	8.3	NSA						NS	NSA	Moderate
	154	Adeq.	391	5.8	6.7			Mild				NS	NSA	N
	206	Incr. (*1)	601	5.5	6.8	NSA						NS	NSA	N
	160	Adeq.	259	6.6	7.3	NSA						NS	NSA	N
	161	Adeq.	391	7.4	7				Moderate			NS	NSA	N
	162	Adeq.	487	6.8	6.5				Moderate			NS	NSA	N
	163	Adeq.	515	6.3	7.2				Many			NS	NSA	N
	165	Adeq.	279	6.2	6.8	NSA						NS	Few	N
	169	Adeq. (*1)	559	5.9	7.2	NSA						NS	NSA	N
	205	Adeq.	309	10.7	7	NSA						NS	NSA	Slight
	207	Adeq. (*1)			6.6	NSA						NS	NSA	Slight
	208													
	209	Adeq. (*1)			7				Mild			NS	NSA	Slight
	210	Adeq. (*1)	117	6.4	7.3			Few				NS	NSA	Slight
	211	Adeq.	414	7.3	7				Mild			NS	NSA	N
	212	Adeq.	351	6.5	7.4	NSA						NS	NSA	N
	215	Adeq. (*1)	242	5.9	7.5	NSA						NS	NSA	Slight
	216	Incr.	671	6.3	6.7	NSA						NS	NSA	Slight
	217	Adeq.	468	6.3	6.5				Mild			NS	NSA	Slight
218	Adeq.	422	6	6.9	NSA						NS	NSA	Slight	
219	Adeq.	583	6.3	7				Mild	Few		NS	NSA	Slight	
Clones	24	Adeq.	398	7.2	7.3	NSA						NS	NSA	Slight
	25	Adeq.	313	6.7	6.8	NSA						NS	NSA	N
	26	Adeq. (*1)			7.2	NSA						NS	NSA	Slight
	27	Adeq. (*1)			7.2	NSA						NS	NSA	N
	33	Adeq.	392	5.8	7.8	NSA						NS	NSA	N
	35	Adeq. (*1)			7.4	NSA						NS	NSA	Slight
	36	Adeq.	335	6.6	7.4	NSA						NS	NSA	N
	38	Adeq. (*1)			7.5	NSA						NS	NSA	Slight
	37	Adeq.	328	12.4	7.5	NSA						NS	NSA	Slight
	55	Adeq. (*1)			6.8	NSA								Slight
	67	Adeq. (*1)			6.9	NSA						NS	NSA	Slight
	69	Incr.	658	5.9	7.7	NSA						NS	NSA	N
	70	Incr.	605	6	7.3	NSA						NS	NSA	N
	98	Adeq. (*1)	395	6.8	6.7				Mild			NS	NSA	N
	99	Incr. (*1)	622	5.5	7.4				Mild			NS	NSA	N
	103	Adeq.	337	11	6.8					Mod		NS	NSA	N
104	Incr.	745	6.4	7.4	NSA						NS	NSA	N	
108	Adeq.	399	14.4	6.8					Few		NS	NSA	Slight	

Table E-200a: Individual Animal Clinical Chemistry Data at Weaning

	58	133	6.8	93	25	22	11	<0.1	9.2	7.4	1.6	7.9	2.2	5.7	0.39	9	118	<2	87	<3
		Sodium	Potassium	Chloride	Bicarbonate	Anion Gap	Urea	Creatine-rb	Calcium	Phosphate	Magnesium-xb	Total Protein	Albumin-bulk	Globulin	A/G	Glucose	AST/P5P	SDH	Alk. Phos.	GGT
Clones	71	140	5.7	102	21	23	9	0.6	10.7	11.2	1.9	5.5	3.6	1.9	1.89	105	64	12	579	12
	72	140	5.5	102	30	14	7	0.5	10.9	11.3	1.9	5.1	3.5	1.6	2.19	121	64	14	616	10
	73	142	6.2	105	28	15	9	0.6	11	11.4	1.8	5.2	3.5	1.7	2.06	123	72	7	474	4
	74	139	4.7	98	29	17	10	0.7	11	10.9	1.7	5.6	3.5	2.1	1.67	113	54	16	531	12
	76	140	5.9	97	24	25	12	0.6	10	9.6	2.2	6.9	3.5	3.4	1.03	22	102	6	191	12
	78	140	5.7	101	27	18	14	0.6	11.2	9.8	2	6.2	3.8	2.4	1.58	119	76	31	627	16
	79	142	6.2	99	25	24	11	1.2	11.1	10.2	1.5	5.4	3.6	1.8	2	51	64	12	511	13
	81	141	5.7	98	25	24	8	0.5	10.9	8.6	2.1	7	3.5	3.5	1	<2	104	14	189	6
	82	140	6.5	100	24	23	11	0.4	10.8	8.3	2.2	7.9	3.2	4.7	0.68	2	110	<2	163	<3
	83	138	6.3	96	25	23	11	0.5	10.1	8.9	1.9	7.6	2.8	4.8	0.58	<2.1	82	9	159	11
	85	142	6	101	26	21	11	0.6	11.2	9	2	6.6	3.5	3.1	1.13	25	80	9	145	8
	87	141	6.1	95	27	25	14	0.4	10.6	11.1	1.9	7.4	3.3	4.1	0.8	17	139	26	222	15
	88	139	6.1	94	27	24	12	0.3	10.2	10.7	1.9	7.2	3.3	3.9	0.85	4	177	15	176	8
	89	137	6.5	95	27	22	11	0.2	10.4	10.1	2	7.1	3.1	4	0.78	22	142	22	212	<3
	90	138	5.3	98	22	23	11	0.3	10.3	8.4	2.2	6.7	3.4	3.3	1.03	14	141	<2	226	<3
	93	137	5.2	94	22	26	9	0.6	10.6	10.4	1.9	7.8	3	4.8	0.63	13	62	13	148	12
	94	139	5.7	99	24	22	11	0.4	10.5	9.5	2.3	6.9	3.4	3.5	0.97	9	116	2	177	<3
	96	137	5.2	95	25	22	17	0.5	10.6	10.3	2.2	7.5	3.7	3.8	0.97	51	85	6	223	<3
	100	135	5.3	96	24	20	6	0.5	10.8	9.1	2.1	8	3.4	4.6	0.74	78	67	11	265	22
	101	140	5.2	100	27	18	10	0.5	10	8.9	1.8	7.2	3.1	4.1	0.76	53	131	30	154	7
	102	138	5.4	96	26	21	15	0.4	10.3	9.5	1.7	6.9	3.3	3.6	0.92	32	110	5	158	<3
	106	138	6.2	101	26	17	10	0.7	10.9	9	1.8	5.9	3.6	2.3	1.57	88	69	11	670	10
	110	140	5.3	99	23	23	16	0.8	10.5	11.3	1.8	6.1	3.5	2.6	1.35	45	103	15	170	6
	111	140	5.4	99	24	22	17	1.1	10.1	11.8	1.8	6.5	3.5	3	1.17	35	94	25	124	14
	114	138	5.3	97	26	20	9	0.7	10.8	9.1	2	6.2	3.7	2.5	1.48	31	95	6	242	6
	115	143	5.2	102	26	20	12	0.6	11.6	9.9	1.9	5.8	3.8	2	1.9	60	79	35	336	10
	116	141	5.2	99	26	21	11	0.7	10	9.6	1.9	6.7	3.5	3.2	1.09	30	131	37	246	3
	117	142	5.4	101	27	19	9	0.4	11.6	9.8	1.8	6.9	3.3	3.6	0.92	38	92	84	261	11
	118	138	5.8	100	23	21	10	0.5	10.5	9.1	1.9	7.1	3.3	3.8	0.87	27	96	11	202	6
	119	138	5	96	28	19	9	0.8	11.3	9.6	1.6	5.7	3.5	2.2	1.59	59	59	25	379	21
	120	137	5	98	24	20	11	0.5	10.5	9	2	7.4	3.3	4.1	0.8	45	80	9	186	8
	121	141	5.5	102	23	22	8	0.6	10.4	9	1.9	6.1	3.8	2.3	1.65	42	81	15	277	11
	124	140	5.2	98	28	19	14	0.6	11.2	9.1	1.8	6.7	3.3	3.4	0.97	43	108	26	254	12
	126	141	6.3	102	24	21	8	0.3	10.9	9.6	2.2	5.9	3.5	2.4	1.46	33	86	<2	261	<3
	127	137	4.7	96	25	21	10	0.6	11.5	8.5	1.8	8	3.2	4.8	0.67	40	70	27	226	12
	128	140	5.5	98	24	24	8	0.4	10.9	9	1.9	6.5	3.6	2.9	1.24	13	94	2	232	10
	129	139	5.5	98	27	20	8	0.5	10.9	9.4	2.1	5.9	3.6	2.3	1.57	60	53	6	251	13
	130	142	5.9	102	25	21	9	0.3	11.7	9.2	2	5.4	3.5	1.9	1.84	60	73	7	693	6
	131	140	5	99	25	21	7	0.6	11.2	8.2	1.8	6.9	3.4	3.5	0.97	38	60	15	270	12
	132	143	5.5	100	25	24	7	0.7	10.7	8.8	2.1	5.9	3.7	2.2	1.68	23	44	9	258	20

Table E-200b: Individual Animal Clinical Chemistry Data at Weaning

0-6 months	Blood Chem	Total Bilirubin	Dir. Bilirubin	Ind. Bilirubin	Amylase	Cholesterol	CK	Iron	TIBC	% Saturation	hBA-random	Lipemia	Hemolysis	Icterus	IGF-1	Estradiol
	Ref Low Ref High Units	0 0.1 mg/dL	0 0 mg/dL	0 0.1 mg/dL		73 280 mg/dL	77 265 U/L	113 226 ug/dL	362 533 ug/dL	28 48 %	9 455 umol/L				34.7 472.27 ng/ml	10 40 pg/ml
Controls																
142		0.1	0	0.1	35	106	124	210	584	36	45.3	25	18	0		
143		0	0	0	38	110	244	184	432	43	18.7	0	31	0		
144		0.1	0	0.1	37	130	225	147	374	39	49.5	25	1	0		
145		0.1	0	0.1	36	119	287	145	370	39	28.4	24	12	0		
146		0	0	0	26	65	207	149	341	44	12.8	18	14	0		
147		0.1	0	0.1	37	75	256	141	412	34	29.7	12	13	0		
148		0.1	0	0.1	26	78	272	158	350	45	25.9	15	16	0		
149		0	0	0	18	92	268	180	438	41	14.9	5	49	w2		
150		0	0	0	86	88	197	156	411	38	26.2	14	8	0		
151		0.1	0.1	0	35	68	373	85	336	25	26.2	16	23	0		
152		0.1	0.1	0	28	66	185	138	371	37	52	15	2	0		
153		0.2	0.1	0.1	22	90	103	45	370	12	70.2	8	9	0		
155		0.1	0	0.1	20	72	213	120	369	33	43.4	19	6	0	114.97	>4.00
156		0.1	0	0.1	32	63	261	182	335	54	29.7	15	46	0	61.68	7.46
157		0.1	0	0.1	20	73	134	112	420	27	13.7	20	17	0	203.71	4.47
158		0.1	0	0.1	15	67	217	194	550	35	78.8	15	20	0	278.97	4.29
159		0.1	0	0.1	29	71	292	147	422	35	33.5	19	16	0	157.34	4
162		0.1	0	0.1	30	63	700	208	389	53	24.5	6	99	0	49.29	5.34
164		0.1	0	0.1	38	73	308	209	475	44	28.5	0	65	0	104.91	5.52
166		0.1	0	0.1	36	59	285	180	431	42	38.8	0	55	0	232.32	4.75
167		0	0	0	29	86	346	88	340	26	42.3	5	31	0	17.99	2.47
168		0.1	0	0.1	26	71	1427	135	403	33	26.6	11	28	0	96.89	6.84
170		0.1	0	0.1	33	78	168	209	449	47	14.2	6	16	0	220.82	7.78
171		0.1	0	0.1	37	84	249	211	459	46	27.8	5	23	0	282.16	4.07
172		0.1	0	0.1	41	81	169	187	487	38	17.5	8	93	0	255.05	3.12
173		0.1	0	0.1	41	78	161	184	449	41	14.1	11	12	0	127.16	8.1
174		0.1	0	0.1	44	74	183	215	511	42	28.2	3	7	0	408.27	10.63
175		0.1	0	0.1	32	76	180	186	417	45	21.1	0	13	0	136.19	7.06
176		0.1	0	0.1	27	83	215	187	391	48	17.6	5	21	0	146.96	7.23
177		0.1	0	0.1	47	96	248	186	423	44	27.1	1	8	0	283.27	4.48
178		0.1	0	0.1	29	81	258	142	345	41	11.7	6	78	0	125.93	6.91
179		0.1	0	0.1	52	60	299	202	467	43	14.6	14	55	0	369.11	7.05
180		0.1	0	0.1	47	68	147	176	411	43	9.7	4	32	0	205.57	12.31
181		0.1	0	0.1	33	91	248	195	386	51	27.1	1	8	0	82.3	4.74
182		0	0	0	35	53	161	129	422	31	11.6	6	19	0	478.03	2.92
183		0.1	0	0.1	12	82	426	199	437	46	24	15	14	0	1.76	5.29
184		0.1	0	0.1	40	80	168	167	424	39	12.8	2	7	0	257.93	14.87
185		0.1	0	0.1	29	37	183	145	395	37	17.6	0	17	0	159.15	2.89
196		0.1	0	0.1	30	50	213	136	316	43	79.5	18	11	0	17.29	18.48
197		0.1	0	0.1	26	58	373	243	442	55	43.1	10	39	0	28.12	7.03
198		0.2	0	0.2	34	54	126	130	337	39	40	15	15	0	25.95	4.82
199		0.1	0	0.1	49	38	233	139	494	28	38.9	11	17	0	8.41	4.17
200		0.1	0	0.1	16	123	184	27	540	5	33.5	17	0	0	108.21	4.82
201		0.1	0	0.1	33	109	72	90	564	16	24.1	17	19	0	204.5	6.29
202		0.2	0	0.2	22	93	165	29	620	5	18.2	20	6	0	216.46	8
203		0.1	0	0.1	24	89	96	22	600	4	21.7	6	14	0	192.22	5.39
204		0.1	0	0.1	32	99	16832	591	5	37.2	15	7	0	0	269.77	8.44
Clones																
39		0.2	0.1	0.1	19	86	281	81	432	19	15.4	0	16	0	113.37	4.73
40		0.1	0	0.1	22	116	230	180	511	35	37.7	9	36	0	252.12	7.04
41		0.1	0	0.1	26	98	243	155	353	44	37	7	51	0	114.43	4.24
42		0.1	0	0.1	28	65	540	188	426	44	17.9	13	18	0	274.83	9.34
56		0.1	0	0.1	18	109	238	244	441	55	18.2	26	67	0	270.54	5.4

Table E-200b: Individual Animal Clinical Chemistry Data at Weaning

	58	0.2	0	0.2	14	52	243	106	209	51	0	17	424	0	7.64	17.99
		Total Bilirubin	Dir. Bilirubin	Ind. Bilirubin	Amylase	Cholesterol	CK	Iron	TIBC	% Saturation	hBA-random	Lipemia	Hemolysis	Icterus	IGF-1	Estradiol
Clones	71	0.2	0	0.2	18	116	375	113	632	18	15.7	18	38	0		
	72	0.2	0	0.2	15	122	189	114	642	18	14.7	30	45	0	109.3	
	73	0.3	0	0.3	16	119	751	145	584	25	12.3	25	103	0		
	74	0.1	0	0.1	15	113	158	153	555	28	30.1	21	21	0	204.52	
	76	0.1	0	0.1	51	94	662	214	384	56	24.1	11	97	0	196.87	7.2
	78	0.2	0	0.2	29	99	209	273	640	43	39	9	27	0		
	79	0.2	0	0.2	28	153	208	131	465	28	19.9	22	58	0	149.6	<4
	81	0.2	0	0.2	23	66	1228	157	376	42	36.4	5	43	0	81.54	8.52
	82	0.2	0	0.2	23	90	505	169	379	45	28.2	14	114	0	92.21	6.01
	83	0.2	0	0.2	17	62	353	83	296	28	32.3	9	19	0	66.45	5.72
	85	0.1	0	0.1	29	77	340	140	416	34	34.6	3	37	0	60.33	10.38
	87	0.1	0	0.1	20	94	1215	203	391	52	17.1	8	9	0	150.05	5.21
	88	0.1	0	0.1	20	105	1265	187	321	58	20.7	10	115	0	110.26	7.77
	89	0.1	0	0.1	18	100	442	243	330	74	16	3	112	0	119.66	5.06
	90	0.2	0	0.2	24	90	2642	209	403	52	14	27	156	0	248.1	7.33
	93	0.1	0	0.1	23	74	151	159	337	47	16.6	16	5	0	124.6	5.14
	94	0.1	0	0.1	26	80	510	193	373	52	25.5	10	111	0	191.99	6.23
	96	0.1	0	0.1	29	74	282	190	465	41	10.5	11	125	0	294.68	5.25
	100	0.1	0	0.1	10	99	110	170	474	36	24.6	10	42	0	436.09	<4
	101	0.1	0	0.1	17	84	1225	164	340	48	17.3	9	102	0	67.59	6.34
	102	0.1	0	0.1	25	98	287	152	407	37	13.1	13	137	0	135.97	4.55
	106	0.2	0	0.2	24	121	556	118	449	26	45.2	17	88	0		
	110	0.1	0	0.1	20	98	238	168	361	47	53.9	2	56	0	256.46	6
	111	0.1	0.1	0	12	76	515	156	320	49	28.4	5	17	0	85.33	9.94
	114	0.1	0	0.1	29	72	234	238	429	55	11	0	70	0	213.19	5.47
	115	0.1	0	0.1	42	95	127	295	459	64	17.8	7	30	0	435	8.37
	116	0.1	0	0.1	35	83	1974	217	388	56	19.3	6	151	0	114.68	4.36
	117	0.1	0	0.1	32	109	127	208	382	54	31.7	21	96	0	260.65	3.63
	118	0.2	0	0.2	38	71	319	162	351	46	35.6	5	107	0	64.08	9.05
	119	0.2	0	0.2	35	121	125	217	431	50	18.1	25	32	0	227.89	<4
	120	0.1	0	0.1	8	71	217	170	358	47	43.1	7	57	0	155.03	5.88
	121	0.1	0	0.1	9	68	685	171	476	36	24.9	4	47	0	244.45	7.11
	124	0.1	0	0.1	31	50	841	143	384	37	103.1	22	33	0	143.7	<4
	126	0.1	0	0.1	24	107	129	204	467	44	27.7	14	253	0	253.54	5.51
	127	0.1	0	0.1	25	84	217	93	424	22	72.9	5	20	0	295.63	<4
	128	0.1	0	0.1	25	135	1056	196	459	43	17.3	8	161	0	113.91	6.71
	129	0.1	0	0.1	24	133	140	187	443	42	32.5	0	42	0	121.47	9.98
	130	0.2	0	0.2	18	127	110	283	547	52	15.4	5	153	0	387.73	8.32
	131	0.1	0	0.1	25	113	152	151	437	35	52.4	0	19	0	216.38	5.21
	132	0.1	0	0.1	28	136	492	197	466	42	24.7	8	24	0	96.56	8.45

Table E-300a: Individual Animal Clinical Chemistry Data at Adulthood

Adult	Blood Chem.	Sodium	Potassium	Chloride	Bicarbonate	Anion Gap	Urea	Creatine-rb	Calcium	Phosphate	Magnesium-xb	Total Protein	Albumin-bulk	Globulin	A/G	Glucose	AST/P5P	SDH
	Ref Low Ref High Units	134 145 mEq/L	3.9 5.3 mEq/L	94 105 mEq/L	22 30 mEq/L	17 24 mEq/L	10 25 mg/dL	0.4 1 mg/dL	8.3 10.4 mg/dL	4.2 7.7 mg/dL	1.7 2.2 mEq/L	7.2 9.4 g/dL	3.2 4.2 g/dL	3.5 5.8 g/dL	0.6 1.2	31 77 mg/dL	53 162 U/L	10 50 U/L
Controls	135	140	4.5	103	21	21	11	0.6	9.6	8.6	2	6.8	3.4	3.4	1	71	120	5
	154	138	4.7	98	27	18	12	0.7	10.9	8.1	2.1	6.7	3.7	3	1.23	88	68	18
	206	136	4.7	97	24	20	5	0.6	10.7	9.6	1.7	6.9	3.4	3.5	0.97	90	83	17
	160	147	5.9	101	23	29	21	0.9	10.1	9.3	1.9	7.3	3.9	3.4	1.15	31	84	23
	161	143	4.7	97	22	29	18	0.7	10.5	9.4	2	6.8	3.7	3.1	1.19	38	88	22
	162	140	4.8	98	23	24	16	0.7	9.7	8.5	1.8	6.4	3.1	3.3	0.94	8	65	13
	163	144	4.8	99	15	35	15	0.7	10.5	9	1.9	7.4	3.5	3.9	0.9	44	86	16
	165	146	4.9	99	18	34	16	0.7	10.6	8.7	1.8	7	3.9	3.1	1.26	22	100	18
	169	143	4.7	96	22	30	13	0.5	11	8.8	1.8	7.3	3.8	3.5	1.09	32	72	25
	205	139	4.5	95	26	23	15	0.8	10.7	8.3	1.8	7.8	3.3	4.5	0.73	81	88	15
	207	137	4.9	95	26	21	10	0.7	10.4	7.7	2	6.6	3.6	3	1.2	83	72	7
	208	139	4.5	96	27	21	10	0.4	10.2	8.9	1.9	7	3.4	3.6	0.94	95	77	4
	209	139	4.8	94	27	23	12	0.6	9.9	9.2	1.8	7	3.2	3.8	0.84	87	109	34
	210	140	4.8	96	26	23	13	0.8	10.1	7.7	2.1	7.1	3.5	3.6	0.97	92	98	15
	211	138	4.3	93	26	23	10	0.6	10.3	8.1	2	7	3.2	3.8	0.84	92	85	19
	212	142	5.5	98	26	24	13	0.6	10.7	8.1	2.1	7.5	3.4	4.1	0.83	90	79	30
	215	141	5.2	100	19	27	16	0.9	10.6	8.8	2	7	3.8	3.2	1.19	49	109	8
	216	143	4.6	101	21	26	18	0.8	9.5	9.5	1.8	6.5	3.5	3	1.17	80	99	25
	217	143	4.6	100	20	28	12	0.7	9.4	10.1	1.8	6.2	3.4	2.8	1.21	74	98	23
	218	141	4.7	99	21	26	18	0.7	10.3	9.7	1.9	6.6	3.7	2.9	1.28	58	74	23
	219	141	4.6	99	21	26	14	0.9	10	9.7	1.9	6.7	3.7	3	1.23	89	88	26
Clones	24	140	4.7	96	24	25	18	1	10.4	8	1.9	7.2	3.6	3.6	1	78	86	13
	25	141	4.3	96	25	24	12	1	10.4	8.1	1.8	6.8	3.6	3.2	1.13	72	95	16
	26	139	4.5	95	25	24	17	1	10	8.6	1.8	7	3.4	3.6	0.94	80	97	19
	27	139	5.2	94	25	25	13	0.9	10.5	8.7	1.9	7.2	3.5	3.7	0.95	78	100	14
	33	143	4.7	98	25	25	14	1.2	10.4	8.3	1.9	7.9	3.6	4.3	0.84	73	68	11
	35	143	4.7	98	26	24	14	0.9	10.2	8.1	2	7.6	3.4	4.2	0.81	75	73	14
	36	140	4.3	96	27	21	12	0.9	10	7.7	1.9	7.6	3.5	4.1	0.85	81	67	15
	38	139	4.2	96	25	22	15	1	9.7	9.1	1.9	7.1	3.4	3.7	0.92	88	75	15
	37	140	5	100	26	19	6	0.5	10.3	8.4	2	6.5	3.6	2.9	1.24	88	92	3
	55	140	5.4	99	25	21	13	0.6	10.8	9.9	2	6.6	3.8	2.8	1.36	45	81	7
	67	145	5	99	23	28	17	0.6	10.6	10.6	1.9	6.9	3.5	3.4	1.03	70	103	15
	69	144	4.8	97	26	26	14	0.7	10	9.3	1.7	7.5	3.1	4.4	0.7	73	85	22
	70	147	4.9	100	27	25	15	0.6	10.7	9.9	2.1	7.2	3.7	3.5	1.06	81	82	20
	98	136	4.2	96	27	17	11	0.6	10	8.5	1.8	6.8	3.7	3.1	1.19	91	59	15
	99	135	4.5	96	23	21	10	0.6	9.4	8.5	1.8	7.4	3.4	4	0.85	92	62	13
	103	144	6.6	100	23	28	13	0.2	10.7	10.2	2.1	7.1	3.7	3.4	1.09	12	133	7
	104	143	6.3	100	27	22	17	0.5	10.8	9.9	2	6.9	3.5	3.4	1.03	32	122	5
	108	137	5.5	97	24	22	18	1.1	10.6	9.2	1.9	6.7	3.7	3	1.23	24	97	15

Table E-300a: Individual Animal Clinical Chemistry Data at Adulthood

Adult	Blood Chem.	Alk. Phos.	GGT	Total Bilirubin	Dir. Bilirubin	Ind. Bilirubin	Amylase	Cholesterol	CK	Iron	TIBC	% Saturation	hBA-random	Lipemia	Hemolysis	Icterus	IGF-1	Estradiol
	Ref Low	23	11	0	0	0		73	77	113	362	28	9				34.7	10
	Ref High	78	39	0.1	0	0.1		280	265	226	533	48	455				472.27	40
	Units	U/L	U/L	mg/dL	mg/dL	mg/dL	U/L	mg/dL	U/L	ug/dL	ug/dL	%	umol/L				ng/ml	pg/ml
Controls	135	121	5	0.1	0	0.1	82	86	176	167	364	46	3.2	20	71	1		
	154	173	15	0.2	0	0.2	32	113	170	164	428	38	50.9	8	10	0	312.99	>4.0
	206	231	11	0.1	0	0.1	44	102	227	212	450	47	28.4	14	45	0	278.28	4.2
	160	182	15	0.1	0	0.1	38	113	153	183	476	38	34.2	5	32	0	324.29	11.14
	161	292	12	0.1	0	0.1	26	95	196	205	493	42	14.6	5	4	0	332.05	8
	162	167	9	0.1	0	0.1	23	69	141	224	343	65	20.4	0	26	0	43.75	4.1
	163	276	11	0.1	0	0.1	36	74	217	157	406	39	27.5	13	19	0	193.53	4.48
	165	191	14	0.1	0	0.1	48	88	207	239	494	48	28.6	12	42	0	291.66	6.93
	169	280	10	0.1	0	0.1	43	74	243	200	508	39	17.7	24	0	0	153.69	5.77
	205	134	16	0.1	0	0.1	45	105	176	114	411	28	15.1	25	14	0	732.75	9.15
	207	193	11	0.1	0	0.1	43	90	117	195	435	45	21.3	13	39	0	752.54	11.41
	208	178	<3	0.1	0	0.1	39	66	242	194	439	44	7.8	24	202	0	793.84	10.36
	209	151	23	0.1	0	0.1	23	67	150	156	410	38	15.1	5	25	0	732.63	10.35
	210	168	18	0.1	0	0.1	33	86	465	170	439	39	32.7	18	64	0	816.99	10.53
	211	169	20	0.1	0	0.1	29	58	168	164	482	34	7.9	17	24	0	861.08	7.91
	212	169	22	0.1	0	0.1	14	77	165	151	529	29	13.5	12	6	0	875.72	12.44
	215	158	<3	0.1	0	0.1	24	172	936	167	403	41	18.9	33	119	1	155.44	>4.0
	216	213	8	0.1	0	0.1	44	141	421	192	407	47	18	15	33	1	215.93	>4.0
	217	197	6	0	0	0	45	181	328	182	401	45	19.5	2	23	1	253.77	4.3
	218	127	8	0.1	0	0.1	44	160	253	180	376	48	20.9	7	41	2	218.14	5.26
	219	254	10	0.1	0	0.1	30	155	640	161	433	37	26.8	9	27	1	338.43	>4.0
Clones	24	109	13	0.1	0	0.1	46	104	181	132	409	32	17.1	16	8	0	924.04	14.16
	25	174	15	0.1	0	0.1	33	113	884	142	388	37	29.4	18	3	0	769.08	11.94
	26	151	13	0.1	0	0.1	34	113	161	130	359	36	20.2	15	2	0	381.04	10.62
	27	163	14	0.1	0	0.1	39	122	205	143	401	36	16.5	15	28	0	618.84	12.02
	33	104	16	0.1	0	0.1	34	81	145	150	470	32	31.5	14	9	0	915.45	24.33
	35	132	13	0.1	0	0.1	34	77	188	192	488	39	16.6	12	10	0	937.54	23.92
	36	113	16	0.1	0	0.1	35	78	103	155	479	32	12.3	11	5	0	875.68	15.4
	38	164	15	0.1	0	0.1	33	125	121	101	353	29	24.9	6	9	0	622.08	7.62
	37	136	3	0.1	0	0.1	37	85	432	179	478	37	14.4	22	84	1		
	55	184	7	0.1	0	0.1	43	129	177	230	466	49	27.9	28	60	0	414.52	9.6
	67	186	17	0.1	0	0.1	29	86	426	193	455	42	12.3	5	34	0	691.78	12.4
	69	138	22	0.1	0	0.1	27	88	263	156	385	41	10.7	25	0	0	445.23	15.82
	70	148	22	0.1	0	0.1	31	99	448	183	464	39	14.3	8	1	0	548.43	10.06
	98	231	11	0.1	0	0.1	39	91	196	132	363	36	23.5	5	8	0	279.26	>4.0
	99	192	11	0.1	0	0.1	37	96	217	129	331	39	20.8	9	5	0	217.88	4.28
	103	182	<3	0.1	0	0.1	33	122	1222	165	390	42	7.7	20	353	0	169.03	6.91
	104	179	<3	0.1	0	0.1	35	108	313	230	430	53	7.3	18	262	0	331.99	7.07
	108	111	12	0.1	0	0.1	33	111	227	165	304	54	29.5	20	30	0	33.25	7.5

Table E-300b: Individual Animal Hematology Data at Adulthood

Hemogram	Hematocrit (HCT)	Hemoglobin (HB)	RBC	MCV	MCH	MCHC	RDW	WBC	Seg. Neut.	Band. Neut.	Lymphocytes	Monocytes	Eosinophils	Basophils	
Ref Low	23	8.6	5.4	36	14	36	16.2	5.6	1.8	0	1.9	0	0	0	
Ref High	25	13.2	8.2	49	19	40	19.7	13.7	7.7	0	7.4	1.2	1.4	0.3	
Units	%	g/dL	mill/uL	fL	pg	g/dL	%	thou/uL	thou/uL	thou/uL	thou/uL	thou/uL	thou/uL	thou/uL	
Controls	135	33	11.8	7.5	44	16	35	18.4	6	3.7	0	1.7	0.1	0.5	0
	154	28	11	8.4	33	13	40	20.7	8.5	2.4	0	6.1	0	0	0
	206	28	10.8	8.9	32	12	39	23.3	13.2	5	0	7.5	0.7	0	0
	160	30	11.8	7.9	38	15	39	19.8	12.4	6	0	6.2	0.2	0	0
	161	34	11.4	8.6	39	13	34	22.2	13.4	3.1	0	10.1	0.3	0	0
	162	35	10.3	8.4	42	12	29	31.4	14.4	2.7	0	10.8	0.7	0	0.1
	163	34	10.3	9.4	37	11	30	28.3	14.6	4.5	0	9.9	0.1	0	0
	165	34	11.6	9.6	35	12	35	31.1	24.9	5.2	0	19.2	0.5	0	0
	169	33	11.5	9.7	34	12	35	21.1	12.2	4.5	0	7	0.2	0.5	0
	205	30	11.4	6.8	44	17	38	17.4	12.7	3.9	0	8.3	0.1	0.3	0.1
	207	29	11	7.3	39	15	38	18.4	8.6	2.6	0	5.8	0.1	0.1	0.1
	208														
	209	32	10.5	8.7	37	12	33	33.1	11.3	4.4	0	6.7	0.2	0	0
	210	34	10.6	9	38	12	31	32.1	10	4.4	0	5.1	0.4	0.1	0
	211	28	10.7	7.8	36	14	38	22.7	9.8	3.7	0	5.8	0.1	0.2	0
	212	34	10.5	8.6	39	12	31	30.2	12.5	5.1	0	6.8	0.6	0	0
	215	46	14.8	10.1	46	15	32	34.6	9.6	4.1	0	5.4	0	0.1	0
	216	32	12.7	9	36	14	40	24.1	8.4	2.8	0	5.5	0.1	0	0
	217	38	12.7	8	47	16	34	23.1	13.6	6.9	0	6.4	0.1	0.1	0
	218	42	13.8	10.2	42	14	33	36	9.9	2.6	0	6.8	0.4	0	0.1
	219	31	12.2	9	34	14	40	22.3	12.4	6.9	0	4.8	0.5	0	0.1
Clones	24	33	11	6.9	48	16	33	20.2	11.6	6.4	0	4.8	0.5	0	0
	25	32	10.7	6.9	46	16	34	23.8	13.9	6.1	0	7.2	0.6	0	0
	26	32	10.6	6.8	47	16	34	21.4	9.5	2.4	0	6.7	0.2	0.2	0
	27	32	10.5	6.7	47	16	33	20.5	7.8	2.5	0	4.8	0.3	0.2	0
	33	33	10.7	7.7	43	14	32	33	10.2	2.1	0	7	0.5	0.4	0.1
	35	34	11.1	7.7	44	15	33	33.9	10.9	2.7	0	7.7	0.2	0.1	0.1
	36	30	9.9	6.9	44	14	33	32.2	9.2	3	0	5.9	0.1	0.1	0.1
	38	31	9.7	6.6	46	15	32	28.3	10.5	4.7	0	5.4	0	0.4	0
	37	35	12.2	8.7	40	14	35	20.1	8.1	2.3	0	5.2	0.6	0.1	0
	55	30	11.1	8.6	35	13	37	21.7	10.5	2.1	0	7.6	0.5	0.3	0
	67	34	11.1	9.5	36	12	32	32.4	9.5	4.8	0	3.8	0.3	0.6	0
	69	31	10.1	9.5	33	11	33	29.5	8.2	2.5	0	5.4	0.1	0.2	0
	70	33	10.8	9.9	34	11	32	31.7	8.2	2.5	0	5.2	0.1	0.3	0
	98	28	10.8	8.9	32	12	38	22.6	8.1	2.1	0	5.4	0.2	0.1	0.3
	99	28	10.6	9.3	30	12	38	20.4	8.5	2.4	0	5.7	0.4	0	0
	103	28	10.5	7.2	40	15	37	21.6	8.6	3.5	0	4.9	0.2	0	0
	104	28	10.9	7.4	38	15	39	19.9	8.8	5.5	0	3	0.2	0.2	0
	108	37	13.8	11.9	31	12	37	19.4	8.2	3.5	0	3.9	0.3	0.2	0.2

Notes:

- NSA No Significant Abnormalities
- NS None Seen
- N Normal
- SCM Smudged Cells-Moderate

Table E-300b: Individual Animal Hematology Data at Adulthood

Hemogram		Platelet Smear	Platelets	MPV	TP-Ref	RBC Morphology	Acanthocytes	Anisocytosis	Echinocytes	Poikilocytes	Schistocytes	Parasites	WBC Exam	Plasma Appearance
Ref Low			232	5.6	5.9									
Ref High			596	8.2	8.1									hemolysis
Units			thou/uL	fL	g /dL									
Controls	135	Incr.	812	16.8	8.3	NSA						NS	NSA	Moderate
	154	Adeq.	391	5.8	6.7			Mild				NS	NSA	N
	206	Incr. (*1)	601	5.5	6.8	NSA						NS	NSA	N
	160	Adeq.	259	6.6	7.3	NSA						NS	NSA	N
	161	Adeq.	391	7.4	7				Moderate			NS	NSA	N
	162	Adeq.	487	6.8	6.5				Moderate			NS	NSA	N
	163	Adeq.	515	6.3	7.2				Many			NS	NSA	N
	165	Adeq.	279	6.2	6.8	NSA						NS	Few	N
	169	Adeq. (*1)	559	5.9	7.2	NSA						NS	NSA	N
	205	Adeq.	309	10.7	7	NSA						NS	NSA	Slight
	207	Adeq. (*1)			6.6	NSA						NS	NSA	Slight
	208													
	209	Adeq. (*1)			7				Mild			NS	NSA	Slight
	210	Adeq. (*1)	117	6.4	7.3			Few				NS	NSA	Slight
	211	Adeq.	414	7.3	7				Mild			NS	NSA	N
	212	Adeq.	351	6.5	7.4	NSA						NS	NSA	N
	215	Adeq. (*1)	242	5.9	7.5	NSA						NS	NSA	Slight
	216	Incr.	671	6.3	6.7	NSA						NS	NSA	Slight
	217	Adeq.	468	6.3	6.5				Mild			NS	NSA	Slight
218	Adeq.	422	6	6.9	NSA						NS	NSA	Slight	
219	Adeq.	583	6.3	7				Mild	Few		NS	NSA	Slight	
Clones	24	Adeq.	398	7.2	7.3	NSA						NS	NSA	Slight
	25	Adeq.	313	6.7	6.8	NSA						NS	NSA	N
	26	Adeq. (*1)			7.2	NSA						NS	NSA	Slight
	27	Adeq. (*1)			7.2	NSA						NS	NSA	N
	33	Adeq.	392	5.8	7.8	NSA						NS	NSA	N
	35	Adeq. (*1)			7.4	NSA						NS	NSA	Slight
	36	Adeq.	335	6.6	7.4	NSA						NS	NSA	N
	38	Adeq. (*1)			7.5	NSA						NS	NSA	Slight
	37	Adeq.	328	12.4	7.5	NSA						NS	NSA	Slight
	55	Adeq. (*1)			6.8	NSA								Slight
	67	Adeq. (*1)			6.9	NSA						NS	NSA	Slight
	69	Incr.	658	5.9	7.7	NSA						NS	NSA	N
	70	Incr.	605	6	7.3	NSA						NS	NSA	N
	98	Adeq. (*1)	395	6.8	6.7				Mild			NS	NSA	N
	99	Incr. (*1)	622	5.5	7.4				Mild			NS	NSA	N
	103	Adeq.	337	11	6.8					Mod		NS	NSA	N
	104	Incr.	745	6.4	7.4	NSA						NS	NSA	N
108	Adeq.	399	14.4	6.8					Few		NS	NSA	Slight	

Cyagra Meat Composition Study Animal Information								
Updated	27-Jul-04							
	Calf #	Breed	Sex	Age (Months)	DOB	Assay Date	Age (Mo)	Sample ID
Clones	33-1	Cross Breed	F	24	3-Apr-02	19-Mar-04	24	A-F1
	39-2	Angus	F	27	12-Jan-02	19-Mar-04	27	A-F2
	263	Brangus	F	43	29-Aug-00	19-Mar-04	43	A-F3
	264	Brangus	F	43	29-Aug-00	19-Mar-04	43	A-F4
	265	Brangus	F	43	29-Aug-00	19-Mar-04	43	A-F5
	144-2	Holstein	F	15	12-Dec-02	19-Mar-04	15	A-F6
	232-3	R&W Holstein	M	17	10-Feb-03	6-Jul-04	17	A-M1
	232-4	R&W Holstein	M	17	13-Feb-03	6-Jul-04	17	A-M2
	248-4	Holstein	M	17	12-Feb-03	6-Jul-04	17	A-M3
	248-8	Holstein	M	17	21-Feb-03	6-Jul-04	17	A-M4
	200-1	Angus	M	12	8-Jul-03	6-Jul-04	12	A-M5
Controls	247bx	Cross Breed	F	>12	adult	14-Apr-04	>12	B-F1
	248bx	Cross Breed	F	>12	adult	14-Apr-04	>12	B-F2
	261bx	Red Angus	F	>12	adult	14-Apr-04	>12	B-F3
	271bx	Cross Breed	F	>12	adult	14-Apr-04	>12	B-F4
	278bx	Cross Breed	F	>12	adult	14-Apr-04	>12	B-F5
	919g	Holstein	F	>12	5-Oct-02		>12	B-F6
	441g	R&W Holstein	M	17	23-Feb-03	6-Jul-04	17	B-M1
	444g	R&W Holstein	M	16	1-Mar-03	6-Jul-04	16	B-M2
	440g	Holstein	M	17	19-Feb-03	6-Jul-04	17	B-M3
	445g	Holstein	M	17	23-Feb-03	6-Jul-04	17	B-M4
	336or	Holstein	M	20	18-Nov-02	6-Jul-04	20	B-M5

Cyagra Meat Study

SampleNo		36108	36115	36116	36117	36118	36119	36120	36121	36122
Marked ID		AF1 - 1	AF1 - 2	AF1 - 3	AF2- 1	AF2- 2	AF2- 3	AF3- 1	AF3- 2	AF3- 3
Analyte	Units	Result	Result	Result	Result	Result	Result	Result	Result	Result
Crude Fat	%	16.7	12.2	46.5	17.5	14.9	30.1	8.1	15.2	27
Moisture	%	62.22	68.38	41.42	61.92	62.86	49.17	69.25	60.93	55.54
Protein - Combustion	%	20.55	20.01	12.14	19.78	18.64	15.34	20.46	18.72	16.15
Protein - Kjeldahl	%	20.38	20.28	12.07	19.31	18.77	15.54	20.69	19.17	16.58
Ash	%	0.95	0.93	0.6	0.89	0.91	0.85	1.16	0.87	1.48
Balance (protein+moist+ash+fat)	%	100.25	101.79	100.59	99.62	97.44	95.66	99.20	96.17	100.60
Amino Acid Profile (results below)										
Tryptophan	%	0.24	0.25	0.14	0.24	0.23	0.2	0.26	0.23	0.19
Aspartic Acid	%	1.82	1.87	1.14	1.79	1.76	1.51	1.97	1.6	1.4
Threonine	%	0.78	0.89	0.55	0.87	0.85	0.74	0.9	0.69	0.6
Serine	%	0.79	0.77	0.47	0.74	0.74	0.62	0.7	0.51	0.45
Glutamic Acid	%	2.82	3.01	1.8	2.86	2.86	2.46	3.17	2.59	2.26
Proline	%	0.8	0.85	0.5	0.83	0.85	0.67	1.05	0.82	0.84
Glycine	%	0.91	0.93	0.57	0.92	0.91	0.69	1.25	0.95	1.03
Alanine	%	1.15	1.19	0.73	1.14	1.12	0.95	1.34	1.05	0.97
Cystine	%	0.23	0.22	0.14	0.2	0.21	0.16	0.23	0.21	0.18
Valine	%	0.58	0.67	0.45	0.7	0.7	0.63	1.08	0.86	0.74
Methionine	%	0.55	0.51	0.31	0.49	0.48	0.37	0.53	0.5	0.41
Isoleucine	%	0.52	0.6	0.39	0.66	0.65	0.57	0.97	0.78	0.66
Leucine	%	1.37	1.46	0.91	1.44	1.43	1.25	1.65	1.34	1.14
Tyrosine	%	0.62	0.65	0.4	0.63	0.62	0.54	0.7	0.56	0.46
Phenylalanine	%	0.71	0.75	0.47	0.74	0.74	0.65	0.88	0.71	0.61
Histidine	%	0.59	0.61	0.41	0.62	0.6	0.52	0.8	0.62	0.54
Lysine, Total	%	1.49	1.61	0.98	1.57	1.57	1.36	1.82	1.49	1.27
Arginine	%	1.11	1.2	0.73	1.13	1.16	0.97	1.4	1.13	1.02
Hydroxyproline	%	0.19	0.17	0.07	0.16	0.25	0.09	0.07	0.3	0.3
Fatty Acid (results below)										
C08:0 Octanoic (Caprylic)	%	0	0	0	0	0	0	0	0	0
C10:0 Decanoic (Capric)	%	0	0	0	0	0	0	0	0	0.01
C11:0 Undecanoic (Hendecanoic)	%	0	0	0	0	0	0	0	0	0
C12:0 Dodecanoic (Lauric)	%	0	0	0.02	0	0	0.01	0	0	0.02
C14:0 Tetradecanoic (Myristic)	%	0.36	0.23	1.02	0.43	0.35	0.74	0.22	0.36	0.67
C14:1 Tetradecenoic (Myristoleic)	%	0.28	0.14	0.5	0.41	0.26	0.51	0.19	0.22	0.2
C15:0 Pentadecanoic	%	0.05	0.04	0.17	0.06	0.05	0.12	0.02	0.06	0.11
C15:1 Pentadecenoic	%	0	0	0	0	0	0	0	0	0
C16:0 Hexadecanoic (Palmitic)	%	3.54	2.53	10.1	3.82	3.36	6.75	1.95	3.71	6.55

Cyagra Meat Study

SampleNo		36108	36115	36116	36117	36118	36119	36120	36121	36122
Marked ID		AF1 - 1	AF1 - 2	AF1 - 3	AF2- 1	AF2- 2	AF2- 3	AF3- 1	AF3- 2	AF3- 3
Analyte	Units	Result	Result	Result	Result	Result	Result	Result	Result	Result
C16:1 Hexadecenoic (Palmitoleic)	%	1.19	0.68	2.33	1.58	1.1	2.05	0.8	1.05	1.12
C16:2 Hexadecadienoic	%	0.1	0.07	0.34	0.11	0.08	0.22	0.04	0.11	0.2
C16:3 Hexadecatrienoic	%	0	0	0	0	0	0	0	0	0
C16:4 Hexadecatetraenoic	%	0	0	0	0.01	0	0	0	0	0
C17:0 Heptadecanoic (Margaric)	%	0.1	0.09	0.4	0.1	0.1	0.24	0.04	0.13	0.28
C17:1 Heptadecenoic Margaroleic	%	0.18	0.12	0.46	0.21	0.16	0.33	0.09	0.15	0.23
C18:0 Octadecanoic (Stearic)	%	1.06	1.06	4.6	0.98	1.11	2.41	0.42	1.34	3.34
C18:1 Octadecenoic (Oleic)	%	7.93	5.6	20.6	8.02	6.65	13	3.57	6.64	11.6
C18:2 Octadecadienoic (Linoleic)	%	0.38	0.31	1.21	0.39	0.35	0.72	0.18	0.41	0.73
C18:3 Octadecatrienoic (Linolenic)	%	0.07	0.05	0.25	0.07	0.06	0.12	0.04	0.08	0.17
C18:4 Octadecatetraenoic	%	0.15	0.09	0.38	0.1	0.08	0.19	0.05	0.1	0.15
C19:0 Nonadecanoic	%	0	0	0	0	0	0	0	0	0
C20:0 Eicosanoic (Arachidic)	%	0	0	0	0	0	0	0	0	0.02
C20:1 Eicosenoic (Gadoleic)	%	0.06	0.05	0.19	0.05	0.04	0.1	0.02	0.04	0.08
C20:2 Eicosadienoic	%	0	0	0.02	0	0	0	0	0	0
C20:3 Eicosatrienoic	%	0	0	0	0	0	0	0	0	0.01
C20:4 Eicosatetraenoic (Arachidonic)	%	0	0	0	0.01	0	0.02	0	0	0.01
C20:5 Eicosapentaenoic	%	0	0	0	0	0	0	0	0	0
C21:0 Heneicosanoic	%	0	0	0	0	0	0	0	0	0
C21:5 Heneicosapentaenoic	%	0	0	0	0	0	0	0	0	0
C22:0 Docosanoic (Behenic)	%	0	0	0	0	0	0	0	0	0
C22:1 Docosenoic (Erucic)	%	0	0	0	0	0	0	0	0	0
C22:2 Docosadienoic	%	0	0	0	0	0	0	0	0	0
C22:3 Docosatrienoic	%	0	0	0	0	0	0	0	0	0
C22:4 Docosatetraenoic	%	0	0	0	0	0	0	0	0	0
C22:5 Docosapentaenoic	%	0	0	0	0	0	0	0	0	0
C22:6 Docosahexaenoic	%	0	0	0	0	0	0	0	0	0
C24:0 Tetracosanoic (Lignoceric)	%	0	0	0	0	0	0	0	0	0
C24:1 Tetracosenoic (Nervonic)	%	0	0	0	0	0	0	0	0	0
Total Monounsatur. Fatty Acids Calc.	%	9.64	6.59	24	10.3	8.21	16	4.66	8.11	13.2
Total Polyunsatur. Fatty Acids Calc.	%	0.71	0.54	2.19	0.71	0.58	1.29	0.32	0.72	1.27
Saturated Fatty Acids	%	5.11	3.95	16.3	5.41	4.98	10.3	2.66	5.61	11
Total Fat (as triglycerides)	%	16.2	11.6	44.6	17.2	14.4	28.9	7.99	15.2	26.8
Calcium	mg/100g	3.4	4	20	3.7	5.2	53	4.7	4.8	15
Iron	mg/100 g	1.9	2	1.6	2.1	4.1	2.2	2.3	4.1	3.8

Cyagra Meat Study

SampleNo		36108	36115	36116	36117	36118	36119	36120	36121	36122
Marked ID		AF1 - 1	AF1 - 2	AF1 - 3	AF2- 1	AF2- 2	AF2- 3	AF3- 1	AF3- 2	AF3- 3
Analyte	Units	Result	Result	Result	Result	Result	Result	Result	Result	Result
Phosphorus	mg/100 g	170	160	110	160	150	150	190	150	240
Zinc	mg/100 g	3.5	4.4	2.2	3.8	4.2	3.1	4	4	3.1
Cholesterol	mg/100g	64.9	58	72.4	77.5	69.9	87.1	52.2	59.6	72.5
Niacin	mg/100 g	5.12	3.95	2.52	4.6	4.45	3.13	5.63	3.47	3.13
Vitamin B1 - Thiamine Hydrochloride	mg/100 g	0.0665	0.0728	0.055	0.0505	0.0592	0.0408	0.0432	0.0522	0.0624
Vitamin B2 - Riboflavin	mg/100 g	0.192	0.2	0.166	0.223	0.231	0.256	0.237	0.167	0.196
Vitamin B6	mg/100 g	0.382	0.32	0.151	0.284	0.23	0.217	0.358	0.307	0.198
Vitamin B12	mg/100 g	0.00229	0.00344	0.00216	0.00248	0.00284	0.00372	0.00267	0.00268	0.00278
Vitamin E	IU/100g	0.505	0.382	0.774	0.841	0.5	0.718	0.615	0.627	0.66

Cyagra Meat Study

SampleNo	36123	36124	36125	36126	36127	36128	36129	36131	36132	36133
Marked ID	AF4- 1	AF4- 2	AF4- 3	AF5- 1	AF5- 2	AF5- 3	AF6- 1	AF6- 2	AF6- 3	AM1- 1
Analyte	Result	Result	Result	Result	Result	Result	Result	Result	Result	Result
Crude Fat	13.5	33	13	10.7	7.7	18.1	9.3	7.5	6.8	6.6
Moisture	62.21	50.03	65.91	67.55	68.82	61.96	69.23	72.23	69.68	69.12
Protein - Combustion	18.82	15.95	20.49	18.84	21	19.34	21.89	20.15	23.04	21.45
Protein - Kjeldahl	18.77	15.73	20.61	19.34	20.97	19.76	22.17	20.35	22.54	22.15
Ash	0.84	0.81	0.9	0.9	0.96	0.88	1.08	1.02	1.07	1.11
Balance (protein+moist+ash+fat)	95.32	99.57	100.42	98.49	98.45	100.70	101.78	101.10	100.09	98.98
Amino Acid Profile (results below)										
Tryptophan	0.22	0.2	0.26	0.23	0.26	0.24	0.27	0.27	0.26	0.27
Aspartic Acid	1.74	1.49	1.91	1.81	2	1.84	2.01	1.98	2.12	2.12
Threonine	0.84	0.72	0.93	0.88	0.97	0.91	0.99	0.97	1.06	0.92
Serine	0.74	0.62	0.8	0.76	0.84	0.77	0.85	0.84	0.87	0.68
Glutamic Acid	2.88	2.42	3.12	2.97	3.26	3	3.33	3.41	3.57	3.44
Proline	0.81	0.64	0.86	0.81	0.83	0.84	0.93	0.92	1.09	1.04
Glycine	0.91	0.68	0.94	0.88	0.84	0.88	0.94	0.95	1.3	1.14
Alanine	1.13	0.94	1.23	1.16	1.24	1.18	1.29	1.26	1.46	1.36
Cystine	0.2	0.17	0.22	0.21	0.23	0.22	0.25	0.24	0.24	0.25
Valine	0.72	0.59	0.76	0.72	0.81	0.78	0.85	0.85	1.06	1.15
Methionine	0.47	0.4	0.53	0.49	0.57	0.53	0.57	0.56	0.57	0.57
Isoleucine	0.68	0.53	0.71	0.65	0.76	0.71	0.8	0.78	0.94	1.04
Leucine	1.43	1.22	1.56	1.48	1.66	1.52	1.68	1.64	1.8	1.79
Tyrosine	0.62	0.54	0.67	0.65	0.73	0.65	0.71	0.71	0.76	0.74
Phenylalanine	0.74	0.63	0.79	0.78	0.88	0.77	0.85	0.84	0.93	0.94
Histidine	0.6	0.54	0.69	0.64	0.71	0.69	0.72	0.64	0.76	0.82
Lysine, Total	1.56	1.31	1.69	1.62	1.81	1.65	1.81	1.8	1.95	1.98
Arginine	1.16	0.96	1.24	1.18	1.29	1.23	1.32	1.34	1.51	1.45
Hydroxyproline	0.21	0.12	0.07	0.1	0.04	0.16	0.15	0.12	0.28	0.19
Fatty Acid (results below)										
C08:0 Octanoic (Caprylic)	0	0	0	0	0	0	0	0	0	0
C10:0 Decanoic (Capric)	0	0.01	0	0	0	0	0	0	0	0
C11:0 Undecanoic (Hendecanoic)	0	0	0	0	0	0	0	0	0	0
C12:0 Dodecanoic (Lauric)	0	0.02	0	0.01	0	0.01	0	0	0	0
C14:0 Tetradecanoic (Myristic)	0.3	0.87	0.33	0.26	0.2	0.48	0.24	0.13	0.14	0.16
C14:1 Tetradecenoic (Myristoleic)	0.21	0.54	0.31	0.09	0.06	0.4	0.04	0.03	0.05	0.04
C15:0 Pentadecanoic	0.05	0.14	0.04	0.05	0.03	0.06	0.04	0.03	0.02	0.03
C15:1 Pentadecenoic	0	0	0	0	0	0	0	0	0	0
C16:0 Hexadecanoic (Palmitic)	3.08	7.85	2.98	2.73	1.97	4.33	2.21	1.61	1.52	1.48

Cyagra Meat Study

SampleNo	36123	36124	36125	36126	36127	36128	36129	36131	36132	36133
Marked ID	AF4- 1	AF4- 2	AF4- 3	AF5- 1	AF5- 2	AF5- 3	AF6- 1	AF6- 2	AF6- 3	AM1- 1
Analyte	Result	Result	Result	Result	Result	Result	Result	Result	Result	Result
C16:1 Hexadecenoic (Palmitoleic)	1.02	2.27	1.32	0.51	0.33	1.66	0.25	0.25	0.33	0.28
C16:2 Hexadecadienoic	0.09	0.26	0.08	0.07	0.05	0.12	0.08	0.05	0.04	0.05
C16:3 Hexadecatrienoic	0	0	0	0	0	0	0	0	0	0
C16:4 Hexadecatetraenoic	0	0	0	0	0	0	0	0	0	0
C17:0 Heptadecanoic (Margaric)	0.1	0.26	0.07	0.1	0.07	0.1	0.1	0.08	0.06	0.07
C17:1 Heptadecenoic Margaroleic	0.13	0.35	0.14	0.09	0.06	0.19	0.05	0.06	0.06	0.05
C18:0 Octadecanoic (Stearic)	0.99	2.52	0.63	1.17	0.92	0.96	1.94	1.1	0.78	1.01
C18:1 Octadecenoic (Oleic)	5.67	14.1	5.67	4.3	3.01	7.94	3.25	3.25	2.82	2.75
C18:2 Octadecadienoic (Linoleic)	0.33	0.77	0.28	0.29	0.2	0.37	0.36	0.24	0.17	0.25
C18:3 Octadecatrienoic (Linolenic)	0.07	0.16	0.06	0.06	0.04	0.08	0.03	0.03	0.02	0.02
C18:4 Octadecatetraenoic	0.09	0.21	0.1	0.05	0.03	0.13	0.03	0.05	0.04	0.03
C19:0 Nonadecanoic	0	0	0	0	0	0	0	0	0	0
C20:0 Eicosanoic (Arachidic)	0	0	0	0	0	0	0.01	0	0	0
C20:1 Eicosenoic (Gadoleic)	0.04	0.11	0.04	0.02	0.02	0.05	0.02	0.03	0.02	0.02
C20:2 Eicosadienoic	0	0	0	0	0	0	0	0	0	0
C20:3 Eicosatrienoic	0	0	0	0	0	0	0	0	0	0
C20:4 Eicosatetraenoic (Arachidonic)	0	0	0	0	0.01	0	0.02	0.02	0.01	0.01
C20:5 Eicosapentaenoic	0	0	0	0	0	0	0	0	0	0
C21:0 Heneicosanoic	0	0	0	0	0	0	0	0	0	0
C21:5 Heneicosapentaenoic	0	0	0	0	0	0	0	0	0	0
C22:0 Docosanoic (Behenic)	0	0	0	0	0	0	0	0	0	0
C22:1 Docosenoic (Erucic)	0	0	0	0	0	0	0	0	0	0
C22:2 Docosadienoic	0	0	0	0	0	0	0	0	0	0
C22:3 Docosatrienoic	0	0	0	0	0	0	0	0	0	0
C22:4 Docosatetraenoic	0	0	0	0	0	0	0	0	0	0
C22:5 Docosapentaenoic	0	0	0	0	0	0	0	0	0	0
C22:6 Docosahexaenoic	0	0	0	0	0	0	0	0	0	0
C24:0 Tetracosanoic (Lignoceric)	0	0	0	0	0	0	0	0	0	0
C24:1 Tetracosenoic (Nervonic)	0	0	0	0	0	0	0	0	0	0
Total Monounsat. Fatty Acids Calc.	7.07	17.3	7.48	5.01	3.48	10.3	3.62	3.62	3.28	3.14
Total Polyunsat. Fatty Acids Calc.	0.59	1.41	0.53	0.49	0.36	0.72	0.53	0.42	0.31	0.38
Saturated Fatty Acids	4.53	11.7	4.06	4.33	3.21	5.95	4.56	2.97	2.54	2.76
Total Fat (as triglycerides)	12.8	31.9	12.7	10.3	7.4	17.8	9.14	7.35	6.43	6.59
Calcium	5.1	4	5.7	3.4	12	6.3	31	3.4	6.4	16
Iron	2.1	2	2	2.5	2.7	2	1.9	2	1.9	2.7

Cyagra Meat Study

SampleNo	36123	36124	36125	36126	36127	36128	36129	36131	36132	36133
Marked ID	AF4- 1	AF4- 2	AF4- 3	AF5- 1	AF5- 2	AF5- 3	AF6- 1	AF6- 2	AF6- 3	AM1- 1
Analyte	Result	Result	Result	Result	Result	Result	Result	Result	Result	Result
Phosphorus	140	150	170	160	170	160	200	170	190	190
Zinc	4.2	2.9	3.8	4.4	4.3	3.4	3.5	4.3	3.6	3.8
Cholesterol	72.5	71.6	61.1	58.6	60.4	61.9	72.7	61.1	70.7	78.3
Niacin	3.99	3.78	5.52	4.52	4.3	4.52	4.47	4.29	5.73	4.93
Vitamin B1 - Thiamine Hydrochloride	0.073	0.0811	0.0775	0.144	0.0918	0.0851	0.0732	0.0929	0.0777	0.0938
Vitamin B2 - Riboflavin	0.216	0.2	0.198	0.184	0.244	0.185	0.272	0.266	0.276	0.309
Vitamin B6	0.195	0.225	0.347	0.321	0.369	0.337	0.369	0.291	0.291	0.358
Vitamin B12	0.00311	0.00333	0.00226	0.00318	0.00299	0.00233	0.00347	0.0033	0.00336	0.00407
Vitamin E	0.611	0.649	0.543	0.37	0.469	0.539	0.382	0.517	0.333	0.302

Cyagra Meat Study

SampleNo	36134	36135	36136	36137	36138	36139	36140	36142	36143	36144
Marked ID	AM1- 2	AM1- 3	AM2- 1	AM2- 2	AM2- 3	AM3- 1	AM3- 2	AM3- 3	AM4- 1	AM4- 2
Analyte	Result	Result	Result	Result	Result	Result	Result	Result	Result	Result
Crude Fat	1.9	6.4	13.7	3.1	3.8	5.5	2.4	1.4	1.4	3.6
Moisture	69.9	71.8	66.28	70.01	72.83	68.5	74.32	73.93	73.37	73.32
Protein - Combustion	26.19	21.4	20.52	25.16	22.52	21.4	21.97	23.44	23.79	22.06
Protein - Kjeldahl	25.67	21.34	20.66	26.44	22.5	21.06	21.95	23.29	23.52	22.41
Ash	1.17	1.11	1.06	1.11	0.97	1.08	1.14	1.21	1.14	1.1
Balance (protein+moist+ash+fat)	98.64	100.65	101.70	100.66	100.10	96.14	99.81	99.83	99.43	100.43
Amino Acid Profile (results below)										
Tryptophan	0.31	0.25	0.25	0.3	0.27	0.26	0.26	0.29	0.3	0.29
Aspartic Acid	2.71	2.11	1.98	2.36	2.16	2.07	2.15	2.34	2.24	2.18
Threonine	1.19	0.98	0.96	1.16	1.02	1	1	1.11	1.1	1.05
Serine	0.85	0.86	0.84	0.99	0.91	0.82	0.91	0.94	0.91	0.91
Glutamic Acid	4.39	3.47	3.29	3.91	3.55	3.44	3.71	3.91	3.68	3.61
Proline	1.33	1.3	0.93	1.17	1.19	0.88	1.01	1.13	1.06	1.07
Glycine	1.41	1.63	1.03	1.29	1.39	0.94	1.18	1.28	1.15	1.14
Alanine	1.73	1.48	1.28	1.55	1.44	1.3	1.38	1.53	1.44	1.39
Cystine	0.32	0.23	0.23	0.28	0.23	0.25	0.25	0.27	0.27	0.26
Valine	1.47	1.01	0.89	1.06	0.92	1.04	1.08	1.19	0.98	0.94
Methionine	0.77	0.48	0.55	0.65	0.55	0.58	0.59	0.62	0.64	0.59
Isoleucine	1.34	0.89	0.81	0.95	0.82	0.98	1	1.13	0.89	0.86
Leucine	2.26	1.7	1.67	1.96	1.74	1.75	1.82	1.96	1.82	1.78
Tyrosine	0.95	0.72	0.72	0.86	0.75	0.76	0.78	0.8	0.8	0.77
Phenylalanine	1.2	0.91	0.86	1.01	0.91	0.92	0.97	1.01	0.94	0.93
Histidine	1.05	0.73	0.72	0.87	0.71	0.78	0.77	0.83	0.77	0.74
Lysine, Total	2.47	1.86	1.81	2.13	1.9	1.93	2.05	2.21	2	1.97
Arginine	1.82	1.54	1.36	1.61	1.51	1.39	1.51	1.57	1.51	1.47
Hydroxyproline	0.17	0.29	0.15	0.28	0.18	0.09	0.24	0.16	0.12	0.22
Fatty Acid (results below)										
C08:0 Octanoic (Caprylic)	0	0	0	0	0	0	0	0	0	0
C10:0 Decanoic (Capric)	0	0	0	0	0	0	0	0	0	0
C11:0 Undecanoic (Hendecanoic)	0	0	0	0	0	0	0	0	0	0
C12:0 Dodecanoic (Lauric)	0	0	0	0	0	0	0	0	0	0
C14:0 Tetradecanoic (Myristic)	0.05	0.14	0.37	0.08	0.09	0.11	0.04	0.03	0.03	0.07
C14:1 Tetradecenoic (Myristoleic)	0.02	0.03	0.14	0.03	0.03	0	0	0	0	0.02
C15:0 Pentadecanoic	0	0.03	0.05	0.01	0.01	0.03	0	0	0	0
C15:1 Pentadecenoic	0	0	0	0	0	0	0	0	0	0.01
C16:0 Hexadecanoic (Palmitic)	0.46	1.41	2.96	0.66	0.79	1.22	0.42	0.3	0.31	0.75

Cyagra Meat Study

SampleNo	36134	36135	36136	36137	36138	36139	36140	36142	36143	36144
Marked ID	AM1- 2	AM1- 3	AM2- 1	AM2- 2	AM2- 3	AM3- 1	AM3- 2	AM3- 3	AM4- 1	AM4- 2
Analyte	Result	Result	Result	Result	Result	Result	Result	Result	Result	Result
C16:1 Hexadecenoic (Palmitoleic)	0.11	0.24	0.75	0.19	0.19	0.12	0.06	0.05	0.04	0.12
C16:2 Hexadecadienoic	0.01	0.05	0.09	0.02	0.02	0.04	0.01	0	0	0.03
C16:3 Hexadecatrienoic	0	0	0	0	0	0	0	0	0	0
C16:4 Hexadecatetraenoic	0	0	0	0	0	0	0	0	0	0
C17:0 Heptadecanoic (Margaric)	0.02	0.06	0.12	0.02	0.03	0.06	0.02	0.01	0.01	0.04
C17:1 Heptadecenoic Margaroleic	0.02	0.05	0.13	0.03	0.04	0.03	0.01	0	0	0.03
C18:0 Octadecanoic (Stearic)	0.21	0.91	1.38	0.26	0.39	1.34	0.36	0.23	0.24	0.59
C18:1 Octadecenoic (Oleic)	0.82	2.74	5.52	1.26	1.61	1.9	0.78	0.53	0.5	1.35
C18:2 Octadecadienoic (Linoleic)	0.07	0.26	0.41	0.1	0.14	0.23	0.08	0.05	0.05	0.15
C18:3 Octadecatrienoic (Linolenic)	0	0.02	0.04	0.01	0.02	0.03	0	0	0	0.02
C18:4 Octadecatetraenoic	0.01	0.03	0.08	0.02	0.02	0.02	0	0	0	0.02
C19:0 Nonadecanoic	0	0	0	0	0	0	0	0	0	0
C20:0 Eicosanoic (Arachidic)	0	0	0	0	0	0	0	0	0	0
C20:1 Eicosenoic (Gadoleic)	0	0.02	0.04	0	0.01	0.01	0	0	0	0
C20:2 Eicosadienoic	0	0	0	0	0	0	0	0	0	0
C20:3 Eicosatrienoic	0	0	0	0	0	0	0	0	0	0
C20:4 Eicosatetraenoic (Arachidonic)	0	0	0.01	0.01	0.01	0.02	0	0	0	0.01
C20:5 Eicosapentaenoic	0	0	0	0	0	0	0	0	0	0
C21:0 Heneicosanoic	0	0	0	0	0	0	0	0	0	0
C21:5 Heneicosapentaenoic	0	0	0	0	0	0	0	0	0	0
C22:0 Docosanoic (Behenic)	0	0	0	0	0	0	0	0	0	0
C22:1 Docosenoic (Erucic)	0	0	0	0	0	0	0	0	0	0
C22:2 Docosadienoic	0	0	0	0	0	0	0	0	0	0
C22:3 Docosatrienoic	0	0	0	0	0	0	0	0	0	0
C22:4 Docosatetraenoic	0	0	0	0	0	0	0	0	0	0
C22:5 Docosapentaenoic	0	0	0	0	0	0	0	0	0	0
C22:6 Docosahexaenoic	0	0	0	0	0	0	0	0	0	0
C24:0 Tetracosanoic (Lignoceric)	0	0	0	0	0	0	0	0	0	0
C24:1 Tetracosenoic (Nervonic)	0	0	0	0	0	0	0	0	0	0
Total Monounsatur. Fatty Acids Calc.	0.97	3.08	6.57	1.53	1.87	2.07	0.87	0.59	0.55	1.51
Total Polyunsatur. Fatty Acids Calc.	0.1	0.38	0.64	0.17	0.23	0.36	0.13	0.07	0.07	0.24
Saturated Fatty Acids	0.74	2.56	4.91	1.03	1.31	2.77	0.84	0.58	0.6	1.45
Total Fat (as triglycerides)	1.9	6.31	12.7	2.86	3.58	5.46	1.93	1.3	1.29	3.35
Calcium	5.2	5.1	43	4	5.1	16	4.1	4.5	6.2	8.2
Iron	4	2.3	2.4	1.9	2.6	2.3	2.2	2	1.7	2.3

Cyagra Meat Study

SampleNo	36134	36135	36136	36137	36138	36139	36140	36142	36143	36144
Marked ID	AM1- 2	AM1- 3	AM2- 1	AM2- 2	AM2- 3	AM3- 1	AM3- 2	AM3- 3	AM4- 1	AM4- 2
Analyte	Result	Result	Result	Result	Result	Result	Result	Result	Result	Result
Phosphorus	230	170	190	190	200	180	180	210	190	180
Zinc	4.6	4.7	3.5	3.5	5.5	3.8	4.7	4.6	3.8	4.5
Cholesterol	60.2	61.3	65.6	62	65.1	58	57.5	52.3	56.6	62
Niacin	7.08	4.51	5.72	7.91	5.69	4.92	5.37	5.89	6.05	5
Vitamin B1 - Thiamine Hydrochloride	0.094	0.086	0.181	0.147	0.145	0.161	0.109	0.112	0.118	0.154
Vitamin B2 - Riboflavin	0.279	0.283	0.31	0.257	0.311	0.272	0.309	0.226	0.266	0.292
Vitamin B6	0.49	0.315	0.373	0.418	0.306	0.276	0.362	0.358	0.368	0.327
Vitamin B12	0.0029	0.00366	0.00359	0.00305	0.00365	0.00293	0.00319	0.00279	0.00283	0.00338
Vitamin E	0.51	0.377	0.319	0.376	0.35	0.498	0.525	0.392	0.507	0.61

Cyagra Meat Study

SampleNo	36145	36146	36148	36149		36150	36151	36152	36153	36154
Marked ID	AM4- 3	AM5- 1	AM5- 2	AM5- 3		BF1- 1	BF1- 2	BF1- 3	BF2- 1	BF2- 2
Analyte	Result	Result	Result	Result		Result	Result	Result	Result	Result
Crude Fat	5.3	8.2	7.2	5.1		4.1	5.3	23.2	21	19.9
Moisture	72.66	69.5	70.22	68.71		71.18	72.44	58.66	58.01	60.84
Protein - Combustion	22.53	21.99	21.18	25.98		22.82	20.05	17.13	19.54	19.11
Protein - Kjeldahl	22.26	21.81	21.23	25.14		22.71	19.92	17.01	19.49	18.93
Ash	1.24	1.06	1.06	1.27		1.08	0.94	0.85	0.95	0.86
Balance (protein+moist+ash+fat)	101.46	100.57	99.71	100.22		99.07	98.60	99.72	99.45	100.53
Amino Acid Profile (results below)										
Tryptophan	0.29	0.24	0.25	0.27		0.26	0.23	0.19	0.21	0.22
Aspartic Acid	2.09	1.97	2	2.38		2.15	1.88	1.55	1.8	1.78
Threonine	1	0.93	0.97	1.18		1.05	0.93	0.75	0.88	0.89
Serine	0.89	0.87	0.86	1.01		0.91	0.81	0.71	0.73	0.75
Glutamic Acid	3.48	3.27	3.34	3.92		3.54	3.15	2.55	2.92	2.96
Proline	1.34	1.39	0.98	1.18		1.03	0.73	0.73	0.66	0.74
Glycine	1.67	1.68	1.09	1.13		1.14	0.95	1.03	0.83	0.87
Alanine	1.52	1.48	1.31	1.5		1.42	1.22	1.08	1.13	1.14
Cystine	0.22	0.22	0.23	0.27		0.23	0.21	0.18	0.19	0.2
Valine	1.03	0.98	0.98	1.16		1.06	0.89	0.74	0.94	0.91
Methionine	0.53	0.52	0.53	0.66		0.56	0.48	0.42	0.47	0.47
Isoleucine	0.92	0.85	0.88	1.06		0.95	0.82	0.68	0.86	0.84
Leucine	1.73	1.61	1.67	2		1.82	1.6	1.31	1.56	1.54
Tyrosine	0.71	0.66	0.71	0.86		0.77	0.69	0.55	0.66	0.66
Phenylalanine	0.93	0.86	0.87	1.05		0.96	0.83	0.68	0.82	0.82
Histidine	0.73	0.71	0.72	0.88		0.83	0.69	0.57	0.71	0.69
Lysine, Total	1.91	1.79	1.89	2.27		2.05	1.79	1.46	1.74	1.73
Arginine	1.56	1.49	1.37	1.6		1.45	1.26	1.08	1.2	1.2
Hydroxyproline	0.13	0.19	0.16	0.15		0.2	0.11	0.12	0.12	0.27
Fatty Acid (results below)										
C08:0 Octanoic (Caprylic)	0	0	0	0		0	0	0	0	0
C10:0 Decanoic (Capric)	0	0	0	0		0	0	0.01	0	0
C11:0 Undecanoic (Hendecanoic)	0	0	0	0		0	0	0	0	0
C12:0 Dodecanoic (Lauric)	0	0	0	0		0	0	0.02	0.01	0.02
C14:0 Tetradecanoic (Myristic)	0.12	0.2	0.16	0.13		0.12	0.13	0.65	0.66	0.63
C14:1 Tetradecenoic (Myristoleic)	0.02	0.03	0.03	0.04		0.06	0.04	0.24	0.23	0.33
C15:0 Pentadecanoic	0.02	0.04	0.03	0.02		0.01	0.02	0.09	0.09	0.09
C15:1 Pentadecenoic	0	0	0	0		0	0	0	0	0
C16:0 Hexadecanoic (Palmitic)	1.15	1.93	1.69	1.22		0.94	1.17	5.39	5.39	5.16

Cyagra Meat Study

SampleNo	36145	36146	36148	36149		36150	36151	36152	36153	36154
Marked ID	AM4- 3	AM5- 1	AM5- 2	AM5- 3		BF1- 1	BF1- 2	BF1- 3	BF2- 1	BF2- 2
Analyte	Result	Result	Result	Result		Result	Result	Result	Result	Result
C16:1 Hexadecenoic (Palmitoleic)	0.16	0.23	0.2	0.21		0.29	0.25	1.09	0.93	1.26
C16:2 Hexadecadienoic	0.04	0.06	0.05	0.03		0.02	0.03	0.15	0.16	0.15
C16:3 Hexadecatrienoic	0	0	0	0		0	0	0	0	0
C16:4 Hexadecatetraenoic	0	0	0	0		0	0	0	0.01	0
C17:0 Heptadecanoic (Margaric)	0.05	0.11	0.09	0.05		0.03	0.05	0.23	0.23	0.2
C17:1 Heptadecenoic Margaroleic	0.03	0.06	0.05	0.05		0.04	0.05	0.22	0.19	0.19
C18:0 Octadecanoic (Stearic)	0.98	1.55	1.37	0.69		0.33	0.59	2.65	2.29	1.99
C18:1 Octadecenoic (Oleic)	1.83	3.05	2.79	1.99		1.79	2.33	10	8.09	7.81
C18:2 Octadecadienoic (Linoleic)	0.19	0.26	0.29	0.13		0.09	0.14	0.59	0.37	0.34
C18:3 Octadecatrienoic (Linolenic)	0.02	0.02	0.03	0.01		0.02	0.02	0.14	0.12	0.11
C18:4 Octadecatetraenoic	0.03	0.03	0.03	0.02		0.02	0.02	0.14	0.07	0.08
C19:0 Nonadecanoic	0	0	0	0		0	0	0	0	0
C20:0 Eicosanoic (Arachidic)	0	0	0	0		0	0	0.01	0.05	0
C20:1 Eicosenoic (Gadoleic)	0.01	0.02	0.02	0.01		0.01	0.01	0.06	0.01	0.04
C20:2 Eicosadienoic	0	0	0	0		0	0	0	0	0
C20:3 Eicosatrienoic	0	0	0	0		0	0	0.02	0	0
C20:4 Eicosatetraenoic (Arachidonic)	0.01	0.01	0.01	0		0.01	0.01	0.02	0.02	0.01
C20:5 Eicosapentaenoic	0	0	0	0		0	0	0	0	0
C21:0 Heneicosanoic	0	0	0	0		0	0	0	0	0
C21:5 Heneicosapentaenoic	0	0	0	0		0	0	0	0	0
C22:0 Docosanoic (Behenic)	0	0	0	0		0	0	0	0	0
C22:1 Docosenoic (Erucic)	0	0	0	0		0	0	0	0	0
C22:2 Docosadienoic	0	0	0	0		0	0	0	0	0
C22:3 Docosatrienoic	0	0	0	0		0	0	0	0	0
C22:4 Docosatetraenoic	0	0	0	0		0	0	0	0	0
C22:5 Docosapentaenoic	0	0	0	0		0	0	0.01	0	0
C22:6 Docosahexaenoic	0	0	0	0		0	0	0	0	0
C24:0 Tetracosanoic (Lignoceric)	0	0	0	0		0	0	0	0	0
C24:1 Tetracosenoic (Nervonic)	0	0	0	0		0	0	0	0	0
Total Monounsatur. Fatty Acids Calc.	2.05	3.4	3.09	2.29		2.2	2.69	11.6	9.46	9.63
Total Polyunsatur. Fatty Acids Calc.	0.31	0.41	0.44	0.2		0.18	0.25	1.08	0.76	0.7
Saturated Fatty Acids	2.33	3.84	3.36	2.12		1.44	1.97	9.06	8.72	8.1
Total Fat (as triglycerides)	4.93	8.02	7.22	4.83		4	5.15	22.8	19.9	19.4
Calcium	53	23	6.7	5		6.7	6.1	35	54	5.6
Iron	2.3	1.4	1.3	1.1		2.4	2.7	2.4	2.7	2.5

Cyagra Meat Study

SampleNo	36145	36146	36148	36149		36150	36151	36152	36153	36154
Marked ID	AM4- 3	AM5- 1	AM5- 2	AM5- 3		BF1- 1	BF1- 2	BF1- 3	BF2- 1	BF2- 2
Analyte	Result	Result	Result	Result		Result	Result	Result	Result	Result
Phosphorus	220	190	180	220		190	170	150	180	160
Zinc	3.8	3.1	3.9	2.8		3.8	4.9	3.4	3.6	4.6
Cholesterol	60.1	65.3	63	70.4		57.7	56.2	73.3	71.1	84.2
Niacin	4.97	5.93	5.18	7.34		5.47	3.95	3.05	4.34	3.59
Vitamin B1 - Thiamine Hydrochloride	0.147	0.114	0.111	0.119		0.108	0.101	0.09	0.12	0.079
Vitamin B2 - Riboflavin	0.308	0.212	0.205	0.172		0.231	0.257	0.224	0.284	0.222
Vitamin B6	0.417	0.361	0.334	0.492		0.344	0.274	0.233	0.301	0.209
Vitamin B12	0.00392	0.00203	0.00198	0.00174		0.00229	0.00403	0.00273	0.00378	0.00342
Vitamin E	0.708	0.353	0.532	0.249		0.389	0.474	0.434	0.331	0.479

Cyagra Meat Study

SampleNo	36155	36156	36157	36158	36170	36171	36172	36173	36174	36175
Marked ID	BF2- 3	BF3- 1	BF3- 2	BF3- 3	BF4-1	BF4- 2	BF4- 3	BF5- 1	BF5- 2	BF5- 3
Analyte	Result	Result	Result	Result	Result	Result	Result	Result	Result	Result
Crude Fat	5.8	10.7	33.7	15.9	10.5	8.8	6.5	4.4	6.2	11.4
Moisture	68.09	64.54	52.56	63.01	66.77	69.1	69.44	69.29	71.24	66.64
Protein - Combustion	24.64	21.94	15.03	20.27	21.39	21.99	23.33	23.97	22.12	20.59
Protein - Kjeldahl	24.49	22.11	15.25	20.08	21.25	21.55	22.56	23.58	21.63	20.76
Ash	1.16	1.04	0.74	0.92	1.01	0.97	0.95	1.12	0.91	1.06
Balance (protein+moist+ash+fat)	99.54	98.39	102.25	99.91	99.53	100.42	99.45	98.39	99.98	99.86
Amino Acid Profile (results below)										
Tryptophan	0.28	0.27	0.2	0.24	0.26	0.27	0.28	0.29	0.25	0.25
Aspartic Acid	2.34	2.12	1.53	1.87	2.04	2.09	2.18	2.42	2.11	2.01
Threonine	1.16	1.03	0.73	0.89	1.03	1.03	1.08	1.2	1.02	0.98
Serine	1	0.83	0.57	0.69	0.86	0.91	0.94	1.05	0.87	0.86
Glutamic Acid	3.8	3.47	2.49	3.09	3.31	3.47	3.55	3.93	3.5	3.28
Proline	0.94	0.98	0.66	0.75	0.77	0.83	0.91	0.95	0.96	0.84
Glycine	1.31	1.11	0.72	0.8	0.91	1.03	1.05	1.12	1.1	1
Alanine	1.61	1.39	0.99	1.17	1.28	1.33	1.4	1.55	1.38	1.32
Cystine	0.24	0.24	0.18	0.23	0.23	0.23	0.23	0.24	0.22	0.21
Valine	1.25	1.1	0.79	0.98	1	0.99	1.05	1.18	1.09	1.05
Methionine	0.6	0.56	0.42	0.55	0.56	0.57	0.56	0.61	0.53	0.5
Isoleucine	1.1	1.02	0.73	0.93	0.93	0.93	0.97	1.08	0.99	0.94
Leucine	2.03	1.79	1.31	1.59	1.76	1.77	1.85	2.07	1.81	1.75
Tyrosine	0.85	0.76	0.54	0.67	0.76	0.76	0.78	0.86	0.75	0.73
Phenylalanine	1.07	0.93	0.7	0.83	0.91	0.91	0.94	1.04	0.93	0.89
Histidine	0.95	0.82	0.57	0.74	0.78	0.73	0.82	0.95	0.81	0.77
Lysine, Total	2.26	1.99	1.43	1.79	1.98	2	2.09	2.35	2.06	1.96
Arginine	1.57	1.44	1.01	1.24	1.34	1.37	1.4	1.56	1.43	1.34
Hydroxyproline	0.14	0.16	0.13	0.11	0.23	0.16	0.1	0.42	0.13	0.21
Fatty Acid (results below)										
C08:0 Octanoic (Caprylic)	0	0	0	0	0	0	0	0	0	0
C10:0 Decanoic (Capric)	0	0	0.01	0	0	0	0	0	0	0
C11:0 Undecanoic (Hendecanoic)	0	0	0	0	0	0	0	0	0	0
C12:0 Dodecanoic (Lauric)	0	0	0.02	0.01	0	0.01	0	0	0	0
C14:0 Tetradecanoic (Myristic)	0.17	0.33	1.04	0.46	0.29	0.23	0.17	0.12	0.15	0.32
C14:1 Tetradecenoic (Myristoleic)	0.13	0.24	0.31	0.28	0.13	0.11	0.14	0.09	0.04	0.11
C15:0 Pentadecanoic	0.02	0.03	0.13	0.05	0.05	0.05	0.03	0.02	0.03	0.06
C15:1 Pentadecenoic	0	0	0	0	0	0	0	0	0	0
C16:0 Hexadecanoic (Palmitic)	1.42	2.5	8.32	3.81	2.69	2.21	1.65	1.05	1.51	2.88

Cyagra Meat Study

SampleNo	36155	36156	36157	36158	36170	36171	36172	36173	36174	36175
Marked ID	BF2- 3	BF3- 1	BF3- 2	BF3- 3	BF4-1	BF4- 2	BF4- 3	BF5- 1	BF5- 2	BF5- 3
Analyte	Result	Result	Result	Result	Result	Result	Result	Result	Result	Result
C16:1 Hexadecenoic (Palmitoleic)	0.52	1.09	1.64	1.34	0.58	0.49	0.56	0.49	0.31	0.64
C16:2 Hexadecadienoic	0.03	0.06	0.24	0.09	0.09	0.07	0.05	0.03	0.05	0.1
C16:3 Hexadecatrienoic	0	0	0	0	0	0	0	0	0	0
C16:4 Hexadecatetraenoic	0	0	0	0	0.01	0	0	0	0	0
C17:0 Heptadecanoic (Margaric)	0.03	0.06	0.35	0.11	0.11	0.09	0.04	0.03	0.07	0.13
C17:1 Heptadecenoic Margaroleic	0.05	0.12	0.31	0.16	0.11	0.09	0.07	0.05	0.06	0.12
C18:0 Octadecanoic (Stearic)	0.34	0.58	4.26	1.21	1	0.84	0.38	0.24	0.72	1.23
C18:1 Octadecenoic (Oleic)	2.41	4.71	14.2	6.99	4.13	3.34	2.69	1.79	2.44	4.48
C18:2 Octadecadienoic (Linoleic)	0.09	0.19	0.7	0.28	0.23	0.19	0.13	0.09	0.16	0.3
C18:3 Octadecatrienoic (Linolenic)	0.02	0.04	0.2	0.06	0.06	0.06	0.03	0.02	0.05	0.08
C18:4 Octadecatetraenoic	0.02	0.06	0.14	0.06	0.05	0.04	0.03	0.02	0.02	0.06
C19:0 Nonadecanoic	0	0	0	0	0	0	0	0	0	0
C20:0 Eicosanoic (Arachidic)	0.01	0	0.03	0	0	0	0	0	0	0
C20:1 Eicosenoic (Gadoleic)	0	0.04	0.13	0.05	0.03	0.03	0.02	0.01	0.02	0.03
C20:2 Eicosadienoic	0	0	0	0	0	0	0	0	0	0
C20:3 Eicosatrienoic	0	0	0	0	0	0	0	0	0	0
C20:4 Eicosatetraenoic (Arachidonic)	0	0	0	0	0.02	0.01	0.01	0	0.01	0.02
C20:5 Eicosapentaenoic	0	0	0	0	0	0	0	0	0	0
C21:0 Heneicosanoic	0	0	0	0	0	0	0	0	0	0
C21:5 Heneicosapentaenoic	0	0	0	0	0	0	0	0	0	0
C22:0 Docosanoic (Behenic)	0	0	0	0	0	0	0	0	0	0
C22:1 Docosenoic (Erucic)	0	0	0	0	0	0	0	0	0	0
C22:2 Docosadienoic	0	0	0	0	0	0	0	0	0	0
C22:3 Docosatrienoic	0	0	0	0	0	0	0	0	0	0
C22:4 Docosatetraenoic	0	0	0	0	0	0	0	0	0	0
C22:5 Docosapentaenoic	0	0	0	0	0	0.01	0	0	0	0
C22:6 Docosahexaenoic	0	0	0	0	0	0	0	0	0	0
C24:0 Tetracosanoic (Lignoceric)	0	0	0	0	0	0	0	0	0	0
C24:1 Tetracosenoic (Nervonic)	0	0	0	0	0	0	0	0	0	0
Total Monounsatur. Fatty Acids Calc.	3.11	6.2	16.6	8.82	4.99	4.04	3.49	2.44	2.87	5.38
Total Polyunsatur. Fatty Acids Calc.	0.18	0.36	1.28	0.51	0.48	0.4	0.28	0.18	0.31	0.58
Saturated Fatty Acids	1.99	3.51	14.2	5.66	4.16	3.44	2.28	1.46	2.51	4.64
Total Fat (as triglycerides)	5.53	10.6	33.6	15.7	10.1	8.28	6.35	4.28	5.97	11.1
Calcium	7	5.4	10	4.9	41	7.4	4.1	4.8	5.7	29
Iron	3.3	2.4	2.4	3	3.3	3	2.9	3.7	4.1	3.6

Cyagra Meat Study

SampleNo	36155	36156	36157	36158	36170	36171	36172	36173	36174	36175
Marked ID	BF2- 3	BF3- 1	BF3- 2	BF3- 3	BF4-1	BF4- 2	BF4- 3	BF5- 1	BF5- 2	BF5- 3
Analyte	Result	Result	Result	Result	Result	Result	Result	Result	Result	Result
Phosphorus	210	180	130	150	190	180	190	220	180	250
Zinc	4.9	4	3.4	4.5	4.6	5.7	4.4	4.3	4.7	4.3
Cholesterol	66.7	62	74.5	62	63.5	70	61.7	61.6	57.6	66.8
Niacin	7.12	5.92	3.55	3.8	5.16	4.98	6.58	5.96	4.32	4.53
Vitamin B1 - Thiamine Hydrochloride	0.121	0.093	0.104	0.1	0.127	0.083	0.121	0.097	0.102	0.11
Vitamin B2 - Riboflavin	0.277	0.239	0.238	0.26	0.352	0.294	0.304	0.323	0.304	0.331
Vitamin B6	0.497	0.419	0.167	0.242	0.412	0.289	0.433	0.582	0.51	0.371
Vitamin B12	0.00365	0.00321	0.00407	0.00347	0.0046	0.00416	0.00369	0.00335	0.00422	0.00399
Vitamin E	0.369	0.482	0.86	0.758	0.608	0.44	0.509	0.28	0.308	0.405

Cyagra Meat Study

SampleNo	36176	36178	36179	36183	36198	36199	36200	36201	36202	36203
Marked ID	BF6- 1	BF6- 2	BF6- 3	BM1- 1	BM1- 2	BM1- 3	BM2 - 1	BM2 - 2	BM2 - 3	BM3 - 1
Analyte	Result	Result	Result	Result	Result	Result	Result	Result	Result	Result
Crude Fat	7.8	22.5	4	6.8	1.4	2.4	0.6	1.1	4.4	3.5
Moisture	70.82	60.58	72.53	70.02	70.42	74.07	73.69	74.35	71.77	74.17
Protein - Combustion	21.14	18.04	22.64	21.48	27.66	22.08	23.83	21.72	22.32	21.25
Protein - Kjeldahl	21.11	17.87	22.35	21.52	26.07	21.71	23.48	22.68	22.39	21.21
Ash	1.05	0.93	1.11	1.16	1.24	1.1	1.16	1.11	1.09	1.11
Balance (protein+moist+ash+fat)	100.78	101.88	99.99	99.50	99.13	99.28	98.93	99.24	99.65	99.99
Amino Acid Profile (results below)										
Tryptophan	0.26	0.21	0.28	0.25	0.31	0.26	0.27	0.25	0.26	0.26
Aspartic Acid	2.11	1.73	2.26	2.05	2.47	2.13	2.17	2.16	2.12	2.07
Threonine	0.99	0.81	1.04	1.03	1.24	1.08	1.1	1.07	1.07	1.02
Serine	0.76	0.7	0.74	0.89	1.09	0.95	0.92	0.91	0.87	0.85
Glutamic Acid	3.51	2.86	3.73	<0.0	4.09	0.01	3.54	3.62	3.47	3.53
Proline	0.89	0.75	0.91	0.94	1.33	0.97	1	0.88	0.89	0.9
Glycine	0.97	0.83	1.03	1.06	1.81	1.12	1.11	1.04	0.96	1.04
Alanine	1.34	1.1	1.46	1.4	1.86	1.4	1.4	1.39	1.34	1.36
Cystine	0.25	0.2	0.25	0.23	0.29	0.28	0.29	0.29	0.27	0.29
Valine	1.11	0.87	1.24	1.05	1.36	1.07	1.09	1.13	1.11	1.07
Methionine	0.57	0.44	0.59	0.53	0.68	0.57	0.61	0.61	0.61	0.58
Isoleucine	1.05	0.82	1.16	0.97	1.17	0.99	1.01	1.04	1.04	1
Leucine	1.78	1.47	1.92	1.79	2.15	1.84	1.87	1.87	1.85	1.79
Tyrosine	0.75	0.6	0.78	0.75	0.9	0.77	0.78	0.78	0.77	0.76
Phenylalanine	0.92	0.75	1.01	0.91	1.11	0.9	0.92	0.93	0.91	0.89
Histidine	0.8	0.64	0.91	0.74	0.93	0.8	0.83	0.84	0.8	0.57
Lysine, Total	2	1.61	2.15	1.96	2.32	2.04	2.08	2.12	2.04	1.99
Arginine	1.42	1.15	1.49	1.42	1.8	1.5	1.5	1.48	1.39	1.4
Hydroxyproline	0.17	0.17	0.04	0.15	0.12	0.18	0.15	0.11	0.09	0.17
Fatty Acid (results below)										
C08:0 Octanoic (Caprylic)	0	0	0	0	0	0	0	0	0	0
C10:0 Decanoic (Capric)	0	0	0	0	0	0	0	0	0	0
C11:0 Undecanoic (Hendecanoic)	0	0	0	0	0	0	0	0	0	0
C12:0 Dodecanoic (Lauric)	0	0.01	0	0	0	0	0	0	0	0
C14:0 Tetradecanoic (Myristic)	0.19	0.59	0.11	0.13	0.03	0.04	0.01	0.02	0.08	0.07
C14:1 Tetradecenoic (Myristoleic)	0.09	0.2	0.06	0.02	0.01	0	0	0	0	0.01
C15:0 Pentadecanoic	0.03	0.13	0.02	0.03	0	0	0	0	0.02	0.02
C15:1 Pentadecenoic	0	0	0	0	0	0	0	0	0	0
C16:0 Hexadecanoic (Palmitic)	1.77	5.22	0.91	1.28	0.28	0.41	0.11	0.24	0.84	0.73

Cyagra Meat Study

SampleNo	36176	36178	36179	36183	36198	36199	36200	36201	36202	36203
Marked ID	BF6- 1	BF6- 2	BF6- 3	BM1- 1	BM1- 2	BM1- 3	BM2 - 1	BM2 - 2	BM2 - 3	BM3 - 1
Analyte	Result	Result	Result	Result	Result	Result	Result	Result	Result	Result
C16:1 Hexadecenoic (Palmitoleic)	0.42	0.9	0.27	0.15	0.06	0.06	0.02	0.03	0.1	0.1
C16:2 Hexadecadienoic	0.06	0.22	0.03	0.05	0	0.01	0	0	0.03	0.02
C16:3 Hexadecatrienoic	0	0	0	0	0	0	0	0	0	0
C16:4 Hexadecatetraenoic	0	0	0	0	0	0	0	0	0	0.01
C17:0 Heptadecanoic (Margaric)	0.08	0.3	0.03	0.07	0	0.02	0	0.01	0.05	0.04
C17:1 Heptadecenoic Margaroleic	0.08	0.22	0.04	0.03	0	0.01	0	0	0.02	0.02
C18:0 Octadecanoic (Stearic)	0.85	3.2	0.34	1.51	0.17	0.39	0.08	0.23	0.9	0.69
C18:1 Octadecenoic (Oleic)	3.28	8.9	1.65	2.34	0.54	0.84	0.18	0.41	1.47	1.23
C18:2 Octadecadienoic (Linoleic)	0.24	0.65	0.11	0.3	0.05	0.12	0.03	0.07	0.21	0.19
C18:3 Octadecatrienoic (Linolenic)	0.06	0.18	0.03	0.04	0	0	0	0	0.02	0.02
C18:4 Octadecatetraenoic	0.03	0.09	0.02	0.02	0	0	0	0	0.02	0.01
C19:0 Nonadecanoic	0	0	0	0	0	0	0	0	0	0
C20:0 Eicosanoic (Arachidic)	0	0.03	0	0.01	0	0	0	0	0	0
C20:1 Eicosenoic (Gadoleic)	0.03	0.07	0.01	0.02	0	0	0	0	0	0
C20:2 Eicosadienoic	0	0	0	0	0	0	0	0	0	0
C20:3 Eicosatrienoic	0	0.02	0	0	0	0	0	0	0	0
C20:4 Eicosatetraenoic (Arachidonic)	0.01	0.02	0	0.02	0	0.01	0	0.01	0.02	0.02
C20:5 Eicosapentaenoic	0	0	0	0	0	0	0	0	0	0
C21:0 Heneicosanoic	0	0	0	0	0	0	0	0	0	0
C21:5 Heneicosapentaenoic	0	0	0	0	0	0	0	0	0	0
C22:0 Docosanoic (Behenic)	0	0	0	0	0	0	0	0	0	0
C22:1 Docosenoic (Erucic)	0	0	0	0	0	0	0	0	0	0
C22:2 Docosadienoic	0	0	0	0	0	0	0	0	0	0
C22:3 Docosatrienoic	0	0	0	0	0	0	0	0	0	0
C22:4 Docosatetraenoic	0	0	0	0	0	0	0	0	0	0
C22:5 Docosapentaenoic	0	0.01	0	0	0	0	0	0	0	0
C22:6 Docosahexaenoic	0	0	0	0	0	0	0	0	0	0
C24:0 Tetracosanoic (Lignoceric)	0	0	0	0	0	0	0	0	0	0
C24:1 Tetracosenoic (Nervonic)	0	0	0	0	0	0	0	0	0	0
Total Monounsatur. Fatty Acids Calc.	3.89	10.3	2.04	2.56	0.62	0.93	0.21	0.45	1.61	1.37
Total Polyunsatur. Fatty Acids Calc.	0.42	1.22	0.2	0.44	0.08	0.18	0.04	0.1	0.31	0.29
Saturated Fatty Acids	2.94	9.48	1.4	3.03	0.5	0.87	0.21	0.52	1.89	1.55
Total Fat (as triglycerides)	7.6	22	3.82	6.33	1.26	2.07	0.48	1.12	4	3.37
Calcium	5.4	22	4.2	39	6.5	6.4	5.4	6.7	20	6.8
Iron	2.6	2.4	2.2	1.7	1.5	1.6	1.6	1.6	1.8	1.7

Cyagra Meat Study

SampleNo	36176	36178	36179	36183	36198	36199	36200	36201	36202	36203
Marked ID	BF6- 1	BF6- 2	BF6- 3	BM1- 1	BM1- 2	BM1- 3	BM2 - 1	BM2 - 2	BM2 - 3	BM3 - 1
Analyte	Result	Result	Result	Result	Result	Result	Result	Result	Result	Result
Phosphorus	170	160	190	200	240	200	200	210	200	190
Zinc	5	3.8	4.2	3.7	4.3	4.7	3.4	4	3	5.3
Cholesterol	57.6	67.9	54.4	71.2	101	73.3	72.8	68.2	69.9	73.4
Niacin	4.56	3.3	4.94	4.4	5.74	4.57	6.38	6.22	6.53	4.1
Vitamin B1 - Thiamine Hydrochloride	0.096	0.078	0.087	0.099	0.093	0.102	0.079	0.072	0.059	0.106
Vitamin B2 - Riboflavin	0.264	0.284	0.231	0.322	0.279	0.323	0.298	0.271	0.335	0.284
Vitamin B6	0.365	0.318	0.556	0.358	0.492	0.5	0.485	0.398	0.509	0.289
Vitamin B12	0.00394	0.0043	0.00385	0.00405	0.00384	0.00411	0.00374	0.00401	0.00443	0.00419
Vitamin E	0.487	0.377	0.724	0.427	0.542	0.431	0.375	0.403	0.387	0.331

Cyagra Meat Study

SampleNo	36204	36206	36207	36208	36209	36210	36211	36213		
Marked ID	BM3 - 2	BM3 - 3	BM4- 1	BM4- 2	BM4- 3	BM5- 1	BM5- 2	BM5- 3		Clone
Analyte	Result	Result	Result	Result	Result	Result	Result	Result		Mean
Crude Fat	4	1.2	5.9	2.9	2.1	22.1	2.3	2.1		11.62
Moisture	72.37	71.42	70.79	72.59	74.99	61.82	73.9	70.71		66.18
Protein - Combustion	21.89	24.91	22.5	23.21	23.31	16.36	22.56	25.89		20.69
Protein - Kjeldahl	21.41	25.39	22.5	23.76	22.99	16.19	21.92	26.39		20.74
Ash	1.18	1.32	1.16	1.16	1.09	0.84	1.02	1.23		1.03
Balance (protein+moist+ash+fat)	98.96	99.33	100.35	100.41	101.17	100.95	99.14	100.43		99.56
Amino Acid Profile (results below)										
Tryptophan	0.27	0.31	0.27	0.28	0.26	0.2	0.27	0.31		0.25
Aspartic Acid	2.11	2.37	2.17	2.27	2.34	1.72	2.12	2.39		1.96
Threonine	1.03	1.19	1.08	1	1.08	0.8	0.98	1.12		0.93
Serine	0.9	1.02	0.9	0.7	0.93	0.71	0.9	1		0.79
Glutamic Acid	3.61	4.05	3.56	3.77	3.64	2.96	3.53	4.02		3.22
Proline	0.98	1.23	0.97	1.04	1.07	0.69	1	1.23		0.97
Glycine	1.17	1.41	1.08	1.22	1.14	0.91	1.13	1.59		1.08
Alanine	1.43	1.61	1.39	1.51	1.4	1.11	1.39	1.63		1.28
Cystine	0.27	0.32	0.23	0.36	0.2	0.2	0.27	0.31		0.23
Valine	1.08	1.25	1.12	1.26	1.13	0.89	1.08	1.26		0.89
Methionine	0.59	0.72	0.57	0.73	0.46	0.42	0.56	0.67		0.54
Isoleucine	1	1.13	1.02	1.15	1.04	0.83	1	1.14		0.81
Leucine	1.84	2.06	1.88	1.91	1.88	1.48	1.79	2.01		1.61
Tyrosine	0.76	0.84	0.78	0.78	0.79	0.61	0.76	0.83		0.69
Phenylalanine	0.91	1.01	0.94	1	0.94	0.77	0.93	1.04		0.84
Histidine	0.58	0.63	0.84	0.85	0.79	0.61	0.78	0.93		0.70
Lysine, Total	2.03	2.31	2.1	2.09	2.09	1.64	1.98	2.24		1.77
Arginine	1.46	1.61	1.44	1.54	1.55	1.17	1.43	1.73		1.33
Hydroxyproline	0.09	0.3	0.11	0.16	0.18	0.08	0.27	0.2		0.17
Fatty Acid (results below)										
C08:0 Octanoic (Caprylic)	0	0	0	0	0	0	0	0		0.00
C10:0 Decanoic (Capric)	0	0	0	0	0	0	0	0		0.00
C11:0 Undecanoic (Hendecanoic)	0	0	0	0	0	0	0	0		0.00
C12:0 Dodecanoic (Lauric)	0	0	0	0	0	0.01	0	0		0.00
C14:0 Tetradecanoic (Myristic)	0.07	0.03	0.13	0.07	0.04	0.49	0.04	0.06		0.28
C14:1 Tetradecenoic (Myristoleic)	0.01	0	0.03	0.03	0	0.08	0.01	0.02		0.15
C15:0 Pentadecanoic	0.02	0	0.02	0	0	0.11	0	0		0.04
C15:1 Pentadecenoic	0	0	0	0	0	0	0	0		0.00
C16:0 Hexadecanoic (Palmitic)	0.79	0.26	1.29	0.62	0.43	5.14	0.47	0.5		2.65

Cyagra Meat Study

SampleNo	36204	36206	36207	36208	36209	36210	36211	36213		
Marked ID	BM3 - 2	BM3 - 3	BM4- 1	BM4- 2	BM4- 3	BM5- 1	BM5- 2	BM5- 3		Clone
Analyte	Result	Result	Result	Result	Result	Result	Result	Result		Mean
C16:1 Hexadecenoic (Palmitoleic)	0.1	0.05	0.18	0.13	0.06	0.57	0.08	0.1		0.69
C16:2 Hexadecadienoic	0.03	0	0.04	0.02	0.01	0.17	0.02	0.01		0.08
C16:3 Hexadecatrienoic	0	0	0	0	0	0	0	0		0.00
C16:4 Hexadecatetraenoic	0	0	0	0	0	0	0	0		0.00
C17:0 Heptadecanoic (Margaric)	0.04	0.01	0.06	0.02	0.02	0.28	0.02	0.02		0.10
C17:1 Heptadecenoic Margaroleic	0.02	0	0.04	0.02	0.01	0.14	0.02	0.02		0.11
C18:0 Octadecanoic (Stearic)	0.77	0.17	1.07	0.35	0.35	4.32	0.35	0.28		1.18
C18:1 Octadecenoic (Oleic)	1.28	0.43	2.12	1.09	0.75	8	0.81	0.79		4.94
C18:2 Octadecadienoic (Linoleic)	0.18	0.05	0.26	0.11	0.12	0.62	0.11	0.08		0.31
C18:3 Octadecatrienoic (Linolenic)	0.02	0	0.02	0	0	0.1	0	0		0.05
C18:4 Octadecatetraenoic	0.01	0	0.02	0.01	0	0.12	0	0		0.07
C19:0 Nonadecanoic	0	0	0	0	0	0	0	0		0.00
C20:0 Eicosanoic (Arachidic)	0	0	0	0	0	0.03	0	0		0.00
C20:1 Eicosenoic (Gadoleic)	0	0	0.01	0	0	0.06	0	0		0.03
C20:2 Eicosadienoic	0	0	0	0	0	0.01	0	0		0.00
C20:3 Eicosatrienoic	0	0	0	0	0	0.01	0	0		0.00
C20:4 Eicosatetraenoic (Arachidonic)	0.02	0	0.02	0.01	0.02	0.03	0.02	0.01		0.01
C20:5 Eicosapentaenoic	0	0	0	0	0	0	0	0		0.00
C21:0 Heneicosanoic	0	0	0	0	0	0	0	0		0.00
C21:5 Heneicosapentaenoic	0	0	0	0	0	0	0	0		0.00
C22:0 Docosanoic (Behenic)	0	0	0	0	0	0	0	0		0.00
C22:1 Docosenoic (Erucic)	0	0	0	0	0	0	0	0		0.00
C22:2 Docosadienoic	0	0	0	0	0	0	0	0		0.00
C22:3 Docosatrienoic	0	0	0	0	0	0	0	0		0.00
C22:4 Docosatetraenoic	0	0	0	0	0	0	0	0		0.00
C22:5 Docosapentaenoic	0	0	0	0	0	0	0	0		0.00
C22:6 Docosahexaenoic	0	0	0	0	0	0	0	0		0.00
C24:0 Tetracosanoic (Lignoceric)	0	0	0	0	0	0	0	0		0.00
C24:1 Tetracosenoic (Nervonic)	0	0	0	0	0	0	0	0		0.00
Total Monounsatur. Fatty Acids Calc.	1.42	0.5	2.38	1.27	0.84	8.86	0.91	0.92		5.92
Total Polyunsatur. Fatty Acids Calc.	0.28	0.08	0.38	0.18	0.19	1.06	0.17	0.13		0.54
Saturated Fatty Acids	1.7	0.47	2.59	1.07	0.84	10.4	0.9	0.86		4.25
Total Fat (as triglycerides)	3.57	1.1	5.61	2.64	1.97	21.3	2.07	2		11.24
Calcium	22	9.3	43	5.5	5.9	24	5.1	8.1		12.01
Iron	1.6	1.5	1.8	1.6	1.7	1.9	1.8	1.7		2.29

Cyagra Meat Study

SampleNo	36204	36206	36207	36208	36209	36210	36211	36213		
Marked ID	BM3 - 2	BM3 - 3	BM4- 1	BM4- 2	BM4- 3	BM5- 1	BM5- 2	BM5- 3		Clone
Analyte	Result	Result	Result	Result	Result	Result	Result	Result		Mean
Phosphorus	210	250	220	210	180	150	180	220		179.09
Zinc	3.6	3.5	3.2	3.8	4.6	3.3	4.3	3.9		3.86
Cholesterol	70.3	71.5	73.9	66.9	66	69.4	67	74.6		64.92
Niacin	5.26	6.33	5.45	5.68	4.66	3.9	4.29	6.47		4.96
Vitamin B1 - Thiamine Hydrochloride	0.132	0.118	0.08	0.064	0.066	0.084	0.077	0.064		0.10
Vitamin B2 - Riboflavin	0.302	0.275	0.327	0.328	0.351	0.291	0.274	0.255		0.24
Vitamin B6	0.266	0.351	0.372	0.431	0.367	0.184	0.367	0.447		0.33
Vitamin B12	0.00434	0.0029	0.00326	0.00392	0.00241	0.00188	0.0041	0.00292		0.00
Vitamin E	0.259	0.303	0.485	0.247	0.328	0.726	0.415	0.302		0.50

Cyagra Meat Study

SampleNo				Clone		Comparator		Clone
Marked ID		Comparator		Females		Females		Males
Analyte	Std dev	Mean	Std dev	Mean	Std Dev	Mean	Std Dev	Mean
Crude Fat	10.08	8.62	8.10	17.10	10.62	12.32	8.50	5.04
Moisture	7.68	68.57	5.51	62.18	8.32	65.87	5.76	70.96
Protein - Combustion	2.96	21.72	2.58	18.96	2.62	20.87	2.46	22.77
Protein - Kjeldahl	2.90	21.58	2.51	19.06	2.60	20.70	2.35	22.76
Ash	0.17	1.05	0.13	0.95	0.18	0.98	0.11	1.12
Balance (protein+moist+ash+fat)	1.72	99.82	0.89	99.29	1.99	99.87	1.04	99.89
Amino Acid Profile (results below)								
Tryptophan	0.03	0.26	0.03	0.23	0.03	0.25	0.03	0.27
Aspartic Acid	0.31	2.08	0.23	1.76	0.25	2.00	0.25	2.19
Threonine	0.15	1.01	0.12	0.84	0.14	0.97	0.13	1.04
Serine	0.14	0.86	0.12	0.72	0.13	0.82	0.12	0.88
Glutamic Acid	0.54	3.33	0.71	2.88	0.44	3.28	0.41	3.63
Proline	0.21	0.91	0.16	0.83	0.13	0.84	0.11	1.13
Glycine	0.27	1.08	0.21	0.92	0.17	0.99	0.15	1.28
Alanine	0.21	1.36	0.18	1.14	0.17	1.30	0.17	1.45
Cystine	0.04	0.24	0.04	0.21	0.03	0.22	0.02	0.25
Valine	0.21	1.07	0.14	0.75	0.16	1.01	0.14	1.06
Methionine	0.09	0.56	0.08	0.49	0.07	0.53	0.06	0.59
Isoleucine	0.20	0.98	0.12	0.69	0.14	0.93	0.13	0.96
Leucine	0.27	1.78	0.20	1.44	0.22	1.71	0.22	1.82
Tyrosine	0.11	0.74	0.08	0.62	0.09	0.72	0.09	0.77
Phenylalanine	0.14	0.91	0.10	0.75	0.11	0.89	0.11	0.95
Histidine	0.12	0.77	0.11	0.63	0.09	0.77	0.11	0.79
Lysine, Total	0.31	1.98	0.23	1.58	0.24	1.91	0.25	2.01
Arginine	0.23	1.41	0.17	1.17	0.18	1.33	0.16	1.52
Hydroxyproline	0.07	0.16	0.07	0.16	0.08	0.17	0.08	0.18
Fatty Acid (results below)								
C08:0 Octanoic (Caprylic)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C10:0 Decanoic (Capric)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C11:0 Undecanoic (Hendecanoic)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C12:0 Dodecanoic (Lauric)	0.01	0.00	0.01	0.01	0.01	0.01	0.01	0.00
C14:0 Tetradecanoic (Myristic)	0.24	0.23	0.24	0.41	0.26	0.35	0.26	0.12
C14:1 Tetradecenoic (Myristoleic)	0.17	0.09	0.10	0.25	0.17	0.16	0.09	0.03
C15:0 Pentadecanoic	0.04	0.04	0.04	0.06	0.04	0.05	0.04	0.02
C15:1 Pentadecenoic	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C16:0 Hexadecanoic (Palmitic)	2.29	2.04	2.00	3.92	2.37	3.00	2.10	1.12

Cyagra Meat Study

SampleNo				Clone		Comparator		Clone
Marked ID		Comparator		Females		Females		Males
Analyte	Std dev	Mean	Std dev	Mean	Std Dev	Mean	Std Dev	Mean
C16:1 Hexadecenoic (Palmitoleic)	0.68	0.45	0.44	1.10	0.67	0.73	0.42	0.20
C16:2 Hexadecadienoic	0.08	0.06	0.06	0.12	0.08	0.09	0.07	0.03
C16:3 Hexadecatrienoic	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C16:4 Hexadecatetraenoic	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C17:0 Heptadecanoic (Margaric)	0.08	0.09	0.09	0.13	0.10	0.12	0.10	0.05
C17:1 Heptadecenoic Margaroleic	0.11	0.08	0.08	0.17	0.11	0.12	0.08	0.04
C18:0 Octadecanoic (Stearic)	0.93	1.05	1.10	1.52	1.07	1.28	1.14	0.77
C18:1 Octadecenoic (Oleic)	4.49	3.43	3.34	7.42	4.72	5.06	3.52	1.96
C18:2 Octadecadienoic (Linoleic)	0.24	0.22	0.18	0.43	0.27	0.27	0.19	0.18
C18:3 Octadecatrienoic (Linolenic)	0.05	0.05	0.05	0.08	0.06	0.07	0.06	0.02
C18:4 Octadecatetraenoic	0.08	0.04	0.04	0.11	0.09	0.05	0.04	0.02
C19:0 Nonadecanoic	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C20:0 Eicosanoic (Arachidic)	0.00	0.01	0.01	0.00	0.01	0.01	0.01	0.00
C20:1 Eicosenoic (Gadoleic)	0.04	0.02	0.03	0.05	0.04	0.03	0.03	0.01
C20:2 Eicosadienoic	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C20:3 Eicosatrienoic	0.00	0.00	0.01	0.00	0.00	0.00	0.01	0.00
C20:4 Eicosatetraenoic (Arachidonic)	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
C20:5 Eicosapentaenoic	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C21:0 Heneicosanoic	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C21:5 Heneicosapentaenoic	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C22:0 Docosanoic (Behenic)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C22:1 Docosenoic (Erucic)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C22:2 Docosadienoic	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C22:3 Docosatrienoic	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C22:4 Docosatetraenoic	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C22:5 Docosapentaenoic	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C22:6 Docosahexaenoic	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C24:0 Tetracosanoic (Lignoceric)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C24:1 Tetracosenoic (Nervonic)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Total Monounsatur. Fatty Acids Calc.	5.42	4.08	3.93	8.99	5.60	6.10	4.08	2.24
Total Polyunsatur. Fatty Acids Calc.	0.44	0.40	0.33	0.76	0.48	0.52	0.36	0.28
Saturated Fatty Acids	3.51	3.47	3.42	6.07	3.75	4.83	3.63	2.08
Total Fat (as triglycerides)	9.76	8.34	7.99	16.60	10.23	12.01	8.41	4.82
Calcium	13.78	14.30	13.67	10.62	12.84	14.35	15.20	13.67
Iron	0.74	2.32	0.71	2.40	0.78	2.87	0.54	2.17

Cyagra Meat Study

SampleNo				Clone		Comparator		Clone
Marked ID		Comparator		Females		Females		Males
Analyte	Std dev	Mean	Std dev	Mean	Std Dev	Mean	Std Dev	Mean
Phosphorus	27.31	191.21	28.12	166.11	27.47	180.56	27.75	194.67
Zinc	0.67	4.14	0.64	3.71	0.60	4.34	0.61	4.04
Cholesterol	7.79	68.43	8.64	66.93	8.57	64.93	7.67	62.51
Niacin	1.18	5.00	1.07	4.28	0.88	4.73	1.15	5.77
Vitamin B1 - Thiamine Hydrochloride	0.04	0.09	0.02	0.07	0.02	0.10	0.01	0.13
Vitamin B2 - Riboflavin	0.05	0.29	0.04	0.22	0.04	0.27	0.04	0.27
Vitamin B6	0.08	0.37	0.11	0.29	0.07	0.36	0.12	0.37
Vitamin B12	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Vitamin E	0.15	0.44	0.15	0.56	0.14	0.48	0.16	0.44

Cyagra Meat Study

SampleNo	Comparator		
Marked ID	Males		
Analyte	Std Dev	Mean	Std Dev
Crude Fat	3.22	4.35	5.26
Moisture	2.39	71.88	3.20
Protein - Combustion	1.78	22.48	2.50
Protein - Kjeldahl	1.74	22.53	2.44
Ash	0.08	1.12	0.11
Balance (protein+moist+ash+fat)	1.32	99.88	0.73
Amino Acid Profile (results below)			
Tryptophan	0.02	0.27	0.03
Aspartic Acid	0.19	2.17	0.18
Threonine	0.09	1.05	0.10
Serine	0.08	0.88	0.10
Glutamic Acid	0.30	3.61	1.01
Proline	0.16	0.99	0.16
Glycine	0.23	1.15	0.24
Alanine	0.12	1.41	0.17
Cystine	0.03	0.28	0.04
Valine	0.14	1.12	0.11
Methionine	0.07	0.59	0.09
Isoleucine	0.14	1.03	0.09
Leucine	0.17	1.85	0.15
Tyrosine	0.07	0.77	0.06
Phenylalanine	0.09	0.93	0.08
Histidine	0.09	0.75	0.12
Lysine, Total	0.19	2.06	0.16
Arginine	0.11	1.48	0.15
Hydroxyproline	0.06	0.16	0.06
Fatty Acid (results below)			
C08:0 Octanoic (Caprylic)	0.00	0.00	0.00
C10:0 Decanoic (Capric)	0.00	0.00	0.00
C11:0 Undecanoic (Hendecanoic)	0.00	0.00	0.00
C12:0 Dodecanoic (Lauric)	0.00	0.00	0.00
C14:0 Tetradecanoic (Myristic)	0.09	0.09	0.12
C14:1 Tetradecenoic (Myristoleic)	0.03	0.02	0.02
C15:0 Pentadecanoic	0.02	0.02	0.03
C15:1 Pentadecenoic	0.00	0.00	0.00
C16:0 Hexadecanoic (Palmitic)	0.72	0.95	1.23

Cyagra Meat Study

SampleNo	Comparator		
Marked ID	Males		
Analyte	Std Dev	Mean	Std Dev
C16:1 Hexadecenoic (Palmitoleic)	0.17	0.13	0.13
C16:2 Hexadecadienoic	0.02	0.03	0.04
C16:3 Hexadecatrienoic	0.00	0.00	0.00
C16:4 Hexadecatetraenoic	0.00	0.00	0.00
C17:0 Heptadecanoic (Margaric)	0.03	0.05	0.07
C17:1 Heptadecenoic Margaroleic	0.03	0.03	0.03
C18:0 Octadecanoic (Stearic)	0.48	0.80	1.06
C18:1 Octadecenoic (Oleic)	1.30	1.55	1.90
C18:2 Octadecadienoic (Linoleic)	0.10	0.17	0.15
C18:3 Octadecatrienoic (Linolenic)	0.01	0.02	0.03
C18:4 Octadecatetraenoic	0.02	0.02	0.03
C19:0 Nonadecanoic	0.00	0.00	0.00
C20:0 Eicosanoic (Arachidic)	0.00	0.00	0.01
C20:1 Eicosenoic (Gadoleic)	0.01	0.01	0.02
C20:2 Eicosadienoic	0.00	0.00	0.00
C20:3 Eicosatrienoic	0.00	0.00	0.00
C20:4 Eicosatetraenoic (Arachidonic)	0.01	0.02	0.01
C20:5 Eicosapentaenoic	0.00	0.00	0.00
C21:0 Heneicosanoic	0.00	0.00	0.00
C21:5 Heneicosapentaenoic	0.00	0.00	0.00
C22:0 Docosanoic (Behenic)	0.00	0.00	0.00
C22:1 Docosenoic (Erucic)	0.00	0.00	0.00
C22:2 Docosadienoic	0.00	0.00	0.00
C22:3 Docosatrienoic	0.00	0.00	0.00
C22:4 Docosatetraenoic	0.00	0.00	0.00
C22:5 Docosapentaenoic	0.00	0.00	0.00
C22:6 Docosahexaenoic	0.00	0.00	0.00
C24:0 Tetracosanoic (Lignoceric)	0.00	0.00	0.00
C24:1 Tetracosenoic (Nervonic)	0.00	0.00	0.00
Total Monounsatur. Fatty Acids Calc.	1.53	1.73	2.10
Total Polyunsatur. Fatty Acids Calc.	0.16	0.27	0.25
Saturated Fatty Acids	1.31	1.92	2.50
Total Fat (as triglycerides)	3.08	4.10	5.08
Calcium	15.12	13.48	12.63
Iron	0.69	1.69	0.12

Cyagra Meat Study

SampleNo	Comparator		
Marked ID	Males		
Analyte	Std Dev	Mean	Std Dev
Phosphorus	17.67	201.67	24.44
Zinc	0.71	3.83	0.63
Cholesterol	6.17	70.33	8.31
Niacin	0.99	5.44	0.94
Vitamin B1 - Thiamine Hydrochloride	0.03	0.08	0.02
Vitamin B2 - Riboflavin	0.04	0.30	0.03
Vitamin B6	0.06	0.37	0.09
Vitamin B12	0.00	0.00	0.00
Vitamin E	0.13	0.38	0.12

Appendix F:
The Viagen Dataset

Appendix F: The ViaGen Dataset

A. Background

ViaGen, Inc., a privately held company engaged in cloning swine and cattle for agricultural purposes, also responded to CVM’s request for additional data on clones and their progeny (for a similar discussion on cattle, see Appendix E: The Cyagra Dataset). ViaGen designed two studies to evaluate the health, growth, and meat composition of swine clones, fertility of boar¹²⁶ clones, and health, growth and meat composition of clone progeny vs. age-matched, genetically related, artificial insemination (AI)-derived comparator animals. Various scientists from CVM with expertise relevant to these studies provided peer-review comments to ViaGen on the overall experimental design, but did not formally approve or disapprove the studies. All of the raw data that CVM received from ViaGen is found at the end of this Appendix.

As described in their report, the company’s objectives in running these studies were to

1. Compare the health of swine clones vs. AI-derived pigs of a related genetic line;
2. Compare the biological characteristics, including laboratory measurements and meat composition, of swine clones vs. AI-derived swine;
3. Assess the reproductive characteristics of clone boars vs. AI-derived boars; and,
4. Compare the biological characteristics, including laboratory measurements and meat composition, of the progeny of clone boars vs. progeny of AI-derived closely-related boars (nuclear donors for clones or progeny of nuclear donor).

B. Experimental Design

Study 1 Overview: Evaluation of Clones

Survival, health, growth and meat characteristics of seven clones (6 “Hamline”¹²⁷ and 1 Duroc (a common breed of swine used in U.S. commercial pork production), all barrows¹²⁸) and 15 conventional barrows (all Hamline) were followed from 50 days after birth through slaughter at approximately 6 months of age. Clones were assigned to this study shortly after they were

¹²⁶ A boar is a reproductively intact male pig.

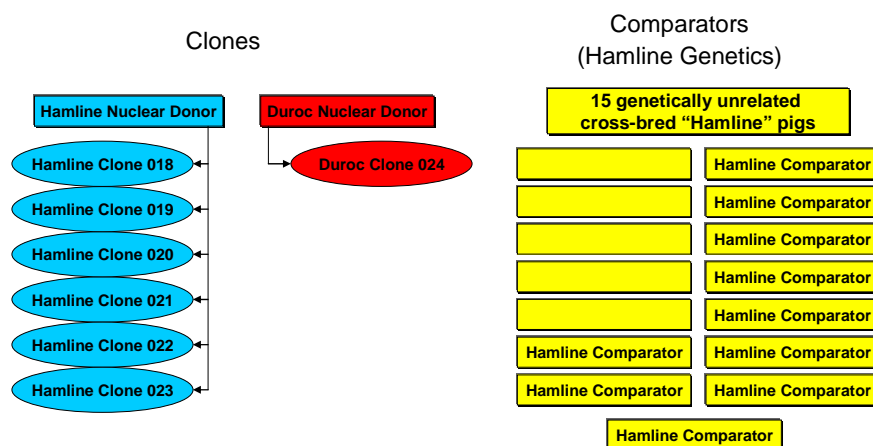
¹²⁷ “Hamline” refers to a specific crossbred line of swine used by ViaGen, Inc. This line was developed by crossing various breeds, including Duroc, European Landrace, Pietran, and European Large White swine.

¹²⁸ A barrow is a castrated male pig.

weaned. Age-matched comparator¹²⁹ pigs were selected from litters sired by the Hamline nuclear donor boar in a conventional breeding (AI) program.

An additional group of four clones (three Hamline and one Duroc) and three AI-derived boars (the Hamline nuclear donor and two AI sons of the Duroc nuclear donor) were used for evaluation of semen quality and fertility. The progeny generated from these boars were used in another experiment to study whether progeny of clones were materially different in growth, health or meat characteristics from conventional swine. Because of the small number of clones, these animals were evaluated on a case-by-case basis and simple means generated when appropriate, rather than applying statistical methods to the data.

Figure F-1: Design of ViaGen Study 1



Study 2 Overview: Evaluation of Clone Progeny

In the second experiment, survival, health, growth, and meat composition of progeny derived from clone boars (three Hamline and one Duroc) and either their nuclear donor (Hamline) or two sons of the nuclear donor Duroc boar were compared. Gilts¹³⁰ sired by six different boars were randomly assigned according to their sire (such that each maternal grandsire was represented in

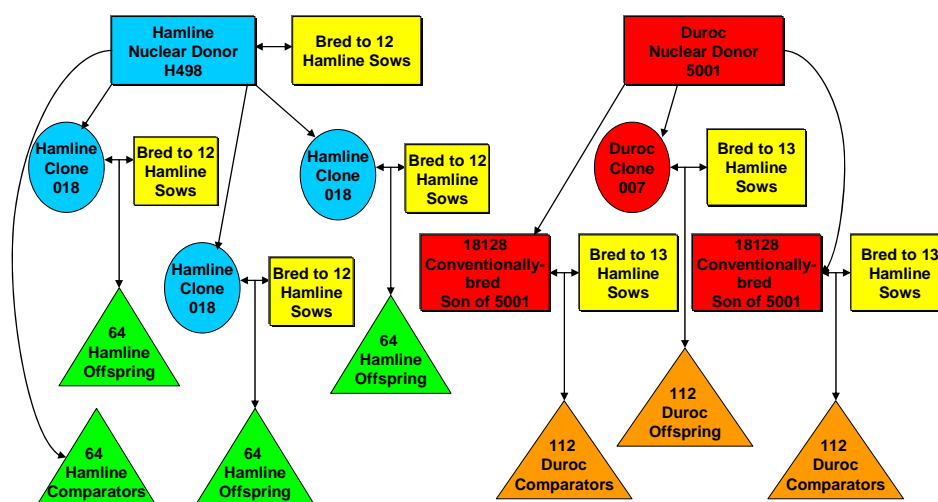
¹²⁹ Although this study is a more controlled experiment than the retrospective review of the Cyagra clones, the word “comparator” is used for consistency with the discussion of Cyagra data, rather than the more common term “control.”

¹³⁰ A gilt is a young female pig that has not given birth (termed “farrowing”).

each breeding group to control for effect of maternal grandsire) to each clone or comparator boar, and bred by AI. Sixty-eight gilts farrowed (gave birth) under standard commercial procedures, yielding 402 total progeny from clone boars and 300 total progeny from comparator boars. This included 284 progeny from Hamline clones, 118 progeny from the Duroc clone, 61 progeny from the Hamline donor sire (Hamline comparator), and 239 progeny from Duroc donor sire sons (Duroc comparators).

Figure F-2: Design of ViaGen Study 2

Study II: Design



1. Study 1: Clones vs. Comparators

Clone pigs were obtained by Caesarean section (C-section) on the day of or the day before the sows' predicted farrowing date. Clone piglets were kept under highly biosecure¹³¹ conditions prior to initiating the experiment. Clones were bottle fed commercial milk replacer and did not receive colostrum at the time of birth. Comparator pigs, however, were born naturally (i.e., vaginally) in a standard commercial farrowing house, and suckled their dams until weaning. By

¹³¹ "Biosecure" connotes conditions that, although not completely sterile, are designed to prevent introduction of most disease-causing organisms. Because animals' immune systems produce antibodies in response to specific stimuli (antigens), animals raised under biosecure conditions lack the full battery of antibodies needed to respond to common pathogens found in conventional environments (although they may have some antibodies if they have been vaccinated, these antibodies will be specific for the antigen used in the vaccine). It can take up to two weeks for an animal to develop a sufficient concentration of antibodies to a specific antigen following exposure.

the start of the test period (~50 days of age), the clones were similar in weight to the comparator pigs (16.4 vs. 15.3 kg for clones and comparators, respectively). In addition, comparator barrows were castrated during the first week of life, as per normal industry practice, but clone barrows were castrated at a later age (~30 days old). Pigs were assigned to this study shortly after they were weaned. Comparator barrows were selected from litters sired by one of the nuclear donor boars in a conventional (AI) breeding program. At the start of Study 1 clones and comparator animals were housed in the same facility. Target slaughter weight was approximately 122 kg.¹³²

a. Animal Health, Hematology, Clinical Chemistry, and Urinalysis

Health records from date of birth were submitted for clones, and included birth weight, heart rate, respiration, body temperature, body weight, and daily health observations. All health problems and treatments were recorded. Necropsy reports were provided for clones that died or were euthanized. Blood samples of clones and comparators were collected at birth and again either approximately one week prior to slaughter, or immediately after animals were slaughtered. Urine samples were collected after slaughter (by bladder puncture). Blood hematology, clinical chemistry, and urine values were assayed as for the Cyagra dataset at the same independent testing laboratory (Cornell University's Animal Health Diagnostic Laboratory, see Appendix E).

b. Boar Semen Evaluation

Semen samples were evaluated in two commercial boar lines (Hamline and Duroc). The Hamline included three clone boars and the sexually derived nuclear donor boar for the clones as a comparator. The Duroc line included one clone boar and two comparators that were AI-derived progeny of the donor (semen from the Duroc nuclear donor was unavailable). Clones and comparator animals were housed at separate facilities. Semen was collected at least once a week from each boar, was diluted with a commercial semen extender to yield 3×10^9 sperm per dose, and cooled to 19°C per standard industry practice. Semen from clones was evaluated pre-freezing and post-freezing (post-shipping) to determine quality. Comparator boar semen was only evaluated post-shipping. Standard tests such as sperm concentration, percent total motility, percent progressive motility, and number of head/tail abnormalities were evaluated.

c. Farrowing Rate

Following semen evaluation, semen from the four clone boars and three comparator boars were used to evaluate pregnancy (number of females diagnosed pregnant divided by number of

¹³² Swine are typically slaughtered in groups. Usually, all swine raised together in a pen will go to slaughter on the same day once the average weight of the animals within that pen has reached a pre-set target weight. The date of slaughter can be affected by the availability of transportation and the capacity of the slaughter facility.

females bred) and farrowing (number of females giving birth to a litter divided by the number of females bred) rates. Three of the clone boars were bred to 12 gilts each and one clone boar was bred to 13 gilts (total of 49 gilts bred). For the comparator boars, two were bred to 14 gilts each and one was bred to 12 gilts (a total of 40 gilts bred). All gilts were bred via AI twice, once on the day of observed estrus and approximately 24 hours later.

2. Study 2: Progeny of Clone Boars vs. Progeny of Conventional Boars

Breeding resulted in 36 litters (402 total progeny) from the clone boars and 25 litters (300 total progeny) from the comparator boars. Reproductive outcomes evaluated included litter size, birth weight, number of stillborns, number of dead/destroyed animals, abnormalities, and nipple counts (another measure of breeding fitness often taken at birth). Animals were slaughtered when they had reached the target weight of approximately 122 kg. Average age at slaughter was 169.3 days for progeny of comparators and 174.9 days for progeny of clones.

a. Statistical Analysis

i. Methodology

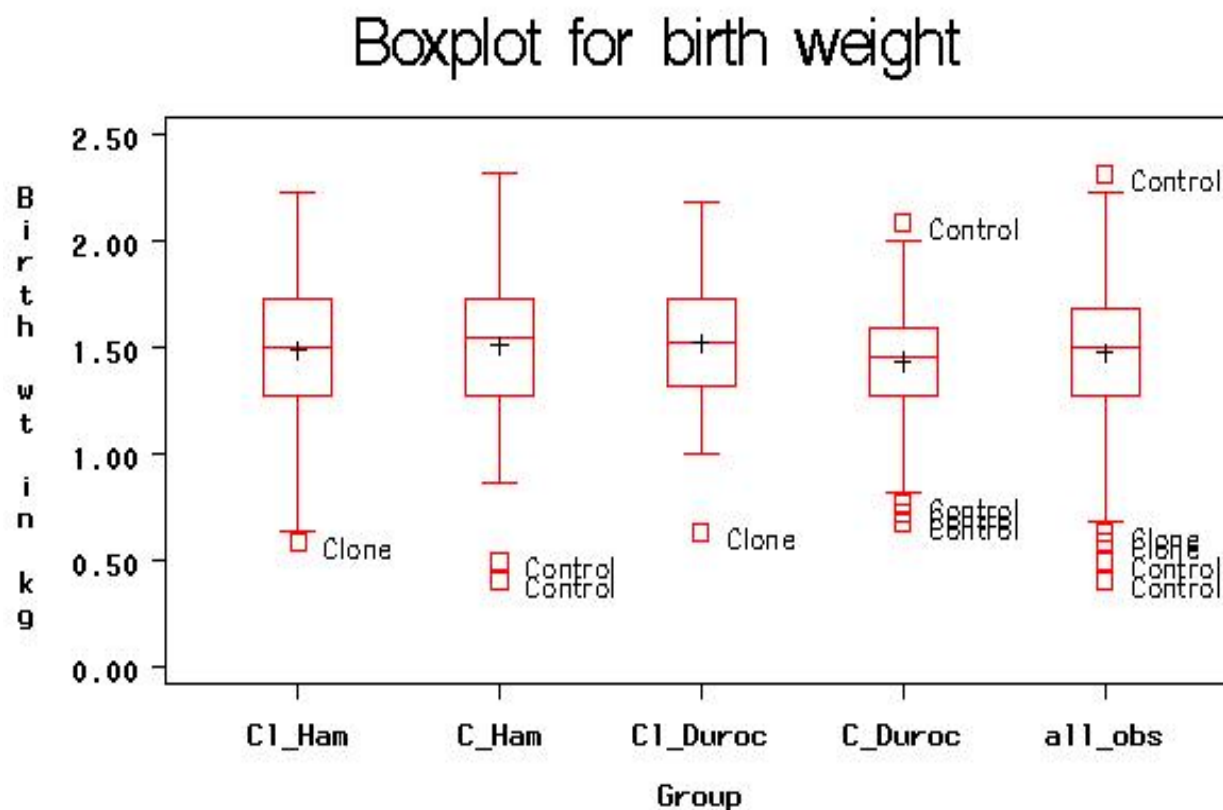
To aid CVM's analysis of the very large sample sizes in Study 2, the Center's biostatisticians produced data summaries to facilitate veterinary and scientific evaluation of the outcomes collected from the progeny. Two types of summaries were used: contingency tables and boxplots. Definitions and examples of both are given below.

Contingency tables present data in a row and column format and are often used to summarize the results of discrete variables. A variable can be defined as discrete if its responses are limited to a few predetermined values. For example, a discrete variable from Study 2 is "Abnormality at birth" which is described according to four possible discrete outcomes: Normal, Atresia, Spraddle legs, or Other. For those progeny for which this variable was recorded, the distributions of "Abnormality" for the live-born progeny of animal clones and progeny of comparators are displayed in the following contingency table (Table F-1).

Table F-1: Contingency Table Data Description for Distribution of Abnormalities for Progeny of Clones and Progeny of Comparators				
Abnormality	Progeny of Clones		Progeny of Comparators	
	Number	Percentage	Number	Percentage
Normal	384	97.5%	291	99%
Atresia	1	0.3%	0	0%
Spraddle Legs	4	1.0%	3	1.0%
Other	5	1.2%	0	0%
Total	394	100%	294	100%

By contrast, continuous variables are those whose responses can take on a continuum of values. Graphical displays are useful in summarizing the responses from continuous variables. The boxplot is one type of graphical display. An example of a continuous variable from Study 2 is “animal weight.” The boxplots of birth weights from the progeny experiment are provided as an example below.

Figure F-3: Boxplot of birth weights of progeny of clones and comparators (in kgs)



In Figure F-3, the boxplot on the far right includes all of the birth weights recorded for clone progeny and comparators. To create a boxplot, the birth weight data was ordered from smallest

to largest. The bottom edge of the box was drawn at a value of birth weight for which 25 percent of the observations have a lower value, which is called the “25th percentile”¹ (or first quartile) of the sample. Similarly, the top of the box was drawn at a value of birth weight for which 75 percent of the observations have a lower value, which is called the “75th percentile”¹³³ of the sample. The value of the observation associated with the 75th percentile minus the value of the observation associated with the 25th percentile is called the interquartile range (IQR). Within the box, a plus sign is used to indicate the sample mean and the line from side to side of the box indicates the sample median which is the value of the observation associated with the 50th percentile below which half the observations lie. Any observation values which lie above the “upper fence,” a point equal to 1.5 times IQR plus the value associated with the 75th percentile or below the “lower fence,” a value equal to the value of the observation associated with the 25th percentile minus 1.5 times IQR, are individually plotted on the graph and are called extreme values. (Note the actual fence values are not plotted.) The whiskers stemming out from the boxes represent the observation just below the upper fence and the observation just above the lower fence. In the example above, the boxplots, from left to right, are for progeny of Hamline clones, progeny of Hamline comparator animals, progeny of Duroc clones, and progeny of Duroc comparator animals.

Boxplots do not require assumptions about the form of the underlying continuous data distributions and are uniformly applicable across a range of different distributions. When boxplots are used to describe a variable sampled from a single population and having a normal distribution, they tend to identify about 1 percent of values above and 1 percent of values below the box as “extreme.”

When the underlying distribution has more data in the tails than a normal distribution, then more than 1 percent of the distribution is identified as extreme. To evaluate the ViaGen data, values from the progeny of clones were compared to the ends of the whiskers in the boxplots for the values from the progeny of comparators.

ii. Utility of Statistical Analysis as a Tool for Assessment of Outcomes

Results (numerical values) for any outcome from the clone progeny were compared to the ends of the whiskers for the distribution of values from progeny. If a result fell within the range defined by the box and whiskers, it was considered “within the same range as the comparators” and therefore posed no significant concern for the health of the animal or the composition of the food product. If a result for a clone fell outside the range identified by the whiskers for the comparators, or was identified because of its value in a contingency table, the appropriate CVM

¹³³ A percentile is defined as $(1/(\text{total sample size})) * 100$

scientist (e.g., veterinarian, animal scientist, chemist, toxicologist) traced back all of the data for that the individual animal, and conducted a thorough examination of all of the relevant clinical information to draw a conclusion regarding the biological or health significance of the value.

In addition, to provide consistency with the presentation of the analysis developed for the Cyagra dataset (Appendix E), box charts were constructed. As described in that Appendix, black boxes or up or down arrows were assigned to values for each clone progeny by the reviewing veterinarian or scientist on the basis of this multi-step review procedure.

b. Specific Methods for the Analysis of Animal Health, Hematology, Clinical Chemistry, and Urinalysis Data

Progeny of clones and comparators were farrowed during a three week period from July 7-27, 2004, at the Meat Animal Research Center, USDA, Clay Center, Nebraska. All available animals were sampled (serum and whole blood) in July, October, and January. Blood samples were obtained between 7:30 and 10:30 AM. After collection, samples were placed on ice until centrifugation at 900 x g for 10 min at 4°C. Serum was collected and stored at -20°C until analysis for estradiol-17 β (E2), and Insulin like growth factor-I (IGF-I). These time points represented early, middle and late stages in the life of the test subjects, which mirrors the lifespan of animals used for commercial pork production in the U.S. When considered in the context of the CBSA approach used in this Draft Risk Assessment, these time points roughly correspond to the Perinatal (July) and Juvenile (October and January) developmental nodes, as most pigs used for food are slaughtered before they reach sexual maturity. July and October blood samples were collected from the cranial vena cava. In January, blood and urine samples were harvested post-mortem. Pigs were individually identified by numbered ear tags, which did not reveal to caretakers or personnel taking samples whether the animals were the progeny of clones or comparators.

c. Meat Composition, Carcass Characteristics, and Meat Quality Assessments for Clones, Comparators, and Progeny.

As in standard US pork production, swine were slaughtered when they reached approximately 122 kg in weight. Samples were collected from the *latissimus dorsi* muscle (the muscle that comprises the large, round-shaped muscle next to the vertebrae end of the ribs that makes up the boneless portion of the “rib eye” roast or steak), frozen, and held at -20°C for approximately two weeks prior to shipping to a commercial laboratory for compositional analysis. Samples were identified by a nine digit code so that personnel at the analytical laboratory did not know which samples belonged to comparators, clones, or progeny of clones. Muscle samples (approximately 0.45 kg, deboned) were held frozen at -20°C for three to 20 days after receipt at the laboratory

before being partially thawed (10 – 18°C overnight) and homogenized using a commercial grade meat grinder with a #12 blade and passed through a 1/8 inch screen. Homogenized meat samples were analyzed for fatty acid and amino acid profiles, cholesterol, vitamins B₆, B₁₂, niacin, calcium, iron, phosphorus, and zinc. Analyses were carried out according to the Association of Analytical Communities International (AOAC) methods for amino acids (982.30), calcium, iron, phosphorus and zinc (965.17 and 985.01), cholesterol (994.10), fatty acid profile (996.06), niacin (944.13), vitamin B₁₂ (952.20) and vitamin B₆ (961.15). With the exception of vitamins B₆, B₁₂, niacin, and cholesterol, values were reported as percentages. Vitamin B₆, niacin and cholesterol were reported as mg/100 g of sample, and vitamin B₁₂ was reported as mcg/100 g of sample. Percentages were based on grams of nutrient per 100 grams of homogenized meat sample.

Economically important traits for the swine carcass evaluation include animal live weight, dressing percentage, carcass length, loin muscle area, backfat depth, extent of muscling, and firmness. These are described in the following paragraphs.

The normal range for dressing percentage¹³⁴ is 68-77 percent (average of 72 percent) with a minimum carcass length of 76.2 cm. The minimum acceptable area for loin muscle is 11.4 cm² and the maximum back fat thickness over the last rib (10th or 11th rib) is 3.8 cm.¹³⁵

Three degrees of pork carcass muscling are recognized in the pork grading standards or USDA¹³⁶ carcass muscling score: muscle score #1 = thin (inferior), muscle score #2 = average, and muscle score #3 = thick (superior). Scores of either 2 or 3 are considered acceptable under USDA inspection standards. Muscle firmness is a subjective score determined by visual observation and physical handling of the meat. Muscle firmness is measured on a three-point scale: score #1 = soft, score #2 = firm, and #3 = very firm. As with muscling score, a firmness score of 2 or 3 is considered desirable.

C. Results

1. Study 1

a. Survival

Seven clones and 16 comparators were raised by ViaGen Inc. The seven clones began the study at 50 days of age, and all seven survived until the end of the observation period (approximately

¹³⁴ Dressing percentage refers to the proportion of the live animal that ends up as the carcass used for food production, or Carcass Weight / Live Weight X 100.

¹³⁵ <http://www.animalrangeextension.montana.edu/Articles/Swine/2-Det-quant.htm>

¹³⁶ <http://ars.sdstate.edu/AnimalEval/Swine/SwineGrade.htm>

195 days of age); however, two clones were euthanized due to chronic health problems at the end of the study. Thus, five clones were slaughtered as per normal industry practice. One of these clones was diagnosed with a lung adhesion post mortem, which, by USDA standards would have led to condemnation of the carcass. Similarly, 15/16 comparators reached the end of the observation period (approximately 171 days of age), with one comparator euthanized approximately four months prior to the end of the study because of a chronic respiratory condition.

Because litters of clones were born approximately six weeks apart, they were separated into two groups of animals and matched with comparators born at approximately the same time. In the first group, five clones and 11 comparators were raised and studied. In this first group, one clone (#21, farrowed 10/13/03) and one comparator (#025435) did not thrive. Clone #21, a Duroc, was euthanized at the end of the study (May 27, 2004) and was not included in the final carcass evaluations because of low weight. However, hematology and clinical chemistry values for this clone were used for analysis of health data. Non-clone comparator #025435 was euthanized on December 4, 2003 because of a respiratory condition that was not improving.

ViaGen raised two clones and five comparators in the second group. Clone #23, a Hamline, farrowed on 11/23/03, was described as a “poor doer” and was euthanized at the end of the study. All comparators from this group survived to slaughter.

b. Animal Health, Growth, Blood Clinical Chemistry, Hematology, and Urinalysis

Clones were smaller on average at birth than their conventionally bred counterparts (1.1 vs. 1.7 kg) (Table F-2). By the start of the test period (~50 days), however, clones were similar in weight to their comparators (16.4 vs. 15.3 kg).

Animal health records indicated that six of the seven clones developed scours (severe diarrhea) approximately two months after birth, which would have been shortly after they were moved from the biosecure environment. The affected clones were #18, 19, 20, 21, 22 and 23. One of these animals (Clone #22) was diagnosed at approximately four months of age with influenza and secondary bacterial septicemia, was periodically treated with antibiotics throughout the experimental period, and was eventually euthanized at the end of the study. One of the comparator animals (#025435) was diagnosed at approximately one month of age with influenza and secondary bacterial septicemia. This animal was periodically treated with antibiotics throughout the experimental period and was eventually euthanized. The clones weighed less at slaughter and took an average of 27 days longer to reach the target slaughter weight. As a result, the calculated average daily (weight) gain (ADG) for clones was lower than for comparators

(0.63 vs. 0.93 kg/day). The clones were slaughtered later than comparators because of the greater length of time needed for them to achieve the target weight, and also because the slaughter facility could only accommodate them on a few specified days. This latter point was because clones are currently withheld from the food supply, and the facility preferred to slaughter the clones on days when no other swine were being processed, in order to prevent accidental mixing of the clones with conventional animals.

	Comparators		Clones	
	Age	Body Weight	Age	Body Weight
Birth weight	—	1.72 ± 0.28a (n=15)	—	1.13 ± 0.09b (n=4)
Post-weaning	22.4 ± 4.81 (n=15)	15.3 ± 5.99 (n=15)	21.2 ± 1.81 (n=4)	17.0 ± 3.40 (n=4)
End of Experiment	77.9 ± 5.62a (n=15)	128.9 ± 7.26 (n=15)	88.5 ± 8.16b (n=4)	122.2 ± 8.93 (n=4)

Note: one comparator is not included because it was euthanized at about 1 month of age. Three clones are not included due to low weight.

As previously mentioned, clones and comparators were reared under different conditions prior to starting the experiment. Clones were delivered by C-section, did not receive colostrum, were fed a commercial milk replacer, were castrated at an older age, and were raised in a biosecure environment prior to initiation of the study. The clones were then housed along with the comparator animals in an environment similar to commercial swine facilities, where they were exposed to a typical range of pathogens. The comparators were farrowed vaginally following natural initiation of labor in a farrowing house similar to commercial swine operations, and the progeny were allowed to nurse off their dams. This disparity likely accounts for some of the differences in growth and health observed. The sudden change from a biosecure environment, with a low immune challenge, to a more conventional barn with a high immune challenge placed considerable stress on the immune systems of the young clones. This stress likely led to the higher incidence of scouring in this group, and may have resulted in decreased food consumption or absorption, and increased energy expenditure to combat illness.

Body temperature, heart and respiration rates were measured in clones, but not in comparators. Because of the absence of comparator data, observations for these measurements are put in the context of common veterinary practice or reference texts or manuals.

Table F-3: Average Body Temperature, Heart Rate, and Respiration Rate for Neonatal Clones During First Eight Days After Birth

Day	Temperature (°F) ¹	Heart Rate (beats/min)	Respiration Rate (breaths/min)
1	100.38 ± 0.18	194.33 ± 1.42	87.56 ± 0.71
2	100.80 ± 0.17	184.22 ± 0.96	79.61 ± 0.53
3	101.40 ± 0.19	179.96 ± 0.92	75.72 ± 0.53
4	101.43 ± 0.19	177.30 ± 1.03	72.52 ± 0.59
5	101.53 ± 0.21	177.28 ± 1.23	71.81 ± 0.71
6	101.41 ± 0.23	177.94 ± 1.23	71.06 ± 0.71
7	101.59 ± 0.22	179.00 ± 1.19	70.29 ± 0.79
8	101.89 ± 0.21	178.11 ± 1.18	72.34 ± 0.68

¹ Temperatures were recorded and analyzed in Fahrenheit. Standard deviations are not convertible to Celsius.

Body Temperature (BT). Clones #18, 19, 20, 22, and 24 were measured for BT 2-9 times per day for the first eight days of life (no records on BT for other clones were available). Body temperature increased over this eight day period from an average of 100.38 ± 0.18°F (37.99°C) on Day 1 to 101.89 ± 0.21°F (38.83°C) on Day 8. The low BT was 97.7° F (36.5°C), and occurred in clones #22 and 24 early in the study (Days 1 and 2 post farrowing). The high temperatures were 104.6°F (40.33°C), 103.8°F (39.89°C), and 103°F (39.44°C) and occurred in one clone (#24), twice on Day 1, 1, and 8 respectively. The Merck Manual's Reference range for swine BT is 101-103°F (38.33 – 39.44°C). The BT indicates an apparent appropriate response to autonomous BT regulation (adjustment to post-natal environment) after delivery from the uterus where BT is not self-regulated, but controlled by the intra-uterine environment. As mentioned in Chapter V, newborn pigs typically cannot regulate their own BT. Generally, it is recommended to maintain the environment of newborn pigs at a temperature of 86-93°F (30-34°C) for the first week to allow for this adaptation period (Merck Veterinary Manual Online).

Heart rates (HR). The heart rates (HRs) of clones also were measured 4-12 times per day for the first eight days of life. Heart rate decreased over time from 194.33 ± 1.42 to 178.11 ± 1.18 beats per minute (bpm). The highest value was 220 bpm for clone #18 on Day 1; the lowest value is 100 bpm for clone #22 on Day 1. This decrease is an appropriate response for neonates. Heart rates decrease with age as fetal circulation (ductus venosus,¹³⁷ foramen ovale,¹³⁸ and

¹³⁷ The ductus venosus carries oxygenated blood from the umbilical vein to the caudal vena cava, bypassing the fetal liver.

¹³⁸ The foramen ovale is a hole between the right and left atria in the fetal heart, for the purpose of by-passing the fetal lungs *in utero*.

ductus arteriosus¹³⁹ closure) normalizes and the neonate develops autonomic control (Medline, Medical Encyclopedia¹⁴⁰). There were no values from the non-clone group for direct comparison. These values should be evaluated with caution because the very act of auscultating (listening to) the thorax of an animal that is seldom handled can raise HR artifactually. In fact, it is possible that the reduction in HR over time represents the pigs becoming more accustomed to handling. Data on HR for non-clone neonatal swine are not routinely reported; however, one study (Foster et al. 2001) recorded HR of day-old piglets before and after insertion of a breathing tube. Prior to insertion of the tube, average HR was 190.75 ± 36.45 bpm. In another study (Aaltonen et al. 2003) examining the effects of meconium aspiration and asphyxia in piglets aged 10 to 12 days. Average HR for the control (untreated) group was 203 ± 23 bpm. Given these data for comparison, the HR of clone pigs during the first eight days of life fell within or slightly below the range of non-clone pigs of similar ages.

Respiration. The same clones as above were evaluated 5-12 times per day for the first eight days of life. Respiration rates decreased over time, ranging from 87.56 ± 0.71 breaths/minute on Day 1 to 70.29 ± 0.79 breaths/minute on Day 7. As with HR, data on swine neonatal respiration rates are not commonly reported. However, one study (McDeigan et al. 2003) measured respiratory responses in pigs from 3 to 7 days of age before and after exposure to either saline (control) or *Escherichia coli* endotoxin. Average pre-treatment respiration rate for control piglets was 44 ± 5 breaths/minute in this study, and thus considerably lower than the average respiration rates of the clones in the ViaGen study. The lowest daily average respiration rate among the neonatal clones in the ViaGen study was 69.6 breaths/min on day 7 (Clone #18). The cautions mentioned for interpreting HR data should also be applied to interpreting respiration rates. Also, animals delivered by C-section have a transient tachypnea (rapid breathing) because the pulmonary surfactant and fluid is not distributed and squeezed out of the lungs as it would in a vaginal birth (Medline, Medical Encyclopedia).

Daily Health Records. Clones #18, 19, 20, 21, 22, and 23 all had scours (severe diarrhea) while on study. Clone #18 herniated a loop of bowel post-castration, which was repaired, and the pig responded well. This pig had an inguinal hernia, sometimes referred to as a “busted pig,” which is not uncommon in swine (Gatphayak et al. 2005). As previously mentioned, clone #21 was described as a “poor doer” (unthrifty, poor weight gain) and was euthanized because of low body weight. He had chronic scours and a skin abscess on his neck. Clone #22 had scours twice while on study. He developed sepsis (a blood infection), and had a low body weight at slaughter. Clone #23 had many health problems, including scours, respiratory disease, cyanotic (bluish) skin

¹³⁹ The ductus arteriosus carries oxygenated blood from the main pulmonary artery to the descending aorta, bypassing the fetal lungs.

¹⁴⁰ <http://www.nlm.nih.gov/medlineplus/ency/article/003399.htm>

color, and grew slowly. He was euthanized because of his low body weight. Clone #24 had some respiratory difficulty toward the end of the study but responded to therapy. One comparator pig (#025435) was euthanized for non-responsive diarrhea and pneumonia early in the study. Another comparator (#027446) had one reported health problem of a swollen dewclaw that responded to therapy.

Although these clones had more health problems than comparators, it is important to note that differences in early rearing are likely to have contributed to these outcomes. First, the clones were not suckled on their dams, but were fed commercial milk replacer. This deprived them of the passive immunity provided by transfer of maternal antibodies in the colostrum. Secondly, pigs raised in a biosecure environment would not have developed antibodies to common environmental pathogens, and would be at increased risk of developing infections until their immune systems adjusted to the environmental challenges when they were moved to less-biosecure conditions at day 50 of age. The comparators, on the other hand, were born in conditions equivalent to commercial swine breeding, and were allowed to suckle on their dams, thereby allowing their immune systems to develop along more conventional lines. We also note that even though some of the clones developed illness, most (5/7) were able to respond to treatment. Further, the hernia observed in clone #18 is not unique to clones, but is considered common among male pigs in the general swine population (Merck Veterinary Manual Online).

Clinical Chemistry and Pathology. Clinical pathology is the term generally used for laboratory findings that includes clinical chemistry, hematology, and urinalysis among others. As described in Appendices E and G, clinical chemistry and hematology responses are best evaluated in the context of the whole animal, including its age, species, breed, husbandry, geographic location, reproductive status, and the laboratory performing the analysis. Laboratory findings complement the subjective physical diagnosis of the patient by providing objective information for the process of differential diagnosis, monitoring treatment, and formulation of a prognosis. “Abnormal” laboratory measurements and examinations are often defined as those values lying outside the limits of the reference range. Determining what constitutes “normal” is more complex than simply comparing an individual value to a reference range derived from a sample of a representative population. In this study, the firm provided serum chemistry data, white cell counts, red cell counts, and urinalysis using a rapid “dipstick” test. Because the number of pigs in the study was low, a statistical analysis of the comparator ranges was not appropriate. Values from clones were compared to those from the comparators.

Across all three sampling periods, 89 percent (281/315) of clones’ hematology measurements were within the range of the comparator population. At the beginning of the study, 80 percent of the clones’ hematology values (84/105) were within the range of the comparators. This

percentage improved at the next sampling period, approximately two months later, such that 88 percent of the clones' values (92/105) were within range. For clinical chemistry, 76 percent of the clones' values (236/315) were within the comparator range across all time periods. As with hematology, the percentage of clone values within the range of comparators was lower at the beginning of the trial, but this ratio improved as the animals grew. At the beginning of the study, 63 percent (66/105) of the clones' values were within the comparator range. By the second sampling period, 83 percent (88/105) of the clones' values were within the comparator range. By the end of the trial it appeared that clones for the most part had stabilized, with 84 percent of the hematology values and 98 percent of clinical chemistry values within the comparator range (Table F-4).

Sampling Period	Number of Variables	Number of Observations	Number of Clones	Number of Out of Range Values	Percentage Out of Range	Number of In Range Values	Percentage In Range
1 st hematology	21	105	5	21	20%	84	80%
1 st clinical chemistry	21	105	5	39	37%	66	63%
2 nd hematology	21	105	5	13	12%	92	88%
2 nd clinical chemistry	21	105	5	17	16%	88	84%
3 rd hematology	21	105	5	17	16%	88	84%
3 rd clinical chemistry	21	84	4	23	2%	82	98%

Seven comparators had elevated WBC counts with no apparent health problem. Clone #20 had a high WBC (32,000 cells/ μ l) during an episode of diarrhea. This is an appropriate clinical response. One clone (#18) and one comparator (#025457) had low platelets at the end of the study. Red blood cell size for both clones and comparators indicated a microcytosis (low MCV). Because the clones and comparators were similar, this value is likely related to the laboratory and not a difference in health.

Clone #21 had elevated BUN levels at the start and middle of the study and elevated liver and muscle enzymes at the start and end of study. He did not show an increase in WBC, and had a slight increase in urine glucose at the end of the study. These findings are consistent with a “poor-doing” animal. Animals in a negative energy balance (due to inadequate nutrition, food

intake or utilization) will start to mobilize muscle protein for energy once their body energy stores are depleted. Blood urea can elevate with increased protein digestion or mobilization. CK (an intracellular enzyme for muscle) will also increase with muscle break down. The elevated liver enzymes indicate some insult to the liver. The cause of these health problems is not evident from the data presented.

Clone #23 exhibited an increase in blood glucose at the start of the study (January) and bile acids at the mid-point of the study (March). By the end of the study (May), clone #23 had glucose and bile acid values within the range of the comparators. Clone #23 was also a “poor doer” (poor weight gain), but had no other clinical pathology abnormalities evident. Although increased blood glucose levels may indicate metabolic disturbances such as diabetes, the increased glucose levels in this study (in blood and once in urine) appear to have been transient and resolved spontaneously. They therefore do not appear to be indicative of an insulin deficiency or resistance. Bile acids are an indicator of liver function (see Appendix G). If the bile acid value is consistently elevated, liver insufficiency may be indicated. However, bile acid levels are also dependent on when the animal was fed, and increase after a meal. Because information on the relationship between blood draws and feeding times was not provided, no conclusion can be made about the single measurement which on its own does not suggest a health problem.

Urinalysis. Urine values were determined by dipstick. There was apparently no Specific Gravity (SG) determination using a refractometer or sedimentation evaluation, so SG was measured only by the dipstick method, which is less accurate than these other methods. With these caveats in mind, two clones and one comparator had glucose in their urine. The percent of clones and comparators with blood and protein in their urine were similar. Seven out of 15 urine samples from clones were positive for blood (47 percent) vs. 16/ 44 (36 percent) samples from comparators. For protein, 6/15 samples from clones (40 percent) vs. 20/44 (45 percent) samples from comparators were positive. Color, turbidity, SG, bilirubin, pH, ketone, nitrite, and leukocytes were all similar between clones and comparators. The presence of blood in the urine may have been due to the fact that these samples were taken after the pigs had been slaughtered.

Organ weights. The clones’ body weights were lighter on average than comparators at slaughter (117 vs. 128.8 kg). Clones #21 and 23 were so light (86.2 and 89.8 kg, respectively), they were not included in the organ weight analyses. Kidneys were smaller for clones vs. comparators on a percent of body weight basis. However, heart, liver, lungs, and spleen were similar as a percent of body weight for clones vs. comparators. With no clinical chemistry indicators of renal insufficiency in clones, the smaller kidney weights are interesting but not conclusive. These findings indicate no appreciable differences between clones and comparators in organ size as a percent of body weight.

IGF-1 and E2. In general, the clones had lower levels of IGF-I than the comparators after birth and before slaughter (Table F-5). However, the values were within the range observed in the comparator group, except for one clone (Clone #19), that had lower levels at the beginning of the study. This clone had levels of IGF-I at the limit of assay sensitivity (low value). Before slaughter, clones #19 and #20 presented levels of IGF-I that were close or below the detectable values by the analytical method.

Although, critical illness can modify and has an important impact on the human endocrine system (Sartin et al. 1998), these animals did not show health-associated conditions at the time of sampling that could explain the low levels of IGF-I. Additionally, the IGF-I axis (the glands, organs and hormones associated with IGF-I), as well as cortisol and gonadal steroids, are endocrine determinants of the growth potential of animals (Mauras and Haymond 2005). Nevertheless, the body weights of the animals with low IGF-I (clones #19 and #20), were not lower than the control animals. This observation suggests that the reduction in IGF-I levels seen before slaughter for these two clones is not biologically important and perhaps is associated with the handling of samples or time of sampling relative to when the animals ate. In addition, limited information has been provided describing the handling and storage of samples in the current experiment, and hemolysis and storage of blood samples have been shown to reduce detectable concentrations of hormones using the assay technique employed in this study (Reimers et al. 1991). Therefore, the possibility that the low levels of IGF-I in clones #19 and #20 may be due to hemolyzed serum used for the hormone analysis cannot be excluded.

Clones had slightly lower levels of E2 than the comparator group; this trend was observed during the perinatal period and before slaughter (Table F-6). However, the E2 values of clones were within the range of the comparators, but one comparator animal (#025461) displayed elevated levels of E2, as a result, the mean values of the comparator group increased. This animal also presented the highest rate of weight gain throughout the study. The body weight at slaughter was higher than the other comparators. If the data for this animal is removed from the comparator group, the apparent difference between animal clones and comparators is considerably diminished. Considering the above observations it can be concluded that the slight reduction in the levels of E2 in the clones was not biologically relevant.

Collection period	Estradiol-17 β (pg/ml)		IGF-I (ng/ml)	
	Clones	Comparators	Clones	Comparators
Start	9.4 \pm 0.4 (n=5)	16.10 \pm 1.4 (n=15) 11.48 \pm 1.6 (n=14)	142 \pm 23.4	235.9 \pm 6.7 (n=15) 222.03 \pm 27.6 (n=14)
End	15.9 \pm 0.9 (n=5)	26.7 \pm 2.4 (n=13)	64.02 \pm 12.8	93.9 \pm 7.2 (n=13)

Clone ID	Start of Study		End of Study	
	Estradiol-17 β (pg/ml)	IGF-I (mg/ml)	Estradiol-17 β (pg/ml)	IGF-I (ng/ml)
18	6.82	45.80	17.84	91.60
19	10.91	114.50	18.71	22.90
20	10.21	68.70	16.06	22.90
21	6.27	22.90	13.76	68.70
22	9.48	137.40	11.05	137.40
23	11.43	0	9.80	91.60
24	9.74	338.17	12.87	160.30

Conclusions for Health and Growth of ViaGen Clones. The results of this study are limited by the small number of animals and the design of the experiment. However, results of this experiment indicate that even though the clone barrows were subjected to a significant immunological challenge after moving from more biosecure conditions to more standard housing conditions, most clones were able to respond appropriately to this stress. This immunological challenge potentially slowed the growth of clones early in this experiment and thus may have resulted in a delay in their maturation.

c. Carcass Characteristics

Carcass characteristics were provided on the four Hamline clones and 15 comparator barrows followed in Study 1; these are summarized in Table F-7. Hot carcass weights¹⁴¹ averaged 85.91 and 90.68 kg for clone and comparator barrows, respectively, with the exception of the animals that were excluded (see discussion in next paragraph). Carcass lengths were 82.4 and 84.5 cm for clones and comparators, respectively. Dressing percentages were 70.1 and 70.2 percent for clones and comparators, and were similar across breed groups. Backfat thickness over the first rib, tenth rib, last rib, and lumbar vertebra were slightly greater for comparators than clones which may, in part, be due to the heavier body weight of comparator barrows at the time of slaughter. Qualitative characteristics including USDA carcass muscle score, firmness, and marbling were similar across groups and are illustrated in Table F-7. All animals received score 2 for carcass muscle. All of the clone and comparator barrows had marbling scores of either 1 or 2.

¹⁴¹ Hot carcass weight refers to the weight of the eviscerated carcass post exsanguination, but prior to chilling.

The Duroc clone barrow carcass was condemned at slaughter due to a lung adhesion, and thus data relating to growth and carcass characteristics from this animal were not included for these variables. The rationale for excluding the carcass for this analysis is that in some cases, lung adhesions are due to bacterial infection, and animals fail to thrive, (i.e., their growth and carcass characteristics are adversely affected). Two other clones (#21 and 23) were approximately 45.45 kg lighter than any of the other animals in the experiment at the time of slaughter, and for this reason were excluded from carcass evaluation.

Measurements of pH at 24 hours post-slaughter on the *longissimus dorsi* muscle were similar. Loin eye area for meat cuts for clones and comparators were quite similar at 116.33 ± 10.16 and 11.76 ± 11.18 cm, respectively.

Table F-7: Comparison of the Carcass Characteristics of Barrows Derived by Somatic Cell Nuclear Transfer (Clones) or Conventional Breeding (Mean \pm standard deviation)		
	Comparators (n=15)	Clones (n=4)
Hot Carcass Weight (kg)	90.5 \pm 6.2	85.7 \pm 6.26
Carcass Length (cm)	84.5 \pm 2.7	82.4 \pm 1.5
Dressing Percentage (%)	70.2 \pm 1.4	70.1 \pm 0.8
Back Fat Thickness (mm)		
First Rib	38.7 \pm 3.1 ^a	35.3 \pm 2.1 ^b
Tenth Rib	22.2 \pm 4.9	18.5 \pm 3.1
Last Rib	23.3 \pm 3.4	20.5 \pm 4.7
Last Lumbar	21.0 \pm 3.1	17.3 \pm 3.2
Loin Eye Area (cm ²)	45.8 \pm 4.0	44.0 \pm 4.4
Longissimus pH at 24 hours	5.7 \pm 0.1	5.6 \pm 0.1
Carcass Muscle Score	2.2 \pm 0.4	2.0 \pm 0.0
NPPC Quality Scores		
Color	2.7 \pm 0.6	3.0 \pm 0.0
Marbling	1.9 \pm 0.5	1.5 \pm 0.6
Firmness	2.9 \pm 0.9	3.5 \pm 0.6
^{a, b} Values with different superscripts are significantly different (P<0.05)		

d. Meat Composition Analysis for Clones and Comparators

Meat composition data were available for the five clones (four Hamline and one Duroc) and 15 comparator animals (all Hamline). Because there were no differences between the Duroc and Hamline clones, data were pooled. All of the values examined (amino acids, fatty acids,

cholesterol, nutritionally important vitamins and minerals (see discussion in Chapter 6) were remarkably similar, and no biologically relevant differences were noted (Table F-8).

Table F-8: Comparison of Nutrient Concentrations in Meat from Swine Clones and Comparators from Experiment 1¹		
Component	Clones mean \pm std. dev.	Comparators mean \pm std. dev.
Amino acids (g)		
Alanine	1.26 \pm 0.04	1.30 \pm 0.04
Arginine	1.41 \pm 0.03	1.47 \pm 0.04
Aspartate	2.55 \pm 0.28	2.43 \pm 0.19
Cystine	0.25 \pm 0.03	0.26 \pm 0.02
Glutamate	3.41 \pm 0.11	3.46 \pm 0.09
Glycine	0.98 \pm 0.04	1.02 \pm 0.10
Histidine	0.99 \pm 0.05	1.03 \pm 0.05
Isoleucine	1.04 \pm 0.05	1.05 \pm 0.03
Leucine	1.74 \pm 0.05	1.79 \pm 0.04
Lysine	1.91 \pm 0.06	1.96 \pm 0.04
Methionine	0.54 \pm 0.06	0.58 \pm 0.03
Phenylalanine	0.86 \pm 0.02	0.89 \pm 0.02
Proline	0.85 \pm 0.03	0.90 \pm 0.06
Serine	0.90 \pm 0.03	0.92 \pm 0.02
Threonine	1.11 \pm 0.04	1.14 \pm 0.03
Tyrosine	0.77 \pm 0.02	0.79 \pm 0.02
Valine	1.10 \pm 0.05	1.12 \pm 0.04
Fatty Acids² (g) and Cholesterol (mg)		
14:0 (Myristic acid)	0.09 \pm 0.06	0.05 \pm 0.03
16:0 (Palmitic acid)	1.31 \pm 0.82	0.95 \pm 0.49
16:1 (Palmitoleic acid)	0.09 \pm 0.04	0.14 \pm 0.05
17:0 (Margaric acid)	0.01 \pm 0.01	0.00 \pm 0.01
17:1 (Margaroleic acid)	0.01 \pm 0.01	0.00 \pm 0.01
18:0 (Stearic acid)	0.66 \pm 0.41	0.55 \pm 0.27
18:1 (Oleic acid)	1.84 \pm 0.84	1.49 \pm 0.50
18:2 (Linoleic acid)	0.26 \pm 0.08	0.19 \pm 0.06
18:3 (Linolenic acid)	0.01 \pm 0.01	0.00 \pm 0.01
20:0 (Arachidic acid)	0.01 \pm 0.01	0.00 \pm 0.01
20:1 (Gadoleic acid)	0.05 \pm 0.03	0.04 \pm 0.02
20:2 (Eicosadienoic acid)	0.01 \pm 0.01	0.01 \pm 0.01
22:6 (Docosahexaenoic acid)	0.02 \pm 0.03	0.01 \pm 0.01

Cholesterol (mg)	55.5 ± 6.95	52.81 ± 2.69
Component	Clones mean ± std. dev.	Comparators mean ± std. dev.
Minerals (g)		
Calcium	0.004 ± 0.000 ³	0.005 ± 0.003
Phosphorus	0.20 ± 0.01	0.21 ± 0.01
Iron	0.001 ± 0.000 ³	0.001 ± 0.001
Zinc	0.002 ± 0.000 ³	0.001 ± 0.000 ³
Vitamins		
Niacin (mg)	10.90 ± 0.83	11.16 ± 1.58
B ₆ (mg)	0.41 ± 0.09	0.48 ± 0.12
B ₁₂ (mcg)	0.21 ± 0.28	ND ⁴
¹ Data expressed as quantities per 100 g of homogenized meat. ² Data presented reflect those fatty acids with detectable levels in pork. ³ Actual standard deviation was < 0.001 ⁴ ND = not detected		

Conclusions for Carcass Characteristics and Meat Composition of Clones. No remarkable differences were observed between clones and comparators for any of the characteristics evaluated. The small differences in backfat thickness and marbling are likely due to the lighter weight of clones vs. comparators at slaughter. Differences in meat nutrient composition were very small and likely not biologically relevant. The lack of biologically relevant differences in the food composition values between muscle of swine clones and comparators supports the conclusion that there is no additional food consumption risk from the consumption of muscle from swine clones compared to their conventional counterparts.

e. Semen and Breeding Evaluation

Sperm concentration, percent total motility (percent of sperm in motion), percent progressive motility (evaluates forward movement), and number of head/tail abnormalities were similar for the four clones and the comparator boars. The clones did not appear to have a reduction in semen quality relative to the comparator boars. The lowest post-shipment semen concentration, percent motility, and percent progressive motility were observed in the Duroc line comparator boars.

Animal	Concentration (sperm/ml x 10⁹)	Total Motility	Progressive Motility	Head Abnormalities	Tail Abnormalities
Clone 2	5.13	82.34%	71.39%	58.00%	90.00%
Clone 3	7.10	79.95%	71.92%	81.75%	91.00%
Clone 5	5.23	86.18%	75.75%	58.50%	87.50%
Clone 7	4.19	79.19%	62.68%	90.50%	79.50%
Control H498	4.51	75.86%	70.45%	66.33%	87.22%
Control 18128	2.53	53.28%	49.42%	92.50%	81.25%
Control 25515	3.19	77.14%	57.20%	97.25%	83.75%

f. Farrowing Rate

The farrowing rate for the gilts inseminated with semen from clones and comparators was 73.5 percent vs. 62.5 percent, respectively. Both these rates are below the industry average of approximately 80 percent. The PigCHAMP swine industry record keeping system farrowing rate average for the second quarter of 2007 was 78.9 percent¹⁴². It should be noted that the PigCHAMP rate included all parities (the number of times a sow has farrowed), and the rate would be expected to be lower for gilts. The lower farrowing rates of the pigs in this study could be attributed to many factors including the feed intake of the gilts as well as the ability of technicians to detect gilts in heat. Farrowing rate is generally more attributable to the female as opposed to the male member of the breeding pair, and takes into account such factors as ovulation rate (number of oocytes released per estrus), age/parity of the sow, and environmental factors (Hafez and Hafez 2000). Additionally, the rate for the comparator-bred group was low relative to the clone-bred group, which was largely attributable to the gilts bred by one boar whose farrowing rate was 41.7 percent. This boar (Hamline nuclear donor) was reported to be five years old at the time of semen collection (relatively old for a breeding boar), which may have contributed to the low farrowing rate for gilts inseminated by his semen. As the clone-bred group had a higher farrowing rate than the comparator-bred group, and the farrowing rate of the clone-bred group was only slightly lower than industry averages, cloning does not appear to negatively impact AI-based boar reproductive performance.

The mean litter size for the progeny of the clone boars was 10.94 pigs and the median was 11.5 pigs. For the pigs from the comparator boars, the mean litter size was 11.76 pigs and the median was 12.0 pigs. The mean litter size for the U.S. in the second quarter of 2007 according to

¹⁴²http://www.pigchamp.com/benchmarking_summaries.html

PigCHAMP records was 11.8 pigs per litter¹⁷. Although the comparator boars had a higher mean litter size, both groups were near the U.S. average for litter size. Litter size appeared to be more variable in gilts inseminated by clones: 11 percent of the litters from the clone boars had more than 14 pigs, whereas none of the comparator litters had more than 14 pigs. Further analysis of litter size showed that the clone boars also had a higher frequency of litters with less than ten pigs. Approximately 27.8 percent (10/36) of the litters from the clone boars had fewer than ten pigs in the litter. For the comparator group, only one litter (out of 25 litters or 4 percent of all comparator litters) had fewer than ten pigs. Many factors can affect litter size in pigs including the estrus cycle in which the gilts are bred, genetics, nutrition, management, environment, and ovulation rate of the gilt (Aherne and Kirkwood 1985). Although many of these factors were controlled (nutrition, parity, management, and environment), other factors besides cloning cannot be ruled out as contributors to the wider distribution of litter size for the litters from the clones. Finally, as semen characteristics appear to be similar between the clones and their comparators, the differences in litter sizes were most likely due to gilt or uncontrollable management variation such as breeding cycle or feed intake.

Table F-10: Results of Breeding Boar Clones and Comparators

Boar	Number of Gilts Bred	Number of Gilts Pregnant Day 30	Percentage of Gilts Pregnant Day 30	Number of Gilts Pregnant Day 110	Percentage of Gilts Pregnant Day 110	Number of Gilts Farrowed	Percentage of Gilts Farrowed
Clone 2	12	9	75	7	58.33	7	58.33
Clone 3	12	10	83.33	10	83.33	10	83.33
Clone 5	12	9	75	8	66.67	8	66.67
Clone 7	13	11	84.62	11	84.62	11	84.62
Control H498	12	7	58.33	7	58.33	5	41.67
Control 18128	14	13	92.86	13	92.86	11	78.57
Control 25515	14	9	64.29	9	64.29	9	64.29

Conclusions for Reproductive Performance of Clones. There were no apparent differences in semen quality between clones and comparator boars. Farrowing rate was slightly higher among gilts bred by semen from clone boars, although the difference between clones and comparators could be traced to the Hamline nuclear donor, which was older than any of the other boars used in this study. Average litter size was similar for clones and comparator boars, and farrowing rate and litter size for clones were similar to national averages. Therefore, reproductive performance does not appear to be affected by the cloning process in these animals.

2. Study 2: Progeny of Clones vs. Comparators

a. Survival

A summary of the survival data from this study is presented in Table F-11. At the start of the study, there were 310 (295 made it to slaughter) live-born clone-derived progeny and 251 (243 made it to slaughter) live-born comparator-derived progeny. The percentage of mummified pigs (dead, desiccated fetuses) at farrowing was 3.3, 2.8, 1.7, and 0 percent for the progeny from Hamline comparator, Hamline clones, Duroc comparators, and Duroc clone boars, respectively. In both breeds of pigs (Hamline and Duroc) the percentage of mummified pigs was slightly higher in the comparator group than in the clone-derived pregnancies. In this study, litters from Hamline clones and comparators and Duroc comparators had higher rates of mummies in their litters than the U.S. average (0.2 percent) (SwineReproNet)¹⁴³; however, the percentage of mummies in the litters derived from the Duroc clone was similar to the U.S. average. A substantial number of pigs were lost around the time of birth, ranging from 17.0 percent-31.4 percent, and in each breed, these losses were slightly higher in the group comprised of progeny derived from clones. Most of these losses were due to the categories of “weakness” or “unknown causes.” Further analysis of the data indicated that an entire litter of 13 progeny from a Duroc clone boar was lost on July 15, 2004, shortly after birth. Reasons provided for the loss of this litter included “unknown” (n=7) and “weak” (n=6). The accompanying animal health records note, however, that between July 13 and 15 sows were stressed due to both high temperature and humidity in the farrowing house. The records also indicate that C- section was performed on one of the heat-stressed gilts, and the gilt and her 13 pigs subsequently died. If these 13 progeny from the one litter that died are removed from the evaluation, the differences in survival rate for progeny from clones and from comparator boars are slight and inconsequential.

¹⁴³ <http://www.triail.uiuc.edu/swinerepronet/paperDisplay.cfm?ContentID=6266>

Table F-11: Comparison of the Survival Data for Progeny of Clone or Comparator Boars					
	Hampshire Comparator	Hampshire Clone	Duroc Comparator	Duroc Clone	Total
Total pigs born	61	284	239	118	702
Mummies	2 (3.3%) ¹	8 (2.8%)	4 (1.7%)	0 (0%)	14 (2.0%)
Died at Birth	11 (18.6%)	62 (22.5%)	40 (17.0%)	37 (31.4%)	150 (21.8%)
Stillborn	5 (8.5%)	21 (7.6%)	22 (9.4%)	15 (12.7%)	63 (9.2%)
Overlay	5 (8.5%)	27 (9.8%)	10 (4.3%)	6 (5.1%)	48 (7.0%)
Weak	0 (0%)	1 (0.4%)	0 (0%)	6 (5.1%)	7 (1.0%)
Unknown	0 (0%)	12 (4.3%)	8 (3.4%)	10 (8.5%)	30 (4.4%)
DDFR	0 (0%)	1 (0.4%)	0 (0%)	0 (0%)	1 (0.1%)
DINJ	1 (1.7%)	0 (0%)	0 (0%)	0 (0%)	1 (0.1%)
Number of Animals Weaned	48 (81.4%)	214 (77.5%)	195 (83.0%)	81 (68.6%)	538 (78.2%)
Died Prior to Slaughter	0 (0%)	5 (1.8%)	7 (3.0%)	0 (0%)	12 (1.9%)
Completed Experiment	48 (81.4%)	209 (75.7%)	188 (80.0%)	81 (68.6%)	526 (76.5%)
Slaughtered	44 (74.6%)	181 (65.6%)	119 (50.6%)	61 (51.7%)	405 (58.9%)
Comparator shipped to market	0 (0%)	0 (0%)	64 (27.2%)	0 (0%)	64 (9.3%)
Tanked at slaughter ²	0 (0%)	21 (7.6%)	0 (0%)	14 (11.9%)	35 (5.1%)
Shipped to Perdue ³	4 (6.8%)	7 (2.5%)	5 (2.1%)	6 (5.1%)	22 (3.2%)
¹ Percentages are of total animals per group.					
² These animals were sent for rendering because there were more progeny born than was needed for the experiment.					
³ These animals were sent at the conclusion of the experiment to Perdue University to be enrolled in an immunology experiment.					

The percentage of stillborns among progeny of clones (36/394) and the progeny of the comparator boars (27/294) was the same (9 percent). This level is within the estimates of industry averages for stillborns (range 5 -10 percent (SwineReproNet)). Sixteen of the 36 litters from the clone boars had at least one stillborn pig (44 percent), while 13 of the 25 litters from the comparator boars had at least one stillborn pig (53 percent). Therefore, the frequency of stillborns in litters from the clone boars was lower than the comparator group. The litters of the clone Hamline boars had an average of 0.8 ± 0.3 rate of stillborn per litter, and were virtually identical to the comparator Hamline boars (0.8 ± 0.07). Similarly, the stillborn rate for the Duroc clone boar litters was 1.4 ± 0.4 per litter, while the rate for the comparator Duroc boar litters was 1.1 ± 0.3 stillborns per litter. According to PigCHAMP, these rates are similar to the average stillborn rate of 0.90 pigs per litter for the U.S. swine industry records for the second quarter of

2007¹⁷. Therefore, cloning does not appear to affect the stillborn rate component of reproductive performance of pigs for either genetic line.

One clone progeny pig was destroyed due to a deformity (DDFR). One comparator pig was destroyed due to an injury (DINJ). The destroyed pigs represent 0.25 percent and 0.3 percent for the progeny of the clone and comparator boars, respectively. The deformity of the destroyed clone progeny was not described and may more accurately fit into the “other abnormalities” category. Adding an additional pig to this category for the progeny of the clone boars does not increase the incidence rate to a level that warrants concern (see discussion on abnormalities). Injuries during nursing are usually due to sow overlays. Consequently, the injured pig reported in its own category may typically be included in the overlay category as discussed below. Adding this pig to the overlay category does not substantially change the frequency of overlays and therefore does not warrant changing the conclusion relative to overlays as discussed below.

The rates of abnormalities in both clone and comparator derived progeny were similar to industry observed levels. Progeny of the clone boars had an abnormality rate of 2.5 percent (10/394), including anal atresia (lack of opening of the anus) (1/394, 0.3 percent), spraddle legs (leg weakness) (4/394, 1 percent), and “other” (5/394, 1.2 percent). Three of the 295 offspring of the comparator boars had abnormalities (1 percent), all of which were recorded as having spraddle legs. The incidence and cause(s) of spraddle legs is not well documented or fully understood, but it may involve several factors including genetics, management, slick flooring, mycotoxins, and a virus or combination of viruses (DeRouchey et al. 2007). Furthermore, the incidence of spraddle legs was the same for both groups. Therefore, the occurrence of spraddle legs does not appear to be related to cloning. Of the two other classes of abnormalities observed in the progeny of clones, all have previously been reported in the literature on swine reproduction. The incidence of anal atresia in the general swine population has been reported to range from 0.1-1.0 percent (Wiedemann et al. 2005). Further investigation of the “other” category revealed that these pigs were from two litters. Three pigs from one litter were described as having “typical leg abnormalities possibly associated with overcrowding in the uterus.” These pigs were from a litter of 18 pigs, which would be considered large compared to industry average (Vonnahme et al. 2002). The other two pigs were from another litter, and were described as having short legs. The frequency of miscellaneous abnormalities in newborn pigs has been reported by Spicer and coworkers (1986) as 1.2 percent, and included cleft palate, anal atresia, renal hypoplasia, hydrocephalus and accidental death. Because similar abnormalities have been reported in the swine industry at a similar frequency to that of this study, the rate of “other” abnormalities in this study is not a high concern.

Seven of the 394 progeny (2 percent), all from the same litter (#339) derived from one of the clone boars were disposed of for unknown reasons. Furthermore, of the seven clone-progeny pigs that were categorized as “disposed of because they were weak” (2 percent), six were from the same litter (#339) as the pigs “disposed of for unknown reasons.” Only one other progeny of the clone boars was disposed of for being weak. No comparator pigs were disposed of for unknown reasons or because of weakness. The fact that 4 percent of the pigs from the clone boars were destroyed due to weakness and unknown reasons compared to 0 percent of comparators could be a matter for concern. However, this effect results primarily from observations from one litter, and the daily observation records indicate that heat stress was a problem in the farrowing house when this litter was delivered. Further, given most of the pigs in this litter were disposed on the day they were born, with only two pigs living for three days, it appears that heat stress of the dam and/or pigs may have contributed to the loss of this litter. The consequent disposal of this litter resulted in the relatively high rate (4 percent) of “unknown” and “weak pig” disposals for the progeny of the clone boars. The data do not, however, fully account for the removal and subsequent care of the pigs in this litter, and this interpretation should be considered preliminary. We also note that unknown disposals or weak pig disposals do not appear to be a problem across litters for the clone progeny group, which would be expected if cloning were a primary contributing factor to the incidence of weak pigs in the progeny of clones.

The frequency of overlays (death due to the sow lying on top of the piglets) for pigs in litters from the clone boars and the litters from the comparator boars were 8.2 percent and 5.0 percent respectively. Industry estimates for pig deaths due to crushing by the sow are between 4.8 and 18 percent (Lay et al. 1999). The frequencies of overlays for both groups in this study are near the low end of the estimated range of deaths due to crushing. Crushing has been reported to be related to several factors including the genetics, activity level of the sow and sow housing (Lay et al. 2002). Secondary factors that potentially influenced the number of pigs that die due to the dam lying on pigs included environmental conditions at the time of farrowing and the number of litters that were born on a single day. The data indicate that 315 of the 688 pigs were born on one of six days where the heat index was above 104°F (40°C). The daily sow/litter observation records indicate that high temperatures in conjunction with poor cooling may have contributed to the number of crushing deaths, as the sows attempted to find a cooler and more comfortable position. The records do not differentiate between the two groups of gilts, and gilt housing was the same for both groups in this study. Therefore, any differences in the number of crushing deaths in this study were probably due to differences in sow activity. Also, the litter observation records indicate normal growth and behavior for all of the pigs and therefore, there is no evidence to indicate an increase in susceptibility to crushing of pigs in the litters from the clone boars.

The total number of disposed pigs (stillborns, destroyed, overlays, unknown deaths and weak pigs) was 21 percent for the pigs from the clone boars and 14 percent for the pigs from the comparator boars. The difference in the two groups is primarily due to the unknown and weak pig disposals and a higher rate of overlays. These categories have been discussed previously. Also, there was a 3.0 percent loss of pigs post-weaning in pigs derived from Duroc comparators and a 1.8 percent loss of pigs obtained from Hampshire clone boars. Based on these data there is no evidence to suggest that progeny of clones are at increased risk for mortality compared to AI-derived pigs.

Initial plotting of the relationship between age and body weight suggested that, although the mean length of time to slaughter was similar for progeny within the four groups, there might be a broader range in the length of time that it took to reach slaughter weight. These data indicate that there was a similar range, from 144 to 210 days, in the length of time that it took for progeny from conventionally bred and clone boars to attain their slaughter weight. Furthermore, the data indicate that there were only small differences in ADG between the four groups of pigs between birth and slaughter or weaning and slaughter. When ADG was calculated over 4 week intervals there were no significant differences prior to 20 weeks of age. The data indicated that slightly less than 50 percent of the progeny from each of the four groups was slaughtered prior to week 24 and that only 27 animals remained on the experiment at week 28. The small differences in percentage of animals could be attributed to the small number of animals remaining in the experiment at this point than to any one of the groups.

Because of its retrospective survey nature, and its smaller size, the Cyagra dataset was evaluated using a slightly different procedure (see description in Appendix E). In that case, values from approximately breed- and age-matched comparators were used to establish a range against which values obtained from clones were compared (The Cyagra approach). To determine how outcomes would differ if the ViaGen data had been analyzed by the same method used to evaluate the Cyagra dataset, both approaches were applied to the clinical chemistry and hematology variables from ViaGen Study 2. The following tables (Tables F-12a, F-12b and F-13) summarize the proportion of values that were identified as outliers for further examination by both procedures. The boxplot procedure identified a slightly higher percentage of values for further examination than the procedure used to describe the Cyagra data. However, the boxplots identified similar percentages of outlier values for progeny of comparators as for progeny of clones.

Table F-12a: Variables, Observations, and Outlier Values of Bloodwork from Progeny of Comparators Using the Box Plot Method

Data	Number of Variables	Number of Observations	Number of Progeny	Number of Outlier Values	Number of Missing Values	Percentage of Outliers	Percentage in Range
July 2004 Hematology	18	439	204	201	0	5.5	94.5
July 2004 Chemistry	35	513	226	267	0	3.4	96.6
October 2004 Hematology	18	452	187	108	2	3.2	96.8
October 2004 Chemistry	35	442	178	295	0	4.7	95.3
January 2004 Hematology	18	222	84	52	10	3.5	96.5
January 2004 Chemistry	35	405	164	156	0	2.7	97.3

To derive percentage of outlier values in Tables F-12a, F-12b, and F-13, multiply the number of variables by number of progeny. For example, from row 1 of Table F-12a: 18 variables x 235 clone progeny sampled = 4230 total observations for July hematology. Then divide the number of outliers in the appropriate column by total observations ($167/4230 = .0394$) and multiply by 100 to obtain the percentage of outlier values.

Table F-12b: Variables, Observations, and Outlier Values of Bloodwork from Progeny of Clones Using the Box Plot Method

Data	Number of Variables	Number of Observations	Number of Progeny	Number of Outlier Values	Number of Missing Values	Percentage of Outliers	Percentage in Range
July 2004 Hematology	18	439	235	167	0	3.9	96.1
July 2004 Chemistry	35	513	287	320	0	3.2	96.8
October 2004 Hematology	18	452	265	151	28	3.2	96.8
October 2004 Chemistry	35	442	264	295	0	3.2	96.8
January 2004 Hematology	18	222	138	122	16	4.9	95.1
January 2004 Chemistry	35	405	241	365	0	4.3	95.7

Data	Number of Variables	Number of Observations	Number of Progeny	Number of Outlier Values	Number of Missing Values	Percentage of Outliers	Percentage in Range
July Hematology	18	439	235	12	0	0.3%	99.7%
July Chemistry	35	513	287	53	0	0.5%	99.5%
October Hematology	18	452	265	36	28	0.8%	99.2%
October Chemistry	35	442	264	62	0	0.7%	99.3%
January Hematology	18	222	138	47	16	1.9%	98.1%
January Chemistry	35	405	241	85	0	1.0%	99.0%

b. Growth, Hematology, Clinical Chemistry, and Urinalysis

i. Growth

The birth weights of clone and comparator progeny were similar. To establish a base population for the comparator group of pigs ($n = 267$), mummified and stillborn pigs were excluded. A birth weight outlier analysis of the progeny of the clones and comparator boars ($n=617$) indicated that two progeny of clone boars were lightweight outliers (0.59 and 0.64 kg). One comparator pig was a lightweight outlier (0.41 kg) and one comparator pig was a heavyweight outlier (2.31 kg). The boxplots for birth weights showed a similar distribution for the progeny of the clones and comparator boars. Additionally, the mean birth weight for the offspring of the clone boars was 1.5 kg and the mean for the comparator group was 1.45 kg. The median for both groups was 1.5 kg. The similarity in birth weights, the birth weight distributions and the low frequency of outliers between the progeny of the clone boars and those of the comparator boars indicate that birth weight is not a health concern for progeny of clones.

Sire	Number of pigs	Birth weight (kg)
Clone 2	78	1.55
Clone 3	124	1.43
Clone 5	75	1.54
Clone 7	118	1.52
Control H498	61	1.52
Control 25515	105	1.48
Control 18128	134	1.41

The progeny from both clones and comparator boars had similar nipple counts with similar distributions of the counts. Nipple counts are important because they are genetically transmitted and indicative of the number of offspring a female can feed. Most of the pigs in this study had between 12 and 16 nipples (95.7 percent and 98.4 percent for the clones (389/394) and comparators (282/294), respectively). The industry standard is for at least six functional teats per side (total 12) of the underline of a gilt (Ahlschwede and Kuhlers 1992). The progeny of the clone boar group had no pigs with fewer than 11 nipples while the progeny of the comparator group had three pigs with 10 or fewer nipples. Nipple counts for the progeny of clone boars are therefore within normal bounds of these pig populations.

Growth characteristics of these animals were also analyzed, with the data evaluated for potential outliers prior to statistical analysis or plotting. Forty-two outliers were identified in the dataset containing body weight measurements (n =2,966). Six of these outliers represented body weights taken prior to the death of 4 animals. Nine of the outliers were data points associated with animals derived from both clone and comparator sires that lost weight near the end of the experiment. No reason for the weight loss was provided, and these animals were excluded from the final analysis. The mean slaughter weights were, 121.7, 119.6, 120.2, and 121.9 kg for progeny derived from the Hampshire comparators, Hampshire clones, Duroc comparators, and Duroc clones, respectively. Body weights at the time of slaughter ranged from 108.8 to 134.7 kg for progeny from the Hampshire comparator boar, 97.4 to 135.7 kg for progeny from Hampshire clones, 97.0 to 136.2 kg for progeny from Duroc comparator boars, and 108.0 to 137.0 kg for progeny from the Duroc clone boar. The mean number of days from birth to slaughter was 173.7, 174.7, 168.2, 175.5 days for progeny obtained from Hampshire comparator, Hampshire clones, Duroc comparators, and the Duroc clone, respectively.

Tabulation of the calculated average daily gain (ADG) for progeny from clones and comparators at various time points throughout their lives is shown in Table F-15. Average daily gains from

birth to slaughter were 0.69, 0.68, 0.71, and 0.69 kg/day for progeny derived from Hampshire comparator, Hampshire clones, Duroc comparators, and Duroc clone boars, respectively. Similar finds were observed for ADG between weaning and slaughter (0.76, 0.73, 0.77, and 0.74 kg/day, respectively). Body weights were also measured every 4 weeks with only small differences between the progeny of clones and comparators.

	Hampshire Comparator	Hampshire Clone	Duroc Comparator	Duroc Clone
Birth to weaning	0.40±0.13 (n=48)	0.44±0.09 (n=208)	0.43±0.10 (n=186)	0.45±0.09 (n=81)
Weaning to 8 wks	0.65±0.13 (n=48)	0.73±0.15 (n=208)	0.82±0.14 (n=186)	0.84±0.14 (n=81)
8 wks to 12 wks	1.59±0.20 (n=48)	1.51±0.21 (n=208)	1.54±0.18 (n=186)	1.55±0.22 (n=81)
12 wks to 16 wks	2.04±0.30 (n=48)	1.96±0.25 (n=208)	2.07±0.28 (n=186)	2.01±0.30 (n=81)
16 wks to 20 wks	2.12±0.35 (n=48)	2.10±0.32 (n=208)	2.18±0.31 (n=186)	1.92±0.30 (n=81)
20 wks to 24 wks	2.11±0.29 (n=24)	2.00±0.38 (n=109)	2.05±0.32 (n=73)	1.98±0.35 (n=54)
24 wks to 28 wks	1.81±0.32 (n=4)	1.77±0.52 (n=11)	0.84±1.88 (n=5)	1.86±0.41 (n=7)
Birth to slaughter	1.53±0.11 (n=48)	1.50±0.11 (n=208)	1.57±0.10 (n=186)	1.52±0.10 (n=81)
Weaning to slaughter	1.67±0.12 (n=48)	1.62±0.13 (n=208)	1.70±0.12 (n=186)	1.63±0.11 (n=81)

ii. Hematology, Clinical Chemistry, and Urinalysis

*First Blood Sampling*¹⁴⁴ (July). The results of blood clinical chemistry and hematology for July, when the progeny were between three and 30 days old, are in Charts F-1 and F-2, respectively. More than 94 percent of these values showed no differences between the progeny of clones and comparators. We identified the following variables to be of interest for clone progeny:

¹⁴⁴ Appendix H has detailed descriptions of clinical chemistry values and what they measure.

hemolysis, lipemia, percent saturation, ALT, AST, CK, Lipase, SDH, LUC, MCHC, and MPV. This is because greater than 5 percent of the clone values were outside the comparator range and therefore would be more than we would expect by chance. In approximately 5 percent of the clone progeny, values for hemolysis, lipemia, percent saturation, ALT, AST, CK, Lipase, SDH, LUC, MCHC, and MPV were outside the comparator range. To determine whether these values indicated concerns for the health of the animals, we compared the amount of variability between the clone progeny and the comparator progeny groups. Following that analysis, we determined that the values for hemolysis, percent saturation, ALT, SDH, LUC, MCHC, and MPV had a similar amount of variability and did not warrant any further concern. The values for lipemia, AST, CK, and lipase were out of range more frequently for clone progeny and required further consideration.

Hemolysis and lipemia can be considered artifacts based on sample handling or drawing samples from animals that have been fed recently, respectively, and can have a significant adverse impact on the quality of other blood data values (Duncan and Prasse 2003). Hemolysis, either from poor collection technique, age of the sample, or poor handling of the sample once in the laboratory, has a serious effect on many blood chemistry tests, including dramatic effects on the enzymes alkaline phosphatase, GGT, and CK. In general, it is advisable to draw a new sample if gross hemolysis is noted. Lipemia (a measure of the amount of fats in the blood) can become elevated if animals have their blood drawn shortly after eating. The increased levels of lipids can falsely raise Na, K, and Cl levels and artifactually lower AST and ALT levels (Shanahan 2004). For that reason, lipemia is not considered a health related variable in food animals (Duncan and Prasse 2003). Creatine kinase (CK) is an enzyme found in muscle tissue and to a lesser extent in liver cells, and elevations are often indicators of muscle injury (including muscle damage during venipuncture) or hemolysis (Duncan and Prasse 2003) (See Appendix E). It is unclear if the small elevations that were noted were due to those reasons, or injury near the time of handling. AST is an enzyme found in liver cells and muscle cells. It has a longer half-life than CK. Although eight more clone progeny (19/242 or 7.8 percent) had elevated AST compared to the non-clone progeny (11/163 or 6.8 percent), neither the level of increase nor the number of animals with increased levels were sufficiently high to indicate a real biological difference. Lipase is a pancreatic enzyme that breaks down fat; its elevation can indicate pancreatic inflammation (pancreatitis). A 2-3 fold increase, however, is considered the threshold for further evaluation to determine whether pancreatic inflammation is responsible. (Duncan and Prasse 2003). The upper level is 93.5 U/l and average elevation is 132 U/l. In pancreatitis, there also is usually an increase in another enzyme (amylase) to corroborate the condition (Duncan and Prasse 2003). Amylase values in this study are within range. As values in this study represented less than a 2 fold increase, we did not consider them biologically relevant. We therefore

concluded that there are no biologically relevant differences in blood values between clone progeny and comparator progeny at this point in their development.

Second blood sampling (October). The results of blood clinical chemistry and hematology for October, when the animals were approximately 12 to 15 weeks old, are in Chart F-3 and F-4, respectively. In comparing the clone to non-clone progeny variability, the CK and basophil values were similar enough to conclude no difference. For the chemistry values in clones, indirect bilirubin/total bilirubin and bile acids had more outliers compared to non-clone progeny. Bilirubin is a breakdown product from the hemoglobin of senescent (old) RBCs. The liver processes this by conjugating the product to a salt and making it water soluble. Elevations in bilirubin can indicate reduced hepatic function (Duncan and Prasse 2003). The clones in this study had low bilirubin. There is no known cause for low bilirubin. For this reason, this finding was considered not clinically relevant. Bile acids were elevated in 25 clone progeny and 13 comparator progeny. Bile acids can be artifactually elevated in response to eating. This value may also indicate some hepatic insufficiency (insufficient number of liver cells to perform the metabolic functions of the liver). If the liver were adversely affected, we would expect to find other corroborating analytes to confirm this possibility. There are no other analytes to confirm hepatic insufficiency in these animals.

Hematology values for basophils for progeny of clones and comparators were similar enough not to warrant further discussion. More clone progeny had lower MCH (mean corpuscular hemoglobin) and MCV (mean corpuscular volume) values and more clone progeny had higher RBC values than the comparators. RBCs are elevated in 12 clone progeny and only 2 comparator progeny. Elevations in RBC can be from excitement (splenic contraction), hydration status (dehydration causes an increase in RBC), or an absolute polycythemia (true increase in production). The cause here is unknown but does not seem to indicate a health problem. MCH is a value derived by dividing hemoglobin by the RBC number. Because the RBC number is high, the MCH must be low. A decrease in mean MCV can mean an iron deficiency. This is usually accompanied by anemia. Anemia is defined as a reduced number of RBCs or decreased Hematocrit/ Packed Cell Volume (PCV). Because there was an increase in these values, its significance is minimal.

Third blood sampling (January). The results of blood clinical chemistry and hematology for January, when the animals were approximately 24 weeks old, are in Charts F-5 and F-6, respectively. Values with similar variation between clone and comparator progeny are Mg⁺⁺, LUC, and RDW. These values require no further discussion.

Sodium:potassium (Na:K) ratio is a value derived from the sodium concentration and compared to the potassium concentration. Neither Na⁺ nor K⁺ were significantly different. Their ratio is used to determine adrenal function (to detect Addison's Disease) in small animals. Its significance in pigs is not listed in clinical pathology texts as being clinically relevant (Duncan and Prasse 2003). ALT, AST, BA, CK, and SDH are analytes with significance for liver and muscle tissues. Because we have seen elevations in these enzymes before and discussed them above, we decided to determine if there was clinical relevance to the increase in clone progeny. As discussed, BA may be increased depending on when the blood was drawn in relation to a meal. Elevations in values for analytes with significance for liver and their effect on body weight are discussed in the next section. Daily health observations were not available. As stated in Appendix E and elsewhere, one can only evaluate lab tests in the context of a complete clinical picture.

In the January hematology, hematocrit and RBCs were elevated in progeny of clones. Elevated hematocrit and RBC values are rarely an adverse health issue. The MCHC (mean corpuscular hemoglobin concentration) had as many high values as low values, which reduces its significance as an indicator of a health problem. The clone progeny had 21/138 (15.2 percent) animals with elevated segmented neutrophils (segs) vs. 4/84 (4.8 percent) for the comparator progeny. Segmented neutrophils are elevated in response to bacterial exposure. With no daily clinical health observations, it is difficult to interpret this observation. However, this may be an appropriate response to some challenge in the pig's environment because no other analytes indicative of active infection (over all white cell count, banded neutrophils, globulin) are elevated.

No differences in the levels of IGF-I (Fig.1), and E2 (Fig. 2) in progeny of swine clones versus comparator animals were found at slaughter. Male progeny of animal clones and their comparators have similar levels of E2. The levels of E2 were slightly, but not significantly, diminished in the female progeny of clones vs. the comparators. This minor decrease was considered to be part of the normal variation in blood levels that may occur depending on reproductive status of the female and time of day. The reproductive status of the female animals was not provided, however, these animals were slaughtered at approximately six months of age, when swine are generally still pre-pubertal. Nevertheless, the levels of E2 in the progeny of swine clones do not differ significantly from the comparators.

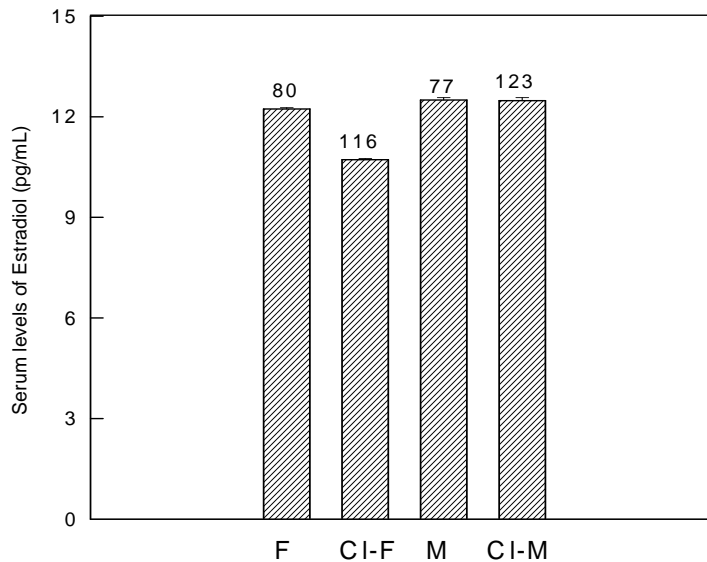


Figure F-4 depicts the levels of IGF-I in the offspring of swine clones and comparators (female and males) at slaughter. CL=offspring clones, F=female, M=male. Values are mean \pm SEM, the numbers above the bars = number of samples (animals) per group.

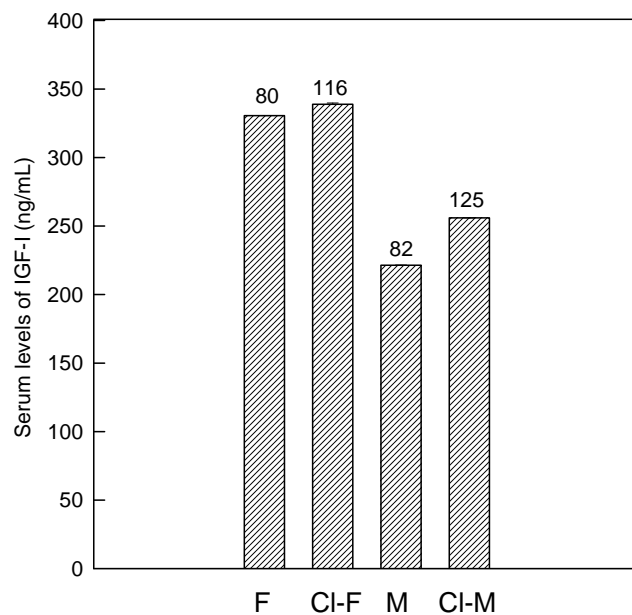


Figure F-5 depicts the levels of Estradiol- β in offspring of swine clones and comparators (female and males) at slaughter. CL=offspring clones, F=female, M=male. Values are mean \pm SEM, the numbers above the bars = number of samples (animals) per group.

Urinalysis. Urine samples were harvested after slaughter, which may account for the presence of blood in some samples. Only one clone progeny had protein in its urine. This is not unrealistic by random chance and not a health issue.

pH	6	6.5	7	7.5	8	8.5	Total
Number of Clone Progeny	10	12	16	54	146	4	242
Percentage	4%	5%	7%	22%	60%	2%	100%
Number of Comparator Progeny	13	11	13	36	89	1	163
Percentage	8%	7%	8%	22%	54.5%	0.5%	100%
Total Number of Animals	23	23	29	90	235	5	405

The distribution of pH values is similar between the clone progeny and the comparator progeny and indicates normal urine variation. This is especially true for animals on an herbivorous diet which is typical of current swine management practice (Duncan and Prasse 2003). No animals in this experiment had glucose in the urine.

Conclusions for Animal Health of Progeny of Clones. Although there was a higher death loss among progeny of clones in this study, most of this loss can be attributed to a single litter farrowed by a heat-stressed sow that did not survive. Causes of death (e.g., stillbirth, overlay, weakness) were similar to national statistics for commercially raised swine, and there were only minor differences between groups. Few animals were noted with abnormalities in either group, and the rates and types of abnormalities were similar to national statistics for commercially raised swine. Growth rates from birth to weaning for progeny of clones and comparators were similar. Differences were noted in both the early (neonatal) and mid-trial (early juvenile) blood values between progeny of clones and comparators in Study 2. The differences during the neonatal period were few and minor. The clone progeny values were considered to be within the range of variation for a normal population of neonatal animals. There are some differences between the clone progeny and comparator progeny during the second blood sampling (early juvenile period). The values for analytes with significance for liver for this second sampling period offered mixed results, none of which confirm liver abnormalities. The blood cell values for this second sampling are also inconsistent, offering no indication of blood cell abnormalities. There are increases in liver-function associated analytes in this dataset (late juvenile period). The other values indicate no negative health impact on progeny of clones.

c. Carcass Characteristics

Given the large variation in live weight at the time of slaughter, one might anticipate that many of the post-slaughter carcass characteristics, such as marbling and backfat thickness, would also vary considerably. Carcass characteristics are provided in Table F-17. Hot carcass weights were 79.9, 79.4, 79.0, and 81.2 kg for progeny from Hamline comparator, Hamline clones, Duroc comparators, and Duroc clone boars, respectively. Carcass length was also similar, 82.7, 81.6, 82.3, and 81.5 cm, respectively. The first rib values were 22.2, 23.4, 23.8, and 25.9 mm for progeny from Hamline comparator, Hamline clones, Duroc comparators, Duroc clone boars, respectively, whereas the last rib values were 16.0, 16.9, 17.4, and 19.0 mm, respectively.

Table F-17: Carcass Characteristics for Progeny Derived from Clone or Comparator Boars				
	Hampshire Comparator (Mean ± standard deviation)	Hampshire Clone (Mean ± standard deviation)	Duroc Comparator (Mean ± standard deviation)	Duroc Clone (Mean ± standard deviation)
Hot Carcass Weight (kg)	79.9±3.9	79.4±3.9	78.9±4.3	81.2±4.1
Carcass Length (cm)	82.7±2.2	81.6±2.1	82.3±2.2	81.5±2.3
Loin Eye Area (cm ²)	6.7 ± 0.8	6.8±0.8	6.6±0.8	7.2±0.9
Back fat Thickness (mm)				
First rib	22.2 ± 4.2	23.4 ± 4.4	23.8 ± 4.1	25.9 ± 4.2
Last rib	16.0 ± 2.9	16.9 ± 3.2	17.4 ± 2.4	19.0 ± 2.8
Last Lumbar	16.6 ± 3.4	17.0 ± 3.2	18.1 ± 2.6	19.3 ± 2.7
Longissimus pH at 24 hours	5.8 ± 0.2	5.7 ± 0.1	5.7 ± 0.1	5.7 ± 0.1
Carcass muscle score	3 ± 0	3 ± 0	3 ± 0	3 ± 0
NPPC Quality Scores				
Color	3 ± 0.3	3 ± 0.2	3 ± 0.1	3 ± 0
Marbling	3 ± 0.7	3 ± 0.8	3 ± 0.8	3 ± 0.9
Firmness	2 ± 0	2 ± 0	2 ± 0	2 ± 0

All animals were given score 3 for carcass muscle score and 2 for firmness. Ninety-three percent of the progeny from the clones and comparator boars had marbling score within the 2 to 4 range. Measurements of pH at 24 hours post-slaughter on the longissimus muscle were similar. Loin eye area for meat cuts for progeny from Hamline comparator, Hamline clones and comparators, as well as Duroc comparators and clones were also very similar. In summary, all of the carcass characteristics evaluated were similar between the offspring of clones and comparators.

d. Meat Composition from the Progeny of Clones and Comparators

Table F-18 provides the comparison of key nutrients between the progeny of clones and their comparators. Data were reported for 412 swine of which 242 were the progeny of clones and 163 were the progeny of comparator boars. The primary comparison was made between the reported nutrient concentrations of these two groups. A secondary comparison was made to reference swine muscle values currently in the food supply (USDA Food Composition Data for pork, fresh, composite of trimmed retail cuts (loin and shoulder blade, separable lean and fat, raw), USDA National Nutrient Database for Standard Reference Release 18¹⁴⁵). The latter comparison is less tightly controlled than the comparison with the comparator, largely due to the differences in cuts, and the unknown nature of the breed(s) of swine used in the USDA dataset.

The composition of the meat from the progeny of clones and comparators indicates that the meat samples were indistinguishable at the level of the key nutrients evaluated. Only two values (alanine and erucic acid) of 56 (0.04 percent) were not virtually identical, less than would be expected by chance alone. Neither of these differences is biologically significant.

Comparing the meat composition of either the progeny of clones or the comparators to the USDA values reveals that neither is as closely comparable to that dataset as they are to each other. For example, values for niacin and vitamin B₁₂ from the progeny of both clones and comparators were higher than the USDA values for a similar type of swine muscle (shoulder blade and loin), while virtually identical to each other. Other nutrients that differ from the USDA database include palmitic acid (16:0), palmitoleic acid (16:1), stearic acid (18:0), oleic acid (18:1), linoleic acid (18:2), arachidonic acid (20:4), and niacin. The levels of these six fatty acids are higher in the database than in the progeny of clones and comparators. Little variability was observed between the other values in the nutrient profiles of the USDA database and those obtained from the progeny of clones and comparators. The differences between the nutrient concentrations in progeny of clones and comparators compared to USDA database may be due to diet, swine genotype, or storage stability effects. The important conclusions from the two comparisons, however, are that (1) there are virtually no differences between the progeny of clones and comparators, (2) the closely genetically related comparators are a better reference point than the USDA data base, and (3) none of the differences pose a food safety concern. These data suggest that there is no increased risk for humans to consume muscle from the progeny of swine clones.

¹⁴⁵ <http://www.nal.usda.gov/fnic/foodcomp/search/>

Table F-18: Comparison of Nutrient Concentrations of Progeny from Clones and Comparators		
Nutrients¹	Progeny from Clone Boars mean \pm std. dev.	Progeny from Comparators Boars Mean \pm std. dev.
Amino Acids		
Aspartic acid	2.31 \pm 0.19	2.29 \pm 0.16
Cystine	0.25 \pm 0.02	0.25 \pm 0.01
Glutamic acid	3.76 \pm 0.34	3.71 \pm 0.27
Glycine	1.14 \pm 0.15	1.12 \pm 0.13
Histidine	0.98 \pm 0.09	0.98 \pm 0.07
Isoleucine	1.03 \pm 0.12	1.03 \pm 0.10
Leucine	1.90 \pm 0.14	1.89 \pm 0.12
Lysine	2.06 \pm 0.17	2.07 \pm 0.16
Methionine	0.61 \pm 0.05	0.62 \pm 0.04
Phenylalanine	0.96 \pm 0.09	0.94 \pm 0.08
Nutrients¹	Progeny from Clone Boars Mean \pm std. dev.	Progeny from Comparators Boars Mean \pm std. dev.
Praline	1.09 \pm 0.13	1.11 \pm 0.13
Serine	0.96 \pm 0.08	0.95 \pm 0.07
Threonine	1.09 \pm 0.09	1.08 \pm 0.07
Tyrosine	0.81 \pm 0.06	0.81 \pm 0.05
Valine	1.09 \pm 0.12	1.10 \pm 0.10
Fatty Acids and Cholesterol		
8:0 (Caprylic acid)	<0.01 ²	0.01
10:0 (Capric acid)	0.01 \pm 0.002	0.01 \pm 0.002
11:0	<0.01	<0.01
12:0 (Lauric acid)	0.01 \pm 0	0.01 \pm 0
14:0 (Myristic acid)	0.08 \pm 0.027	0.08 \pm 0.029
14:1 (Myristoleic acid)	<0.01	<0.01
15:0	<0.01	<0.01
15:1	<0.01	<0.01
16:0 (Palmitic acid)	1.39 \pm 0.38	1.40 \pm 0.49
16:1 (Palmitoleic acid)	0.17 \pm 0.06	0.16 \pm 0.05
17:0 (Margaric acid)	0.01 \pm 0.003	0.01 \pm 0.002
17:1 (Margaroleic acid)	0.01 \pm 0.003	0.01 \pm 0.002
18:0 (Stearic acid)	0.66 \pm 0.24	0.68 \pm 0.25
18:1 (Oleic acid)	2.26 \pm 0.76	2.20 \pm 0.72
18:2 (Linoleic acid)	0.3 \pm 0.11	0.29 \pm 0.11
18:3 (Linolenic acid)	0.02 \pm 0.001	0.01 \pm 0.005

Nutrients ¹	Progeny from Clone Boars Mean \pm std. dev.	Progeny from Comparators Boars Mean \pm std. dev.
18:4	0.01 \pm 0.0001	0.01 \pm 0.004
20:0 (Arachidic acid)	0.01 \pm 0.005	0.01 \pm 0.005
20:1 (Gadoleic acid)	0.08 \pm 0.04	0.07 \pm 0.04
20:2 (Eicosadienoic acid)	0.02 \pm 0.01	0.02 \pm 0.005
20:3 (Eicosatrienoic acid)	0.01 \pm 0.01	<0.01
20:4 (Arachidonic acid)	0.01 \pm 0.003	0.01 \pm 0.002
20:5 (Eicosapentaenoic acid)	0.01 \pm 0	0.01 \pm 0.004
21:5 (Heneicosapentaenoic acid)	0.01 \pm 0	<0.01
22:0 (Behenic acid)	<0.01	<0.01
22:1 (Erucic acid)	0.01 \pm 0.006	0.02 \pm 0.006
22:2 (Docosadienoic acid)	<0.01	0.01 \pm 0.01
22:3 (Docosatrienoic acid)	<0.01	<0.01
22:4 (Docosatetraenoic acid)	<0.01	<0.01
22:5 (Docosapentaenoic acid)	<0.01	<0.01
22:6 (Docosahexaenoic acid)	0.02 \pm 0.01	0.02 \pm 0.01
24:0 (Lignoceric acid)	<0.01	<0.01
24:1 (Nervonic acid)	<0.01	<0.01
Cholesterol (mg/100 g)	57.93 \pm 5.46	59.39 \pm 5.04
Minerals		
Calcium	0.01 \pm 0.003	0.01 \pm 0.002
Iron	0.00 \pm 0.0005	0.000 \pm 0.003
Phosphorus	0.18 \pm 0.082	0.16 \pm 0.082
Zinc	0.00 \pm 0.0003	0.00 \pm 0.0001
Vitamins		
Niacin (mg/100g)	10.68 \pm 1.23	10.64 \pm 1.03
Vitamin B ₆ (mg/100 g)	0.40 \pm 0.07	0.38 \pm 0.07
Vitamin B ₁₂ (mcg/100 g)	1.01 \pm 0.25	0.97 \pm 0.28
¹ Unless otherwise specified, quantities are expressed as g/100g homogenized meat.		
² Values marked with "<" indicate concentrations below the level of detection for the instrument used in the assay.		

Table F-19: Nutrient Concentrations for Pork, Fresh, Composite of Trimmed Retail Cuts (Loin And Shoulder Blade), Separable Lean and Fat, Raw as Listed in USDA National Nutrient Database	
Nutrients¹	USDA²
Amino Acids	
Aspartic acid	1.795
Cystine	0.246
Glutamic acid	3.011
Glycine	1.011
Histidine	0.761
Isoleucine	0.900
Leucine	1.556
Lysine	1.748
Methionine	0.508
Phenylalanine	0.776
Proline	0.830
Serine	0.807
Threonine	0.882
Tyrosine	0.668
Valine	1.052
Fatty Acids and Cholesterol	
8:0 (Caprylic acid)	0.000
Nutrients¹	USDA²
10:0 (Capric acid)	0.010
11:0	not listed
12:0 (Lauric acid)	0.010
14:0 (Myristic acid)	0.160
14:1 (Myristoleic acid)	not listed
15:0	not listed
15:1	not listed
16:0 (Palmitic acid)	2.79
16:1 (Palmitoleic acid)	0.37
17:0 (Margaric acid)	not listed
17:1 (Margaroleic acid)	not listed
18:0 (Stearic acid)	1.46
18:1 (Oleic acid)	5.27
18:2 (Linoleic acid)	1.140
18:3 (Linolenic acid)	0.09
18:4	0.00

Nutrients¹	USDA²
20:0 (Arachidic acid)	not listed
20:1 (Gadoleic acid)	0.100
20:2 (Eicosadienoic acid)	not listed
20:3 (Eicosatrienoic acid)	not listed
20:4 (Arachidonic acid)	0.08
20:5 (Eicosapentaenoic acid)	0.00
21:5 (Heneicosapentaenoic acid)	not listed
22:0 (Behenic acid)	not listed
22:1 (Erucic acid)	0.00
22:2 (Docosadienoic acid)	not listed
22:3 (Docosatrienoic acid)	not listed
22:4 (Docosatetraenoic acid)	not listed
22:5 (Docosapentaenoic acid)	0.000
22:6 (Docosahexaenoic acid)	0.000
24:0 (Lignoceric acid)	not listed
24:1 (Nervonic acid)	not listed
Cholesterol (mg/100 g)	64
Minerals	
Calcium	0.019
Iron	0.00082
Phosphorus	0.195
Zinc	0.00187
Vitamins	
Niacin (mg/100g)	4.492
Vitamin B ₆ (mg/100 g)	0.456
Vitamin B ₁₂ (mcg/100 g)	0.63
¹ Data expressed as quantities per 100 g of homogenized meat.	
² USDA means taken from the USDA National Nutrient Database for Standard Reference Release 18, item number 10226.	
³ Values marked with < indicate concentrations below level of detection for instrument used in assay.	

Conclusions for Carcass Characteristics and Meat Composition for Progeny of Clones and Comparators

Although some minor differences in backfat thickness were noted for progeny of clones vs. comparators, they have no significance for food safety. The increased values for niacin and B₁₂ and decreased values for six fatty acids compared to USDA values were similar for progeny of clones and comparators in this experiment, and may reflect differences in diet, genotype, or sample handling compared to the national average. Because these values were similar between

the two groups involved in this study, there is no increased risk associated with meat from progeny of clones vs. contemporary comparators. All other meat composition values were similar between groups, indicating no increased risk associated with meat from progeny of clones vs. contemporary comparators.

D. Conclusions from the ViaGen Dataset

1. Study 1: Clones vs. Comparators

a. Animal Health

The interpretation of the results of the study comparing conventionally bred and clone barrows is limited because it was not initiated until the animals were approximately 50 days of age, clones were raised under different conditions prior to initiating the experiment, and there were a limited number of clones. The conventionally derived barrows were selected based on two criteria: (1) their sire was one of the donor boars for SCNT; and (2) the pigs were similar in age and weight to the clone barrows. Retrospective evaluation of the birth weights for the seven clone barrows indicated that these animals were smaller at birth than their conventionally bred counterparts. The growth rate data would suggest that clone barrows grew as well as conventionally bred animals prior to weaning, as these animals reached the same body weight at around 50 days of age. However, after the clones were moved to the more conventional rearing facility to be raised with the AI-derived barrows, it took the clone barrows on average 27 days longer to reach their slaughter weight, and the clone barrows were on average 18.2 kg lighter when they were slaughtered than the conventionally bred barrows.¹⁴⁶ Furthermore, three of the seven clone barrows were not processed at the end of the experiment. One of the clones was condemned at slaughter due to a lung adhesion and the other two animals were approximately 45 kg lighter than their counterparts. The health records for the clones indicate that these animals developed several health issues including scouring. These clones were born and maintained under highly biosecure conditions until the beginning of the study (at approximately 50 days of age), were potentially premature at delivery, and were deprived of colostrum. Thus, moving these animals to a conventional production system could have had a dramatic effect on their growth rate. However, four of the seven clone barrows responded appropriately and overcame the pathogenic challenge.

¹⁴⁶ As mentioned previously, because animal clones are voluntarily withheld from entering the food supply, the slaughter facility could only make certain dates available to slaughter these animals. Thus, the clones were slaughtered as a group on one of the pre-selected, available dates once their average weight approached the target weight.

b. Food Safety

The most significant difference between the comparator and clone barrows at slaughter was a trend for higher backfat thickness in the conventionally bred animals, consistent with the observation that the conventionally bred animals were heavier at the time of slaughter than clones.

Data were presented on the key nutrient levels of *latissimus dorsi* muscle from swine clones and comparators. Fifty-six nutrients were measured in tissue samples of five clones (one animal was euthanized for health reasons) and 15 comparator swine. There were limitations in the usefulness of the data due to the study design and data reporting. A comparison of reported values to reference swine muscle values (USDA National Nutrient Database for Standard Reference Release 18¹⁴⁷) was possible for only four nutrients because the other nutrients were reported as percentages. Values for niacin and vitamin B₁₂ in both clones and comparator swine were above USDA values for a similar type of swine muscle (shoulder blade and loin), but were similar to each other. Values for cholesterol and vitamin B₆ were similar to the USDA values. Little variability between reported values was observed when the data were examined by nutrient.

The lack of variability observed in the food composition values between muscle of swine clones and comparators supports a conclusion that there is no additional risk in the human consumption of muscle from swine clones. However, limitations in the study design, reporting of data, and the elevated niacin and vitamin B₁₂ concentrations in both clone and comparator muscle compared to the USDA reference values diminish the confidence of this conclusion. A more definitive comparison between the food composition of clone and comparator swine muscle could be made if more of the analyses could be compared to values in reference (USDA) swine muscle.

2. Progeny of Clone Boars vs. Progeny of Comparator Boars

This experiment was designed to determine whether progeny from clones performed as well as progeny from comparator boars and if food products from progeny of clones would pose any additional risk relative to corresponding products derived from comparator animals. Data were provided on 300 and 402 progeny derived from comparator and clone boars, respectively.

a. Animal Health

Although there was a higher percentage loss of pigs at birth for progeny derived from clone boars, this difference was primarily due to the loss of an entire litter of 13 pigs. Secondary factors that potentially influenced the number of pigs that died due to the dam lying on pigs

¹⁴⁷ Ibid.

included environmental conditions at the time of farrowing and the number of litters that were born on a single day. Although there was a slight increase in the percentage of progeny of clones that were crushed by their dam, there was a similar number of pigs/litter crushed by their dams among comparators, and these values were similar to values that are commonly found within the swine industry. Thus, there do not appear to be any differences in the survival of progeny from clone boars when compared to progeny from conventionally derived boars. There was a similar percentage of mummified pigs presented at birth. The survival and growth rate data do not show any animal health concerns for progeny of clone boars when compared to progeny from conventionally derived boars. Hematology, clinical chemistry and urinalysis values for clone progeny were considered to be within the range of variation for a normal population of animals.

b. Food Safety

Data were presented on the food composition of *latissimus dorsi* muscle from the progeny of swine clones and controls. Fifty-eight nutrients were measured in the tissue samples of 242 AI-derived comparators and 163 clone progeny. A positive aspect of the study is the large numbers of test swine and the numerous nutrients analyzed. Negative aspects of the study are the lack of method performance data in the test matrix (meat), the choice of *latissimus dorsi* as the test matrix instead of a retail pork cut and/or another edible tissue, and the lack of storage stability data.

The nutrient concentrations of clone progeny and comparator swine are very similar. A few nutrients did have differences in the variability and distribution of values between clone progeny and comparator swine. We evaluated the differences, and determined they were minor and not biologically significant.

Most of the nutrient concentrations were similar to USDA reference values. Six fatty acids had lower concentrations in both the clone progeny and comparator swine compared to the USDA values. One B vitamin was higher in the clone progeny and comparator swine than the USDA value. The difference between these nutrient concentrations in clone progeny and comparator progeny compared to USDA values may be due to effect of diet, swine breed, or storage stability effects on method performance.

Based on the lack of difference in the nutrient concentrations between muscle of progeny from AI-derived comparator and clone boars, we conclude that there is no increased risk for humans to consume muscle from the progeny of clone swine. The current study provides no information regarding the food composition of other swine edible tissue (liver, kidney, fat). Therefore, food safety conclusions about muscle cannot be extrapolated to other edible tissues of swine.

E. Addendum

On January 5, 2006, ViaGen Inc. faxed several pages of data from the re-assay of samples which they had identified as outliers using the criteria outlined for the Cyagra dataset (Appendix E). Samples from 15 clone progeny were re-assayed because the values for specific nutrients were > 10 percent above or below the range of values for comparators. Table F-20 provides a comparison of the original and re-assay values for the nutrients assayed by animal.

Table F-20. Comparison of original and re-assay values of select nutrients from meat of clone progeny.			
Nutrient¹	Animal ID	Original Value	Re-assay Value
Amino Acids			
Aspartic Acid	200437509	3.33	2.30
Fatty Acids			
10:0 (Capric)	200430710	0.01	<0.01
	200438107	0.02	0.01
14:0 (Myristic)	200430710	0.15	0.12
	200431409	0.03	0.05
	200430701	0.04	0.07
	200431606	0.12	0.09
	200432702	0.07	0.09
	200438107	0.21	0.20
16:0 (Palmitic)	200430710	2.40	2.03
	200431409	0.45	0.99
	200430701	0.53	1.00
	200432702	1.10	1.52
	200438107	3.62	3.63
16:1 (Palmitoleic)	200430710	0.27	0.15
	200431409	0.06	0.12
	200430701	0.06	0.15
	200431606	0.25	0.17
	200432702	0.12	0.20
	200438107	0.38	0.36
17:1 (Margaroleic)	200430710	0.02	0.01
	200431606	<0.01 ²	0.01
	200438107	0.02	0.01
18:0 (Stearic)	200430710	1.17	1.11
	200431409	0.21	0.49
	200430701	0.23	0.42
	200431606	0.99	0.68
	200432702	0.48	0.68
	200438107	1.77	1.78

Nutrient¹	Animal ID	Original Value	Re-assay Value
18:1 (Oleic)	200430710	4.63	3.30
	200431409	0.79	1.71
	200430701	0.69	1.76
	200431606	3.53	2.43
	200432702	1.38	2.45
	200438107	5.44	5.67
18:2 (Linoleic)	200430710	0.70	0.46
	200431409	0.11	0.32
	200430701	0.07	0.29
	200431606	0.92	0.39
	200432702	0.06	0.33
	200438107	0.55	0.61
18:3 (Linolenic)	200430710	0.04	0.01
	200431409	<0.01	0.02
	200430701	<0.01	0.01
	200431606	0.05	0.01
	200432702	<0.01	0.02
	200438107	0.02	0.03
18:4	200430710	0.01	<0.01
20:0 (Arachidic)	200431606	0.02	0.01
	200438107	0.03	0.02
20:1 (Gadoleic)	200430710	0.14	0.07
	200430701	0.05	0.03
	200431606	0.07	0.04
	200432702	0.04	0.03
	200438107	0.13	0.09
20:2 (Eicosadienoic acid)	200430710	0.04	0.06
	200431409	<0.01	0.03
	200430701	<0.01	0.01
	200431606	0.04	0.03
	200432702	<0.01	0.04
	200438107	0.03	0.03
20:3 (Eicosatrienoic acid)	200431606	0.01	<0.01
Nutrient¹	Animal ID	Original Value	Re-assay Value
20:4 (Arachidonic)	200430701	<0.01	0.01
	200431606	0.02	<0.01
22:1 (Erucic)	200438107	0.02	<0.01
22:6 (Docosahexaenoic)	200430710	0.01	<0.01
	200432702	0.03	<0.01
	200438107	0.05	<0.01
Minerals			
Calcium	200430609	0.034	0.0042
	200430708	0.021	0.0045
Phosphorus	200433206	0.72	0.021
Zinc	200438104	0.0025	0.0014
	200431008	0.0046	0.0015

Nutrient ¹	Animal ID	Original Value	Re-assay Value
Vitamins			
Niacin (mg/100g)	200433002	19.1	8.78
Vitamin B ₁₂ (mcg/100 g)	20045803	2.20	1.20
¹ Unless otherwise specified, quantities are expressed as g/100g homogenized meat.			
² Values marked with "<" indicate concentrations below the level of detection for the instrument used in the assay.			

CVM conducted a follow-up analysis of the data using the new values and found they had only very minor effects on the average nutrient values for clone progeny. The reanalyzed means are presented in Table F-21, and compared to the original means for progeny of clones and comparators.

Table F-21: Comparison of Nutrient Concentrations of Progeny from Clones and Comparators			
Nutrients ¹	Progeny from Clone Boars (Original) mean \pm std. dev.	Progeny from Clone Boars (Reanalyzed) mean \pm std. dev.	Progeny from Comparators Boars Mean \pm std. dev.
Amino Acids			
Aspartic acid	2.31 \pm 0.19	2.30 \pm 0.17	2.29 \pm 0.16
Fatty Acids			
10:0 (Capric acid)	0.01 \pm 0.002	0.00 ² \pm 0.003	0.01 \pm 0.002
14:0 (Myristic acid)	0.08 \pm 0.027	0.08 \pm 0.027	0.08 \pm 0.029
16:0 (Palmitic acid)	1.39 \pm 0.38	1.39 \pm 0.47	1.40 \pm 0.49
16:1 (Palmitoleic acid)	0.17 \pm 0.06	0.17 \pm 0.06	0.16 \pm 0.05
17:1 (Margaroleic acid)	0.01 \pm 0.003	0.00 \pm 0.004	0.01 \pm 0.002
18:0 (Stearic acid)	0.66 \pm 0.24	0.66 \pm 0.23	0.68 \pm 0.25
18:1 (Oleic acid)	2.26 \pm 0.76	2.26 \pm 0.74	2.20 \pm 0.72
18:2 (Linoleic acid)	0.3 \pm 0.11	0.3 \pm 0.10	0.29 \pm 0.11
18:3 (Linolenic acid)	0.02 \pm 0.001	0.01 \pm 0.009	0.01 \pm 0.005
18:4	0.01 \pm 0.000	0.00 \pm 0.002	0.01 \pm 0.004
20:0 (Arachidic acid)	0.01 \pm 0.005	0.01 \pm 0.008	0.01 \pm 0.005
20:1 (Gadoleic acid)	0.08 \pm 0.04	0.08 \pm 0.04	0.07 \pm 0.04
20:2 (Eicosadienoic acid)	0.02 \pm 0.01	0.01 \pm 0.01	0.02 \pm 0.005
20:3 (Eicosatrienoic acid)	0.01 \pm 0.01	<0.01 ³	<0.01
20:4 (Arachidonic acid)	0.01 \pm 0.003	0.00 \pm 0.002	0.01 \pm 0.002
22:6 (Docosahexaenoic acid)	0.02 \pm 0.01	<0.01	0.02 \pm 0.01

Nutrients¹	Progeny from Clone Boars (Original) mean \pm std. dev.	Progeny from Clone Boars (Reanalyzed) mean \pm std. dev.	Progeny from Comparators Boars Mean \pm std. dev.
Minerals			
Calcium	0.01 \pm 0.003	0.01 \pm 0.002	0.01 \pm 0.002
Phosphorus	0.18 \pm 0.082	0.18 \pm 0.075	0.16 \pm 0.082
Zinc	0.00 \pm 0.0003	0.00 \pm 0.0002	0.00 \pm 0.0001
Vitamins			
Niacin (mg/100g)	10.68 \pm 1.23	10.64 \pm 1.11	10.64 \pm 1.03
Vitamin B ₁₂ (mcg/100 g)	1.01 \pm 0.25	0.93 \pm 0.34	0.97 \pm 0.28
¹ Unless otherwise specified, quantities are expressed as g/100g homogenized meat. ² Values of 0.00 reflect means less than 0.01. ³ Values marked with “<” indicate concentrations below the level of detection for the instrument used in the assay.			

Of the 22 nutrient values that were reanalyzed, 12 means were unchanged compared to the original values. Nine values were changed (0.01 g/100g) compared to the original values. Only the mean for Vitamin B₁₂ differed by more than 0.01 g/100 g; however, the reanalyzed mean was similar to the mean for comparators (0.93 \pm 0.34 vs. 0.97 \pm 0.28 g/100 g). All 22 values were similar to values for comparators. None of these minor changes affect the assumptions regarding the safety of meat from clone progeny.

ViaGen Data Chart Interpretations

Chart F-1 (Page 1): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (July 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary			
200430601	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200430602	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200430603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430605	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	2	/	35	
200430606	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430607	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430608	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200430609	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	↓	↓	■	↑	■	■	■	■	■	■	■	↓	■	■	↓	■	■	■	7	/	35		
200430610	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430611	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430701	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430702	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430703	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430704	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430705	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430706	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200430707	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	↑	2	/	35
200430708	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430709	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35	
200430710	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200430711	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200430801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430802	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430803	■	■	↑	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	↓	■	■	■	■	■	■	4	/	35	
200430804	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200430805	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-1 (Page 2): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (July 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary											
200430901																																															
200430902	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35			
200430903	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35		
200430904																																															
200430905	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35		
200430906	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200431001	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200431002	■	■	■	■	■	■	■	↑	↓	■	↑	■	↑	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	6	/	35			
200431003	■	■	↑	↑	■	↑	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	7	/	35			
200431004	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35		
200431005	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200431006	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35	
200431007	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200431008	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200431009	■	■	■	■	■	■	■	↑	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3	/	35		
200431010	■	■	↑	■	■	■	■	■	↓	↓	■	↓	■	■	■	■	↓	■	■	■	■	■	■	↓	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	7	/	35			
200431011	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200431012	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35		
200431013																																															
200431401	■	↑	■	■	■	■	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	5	/	35	
200431402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35	
200431403	■	■	■	↑	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3	/	35	
200431404	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200431405	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200431406	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200431407	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200431408	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35		

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-1 (Page 3): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (July 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary
200431409	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200431410	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200431411	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	2 / 35	
200431501	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200431502	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	↑	■	■	4 / 35	
200431503	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200431504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	1 / 35	
200431505	■	■	■	■	■	↑	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3 / 35	
200431506	■	■	■	■	■	↑	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↓	■	■	■	■	■	4 / 35	
200431601	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200431602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200431603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200431604	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200431605	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	1 / 35	
200431606	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	2 / 35	
200431607	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200431608	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200431609																																				
200431610	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35	
200431701	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200431702	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200431703	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200431704	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200431705																																				
200431706	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200431707																																				
200431708	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	↑	■	■	■	■	■	↑	■	■	■	■	■	■	■	3 / 35	

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-1 (Page 4): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (July 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary		
200432001	■	■	■	↑	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3	/	35
200432002	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200432003	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3	/	35
200432004	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	2	/	35	
200432005	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35	
200432006	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200432007	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3	/	35
200432008	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35	
200432101	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200432102	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200432103	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200432104	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200432105	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35	
200432106	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	2	/	35	
200432107																																						
200432108	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200432109	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200432301	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200432302	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200432303	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200432304	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200432305	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200432306	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	4	/	35	
200432307	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35	
200432308	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200432309	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200432310	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-1 (Page 5): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (July 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary	
200432311	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432312	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	1 / 35	
200432401	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432403	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432404	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	2 / 35	
200432405	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	2 / 35	
200432406	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432407	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200432408																																					
200432501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200432503	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200432504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200432505																																					
200432506	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200432507	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200432508	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200432701	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200432702	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200432703	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200432704	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200432705	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200432706	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200433001	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	↑	■	■	■	■	■	■	■	■	■	2 / 35
200433002	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	1 / 35
200433003	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-1 (Page 6): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (July 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary
200433004	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200433005	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200433006	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	1 / 35
200433007	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200433008	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200433009																																				
200433010	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	↓	■	■	■	■	■	■	■	3 / 35
200433011	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	3 / 35
200433201	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200433202	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200433203	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	1 / 35
200433204	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200433205	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200433206	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	2 / 35
200433207	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	2 / 35
200433208	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	↑	■	↑	■	■	■	■	■	■	■	■	↑	■	■	↑	↑	■	■	■	■	6 / 35
200433601																																				
200433602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1 / 35
200433603																																				
200433604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1 / 35
200433605	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1 / 35
200433701	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	2 / 35
200433702	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1 / 35
200433703	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	2 / 35
200433704	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	2 / 35
200433705	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	3 / 35
200433706	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	3 / 35

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-1 (Page 7): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (July 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary			
200433707	■	■	■	■	■	■	↑	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3	/	35	
200433708	■	■	■	↑	■	■	■	■	■	■	■	↑	■	↑	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	6	/	35
200433709	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35
200433710	■	↓	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3	/	35	
200433711	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200433712	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35
200433801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200433802	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35
200433803	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35
200433804	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	3	/	35
200433805																																							
200433806	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35
200433807																																							
200433808	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35
200433809	■	↓	■	■	■	■	■	■	■	■	■	↑	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	5	/	35
200434101	■	■	■	■	■	■	■	■	↑	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	4	/	35
200434102	■	■	↑	■	■	■	■	■	↑	■	■	↑	■	■	↑	■	■	■	■	↑	■	■	■	■	■	■	↑	■	↓	■	■	■	■	■	■	■	7	/	35
200434103	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35
200434104	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200434105	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200434106	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434107	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434108	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434109	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434110																																							
200434111	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200434112	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-1 (Page 8): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (July 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary			
200434113	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	↓	■	■	■	■	■	■	■	■	■	■	■	5	/	35	
200434114	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200434115	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434401	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35	
200434402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200434403	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434501	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3	/	35
200434502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	1	/	35
200434503	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200434504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434505																																							
200434506	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35
200434507	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200434508	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434509	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434510	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200434511	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200434512	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434601	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434603																																							
200434604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200434605	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200434606	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434607	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	↑	3	/	35
200434608	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200435801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-1 (Page 9): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (July 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary				
200435802	↑	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3	/	35		
200435803	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436001	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436002	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436003	↑	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	4	/	35		
200436004	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	↑	■	■	■	↓	■	↑	■	■	■	■	■	■	■	■	↑	5	/	35	
200436005	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	1	/	35	
200436006	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200436007	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	2	/	35	
200436008	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436009	■	■	■	↑	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	3	/	35	
200436010	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436011	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436012	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200436013	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	1	/	35	
200436014																																								
200436401	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	2	/	35	
200436402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200436403	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200436404	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200436405	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200436601	■	■	■	■	■	■	■	■	■	■	■	↑	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	1	/	35	
200436602	■	■	■	↑	■	■	■	■	■	■	■	↑	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	4	/	35	
200436603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436605	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436606	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-1 (Page 10): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (July 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary			
200436607	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436608	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	↑	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	3	/	35	
200436609	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436610	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436611	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35		
200436612	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436901																																							
200436902	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200436903	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436904	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436905	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436906	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436907	■	■	■	↑	■	■	■	↑	■	■	↑	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	5	/	35		
200436908	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436909	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	↑	■	■	■	■	■	■	2	/	35	
200436910	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436911	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436912	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200436913	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200437401	■	■	■	■	↑	↑	■	■	■	■	■	↑	■	↑	↑	■	■	■	↑	↑	■	■	■	■	↓	■	↑	■	↓	■	■	↑	■	↑	■	#	/	35	
200437402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200437403	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200437404	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	1	/	35	
200437405	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	2	/	35
200437406	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	1	/	35
200437407	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200437408	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
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Chart F-1 (Page 11): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (July 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary						
200437409	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35				
200437410	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35			
200437411	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35			
200437501	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	2	/	35			
200437502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35			
200437503																																										
200437504																																										
200437505	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35		
200437506	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200437507	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200437508																																										
200437509	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200437510	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200437801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200437802	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200437803	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200437804	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	2	/	35
200438101	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200438102	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200438103	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200438104	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200438105	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200438106	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200438107	■	■	■	■	■	↓	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35
200438601	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200438602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200438603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-1 (Page 12): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (July 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary
200438604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200438605	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200438606	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200438607	■	■	■	↑	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3 / 35	
200438608	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200438609	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200438610	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200438611	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200439501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200439502	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35
200439503																																				
200439504	■	■	■	↑	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3 / 35
200439505	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
Summary	2 / 287	4 / 287	4 / 287	15 / 287	1 / 287	9 / 287	21 / 287	6 / 287	15 / 287	2 / 287	11 / 287	17 / 287	1 / 287	19 / 287	14 / 287	1 / 287	3 / 287	7 / 287	6 / 287	28 / 287	0 / 287	4 / 287	3 / 287	5 / 287	11 / 287	37 / 287	7 / 287	9 / 287	7 / 287	26 / 287	10 / 287	0 / 287	5 / 287	1 / 287	9 / 287	

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-2 (Page 1): Summary of Hematology Data for Viagen Clone Progeny and Comparators (July 2004)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary
200430601	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430602	■	■	■	■	■	■	■	■	↑	■	■	↓	↓	■	■	■	■	■	3 / 18
200430603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430605	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	1 / 18
200430606	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200430607	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200430608	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430609																			
200430610	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	1 / 18
200430611	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430701	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200430702	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430703	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430704	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430705	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430706	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430707																			
200430708	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	1 / 18
200430709																			
200430710	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430711	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430802																			
200430803	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200430804																			
200430805	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430901																			
200430902	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430903	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430904																			
200430905	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430906	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200431001	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200431002																			
200431003	■	■	■	■	■	↓	■	■	■	■	■	■	↓	■	■	■	■	■	2 / 18
200431004	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431005	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200431006																			
200431007	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-2 (Page 3): Summary of Hematology Data for Viagen Clone Progeny and Comparators (July 2004)

200432004	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200432005	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200432006	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432007																					
200432008																					
200432101	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	↑	■	■	■	2	/	18
200432102																					
200432103	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432104	■	■	■	■	↓	↑	■	■	■	■	■	↑	■	■	↑	■	■	■	4	/	18
200432105																					
200432106	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432107																					
200432108	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200432109	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432301	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432302	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432303	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432304	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432305																					
200432306																					
200432307																					
200432308																					
200432309	■	■	■	■	■	■	■	■	■	■	■	↑	↑	■	■	■	■	■	2	/	18
200432310																					
200432311	■	■	■	■	■	■	■	■	■	■	■	↑	■	↑	■	■	■	■	2	/	18
200432312	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200432401	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432402																					
200432403	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200432404	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432405	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200432406																					
200432407	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	1	/	18
200432408																					
200432501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432503	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432505																					
200432506	■	■	↑	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	2	/	18
200432507	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432508	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432701	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200432702	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-2 (Page 5): Summary of Hematology Data for Viagen Clone Progeny and Comparators (July 2004)

200433805																					
200433806	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200433807	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200433808	■	■	■	■	■		■	■	■	■	■	↑	■	■	■	■	■	■	1	/	18
200433809																					
200434101	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434102	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434103	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434104	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434105	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434106	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434107	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434108	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434109	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434110	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434111	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434112	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434113	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434114	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434115	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	2	/	18
200434401	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434402																					
200434403	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434501																					
200434502																					
200434503																					
200434504																					
200434505																					
200434506																					
200434507	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434508																					
200434509	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434510	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434511	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434512	■	■	■	■	↓	↑	↑	■	■	■	■	■	■	↑	■	↑	■	■	5	/	18
200434601	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434605	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434606	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434607																					
200434608	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200435801	■	■	↑	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	2	/	18

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-2 (Page 6): Summary of Hematology Data for Viagen Clone Progeny and Comparators (July 2004)

200435802	■	■	■	■	■	↓	■	■	■	■	■	↑	■	■	■	■	■	■	2 / 18
200435803	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436001	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436002	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200436003																			
200436004	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	1 / 18
200436005	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436006	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	↑	■	■	■	2 / 18
200436007	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436008																			
200436009																			
200436010	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	1 / 18
200436011	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436012	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	1 / 18
200436013	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436014																			
200436401	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200436402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436403	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200436404																			
200436405	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436601	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436602																			
200436603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436605	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436606	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436607	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200436608	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436609	■	■	■	↓	↓	↓	■	■	■	↑	■	■	■	↑	■	↑	■	■	6 / 18
200436610	■	■	■	↓	↓	↓	■	■	■	■	■	■	■	↑	■	↑	■	■	4 / 18
200436611	■	■	↑	↓	↓	↓	■	■	↑	↓	■	■	■	↓	↑	■	■	■	8 / 18
200436612	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436901	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436902	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436903	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436904	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436905																			
200436906	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	↑	■	■	2 / 18
200436907																			
200436908	■	■	■	■	■	↑	↑	■	■	■	■	■	■	■	■	■	↑	■	3 / 18
200436909	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436910	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200436911																			

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-2 (Page 7): Summary of Hematology Data for Viagen Clone Progeny and Comparators (July 2004)

200436912	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436913																				
200437401	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	2	/	18
200437402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200437403	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200437404	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	1	/	18
200437405	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	1	/	18
200437406	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200437407	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200437408																				
200437409	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1	/	18
200437410	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200437411																				
200437501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200437502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200437503																				
200437504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200437505	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	1	/	18
200437506	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200437507	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200437508																				
200437509	■	■	■	■	■	↓	■	■	■	■	■	↑	■	■	■	■	■	2	/	18
200437510	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200437801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200437802	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200437803	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200437804	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200438101	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	1	/	18
200438102	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200438103	■	■	■	■	■	■	■	↓	■	↑	■	■	■	■	■	↑	↑	4	/	18
200438104																				
200438105	■	■	↑	■	■	↓	■	■	↓	■	■	■	■	■	■	■	■	3	/	18
200438106	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200438107	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200438601	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200438602	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200438603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200438604	■	■	■	↓	↓	■	■	■	■	■	■	↑	↓	↑	■	■	■	5	/	18
200438605	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200438606	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	1	/	18
200438607																				
200438608																				
200438609	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200438610	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	1	/	18

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-2 (Page 8): Summary of Hematology Data for Viagen Clone Progeny and Comparators (July 2004)

200438611	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200439501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200439502																					
200439503	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200439504																					
200439505	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
Summary	0	0	9	3	7	76	4	0	12	1	3	15	9	7	10	1	7	3			
	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/			
	235	235	235	235	235	235	235	235	235	235	235	235	235	235	235	235	235	235			

■ In Comparison Range

↑/↓ Animal is above/below reference range

Blank rows indicate values that were missing

Chart F-3 (Page 5): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (October 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary	
200432311	■	■	■	■	■	↓	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3 / 35		
200432312	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35		
200432401	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200432402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35		
200432403	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200432404	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200432405	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200432406	■	■	■	■	■	↓	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	3 / 35	
200432407	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200432408																																					
200432501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200432502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432503	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432505																																					
200432506	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35		
200432507	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	2 / 35	
200432508																																					
200432701	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35		
200432702	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432703	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200432704	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200432705	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432706	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200433001	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	2 / 35
200433002	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200433003	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35	

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-3 (Page 7): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (October 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary			
200433707	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35		
200433708	↓	■	↑	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3	/	35	
200433709	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200433710	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200433711	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35	
200433712	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200433801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200433802	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	2	/	35	
200433803	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200433804	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	2	/	35	
200433805																																							
200433806	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	↑	4	/	35
200433807	■	■	■	■	■	↓	■	■	■	■	■	↑	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	4	/	35
200433808	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	2	/	35
200433809	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	2	/	35
200434101	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	1	/	35
200434102	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434103																																							
200434104	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434105	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434106																																							
200434107	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434108																																							
200434109	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200434110																																							
200434111	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35
200434112																																							

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-3 (Page 11): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (October 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary			
200437409																																							
200437410																																							
200437411																																							
200437501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200437502	■	■	■	↑	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	3 / 35		
200437503																																							
200437504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200437505																																							
200437506	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35		
200437507																																							
200437508																																							
200437509	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35		
200437510	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	2 / 35	
200437801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200437802	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200437803	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200437804	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	2 / 35	
200438101	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200438102	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35	
200438103																																							
200438104	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200438105	■	■	■	↑	■	■	■	■	■	■	■	↑	■	↑	■	■	↑	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	5 / 35	
200438106	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35	
200438107	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3 / 35	
200438601																																							
200438602	■	■	■	■	■	↓	↑	■	■	↓	■	■	■	■	■	■	■	↓	↓	■	■	■	■	■	■	■	■	■	■	↓	■	↓	■	↓	↓	■	■	9 / 35	
200438603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	1 / 35

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-3 (Page 12): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (October 2004)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary				
200438604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35		
200438605	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	↑	↑	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	11	/	35	
200438606	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200438607	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200438608	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200438609	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
200438610	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200438611	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35	
200439501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	4	/	35
200439502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35	
200439503	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35	
200439504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	4	/	35
200439505	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35	
Summary	5	2	9	13	0	42	9	4	5	6	6	13	2	11	25	1	12	11	3	20	5	0	2	4	1	11	1	1	6	4	8	1	42	6	4					
	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/			
	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264	264			

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-4 (Page 1): Summary of Hematology Data for Viagen Clone Progeny and Comparators (October 2004)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary
200430601	■	■	■	■	■	■	■	↓	■	■	■	■	↑	■	■	■	■	■	2 / 18
200430602																			
200430603	■	■	■	■	■	■	■	↓	■	↓	■	■	■	■	■	■	■	■	2 / 18
200430604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430605	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430606																			
200430607	■	■	↑	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	3 / 18
200430608	■	■	■	■	■	■	■	↓	■	↓	■	■	■	■	■	■	■	■	2 / 18
200430609	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430610	■	■	■	■	■	■	■	↓	■	↓	■	■	■	↑	■	■	■	■	3 / 18
200430611	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430701																			
200430702	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430703	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	1 / 18
200430704	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430705	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	1 / 18
200430706	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430707	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430708	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430709																			
200430710	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430711	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430802	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200430803	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430804	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430805	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430901																			
200430902	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430903	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430904																			
200430905	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430906																			
200431001	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431002	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	1 / 18
200431003	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200431004	■	■	■	■	■	■	■	■	■	■	■	↓	↓	■	■	■	■	■	2 / 18
200431005	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431006	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431007	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	1 / 18

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-4 (Page 2): Summary of Hematology Data for Viagen Clone Progeny and Comparators (October 2004)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary	
200431008	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200431009	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431010	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431011	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431012	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431401	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431403	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431404	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431406																				
200431407	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431408	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431409	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 18
200431410	■	■	■	■	■	■	■	↓	■	↓	■	■	■	↑	■	■	■	■	■	3 / 18
200431411	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431501																				
200431502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431503	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431505	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431506	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431601	■	■	■	■	■	■	■	↓	■	↓	■	■	■	■	■	■	■	■	■	2 / 18
200431602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431603	■	■	■	■	■	■	■	↓	■	↓	■	■	■	■	■	■	■	■	■	2 / 18
200431604	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200431605	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	2 / 18
200431606	■	■	■	↓	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 18
200431607																				
200431608	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431609																				
200431610	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431701	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431702	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431703	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431704	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431705																				
200431706	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431707																				
200431708	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432001	■	↑	↑	■	■	↑	↑	■	■	■	■	■	■	■	■	■	■	■	↑	5 / 18
200432002	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-4 (Page 3): Summary of Hematology Data for Viagen Clone Progeny and Comparators (October 2004)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary
200432003	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432004	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432005	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432006	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432007	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432008	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	1 / 18
200432101	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432102																			
200432103																			
200432104																			
200432105	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432106	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200432107																			
200432108	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432109	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432301																			
200432302	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432303																			
200432304	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432305	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432306	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	1 / 18
200432307	■	■	■	■	■	■	■	↓	■	↓	■	■	■	■	↑	■	■	■	3 / 18
200432308	■	↑	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	2 / 18
200432309	■	■	■	■	■	■	■	↓	■	↓	■	■	■	■	↑	■	■	■	3 / 18
200432310	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432311	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432312	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432401	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432403	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432404	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432405	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432406	■	■	■	↓	↓	■	■	■	■	■	■	■	■	↓	■	■	■	■	3 / 18
200432407	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432408																			
200432501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432503	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	2 / 18
200432504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432505																			

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-4 (Page 4): Summary of Hematology Data for Viagen Clone Progeny and Comparators (October 2004)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary
200432506	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432507	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200432508																			
200432701	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432702	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432703	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200432704	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432705	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432706	■	■	■	■	■	■	■	↑	■	↑	■	■	■	■	■	■	■	■	2 / 18
200433001	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433002	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200433003	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200433004	■	↑	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	2 / 18
200433005	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433006	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433007	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433008	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433009																			
200433010	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	1 / 18
200433011	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200433201	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433202	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200433203	■	↑	↑	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	↑	4 / 18
200433204	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433205	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200433206	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433207	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433208	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433601	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433602	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200433603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433605	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200433701	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433702	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433703	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433704	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433705	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200433706	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433707	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-4 (Page 5): Summary of Hematology Data for Viagen Clone Progeny and Comparators (October 2004)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary
200433708	■	■	■	■	■	↑	■	↓	■	↓	■	■	■	■	■	■	■	■	3 / 18
200433709	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433710	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433711	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433712	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433802	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433803	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433804	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433805																			
200433806	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200433807	■	■	■	■	■	■	■	↓	■	↓	■	■	■	■	↑	■	■	■	3 / 18
200433808	■	■	■	■	■	■	■	↓	■	↓	■	■	■	↑	↑	■	■	■	4 / 18
200433809	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	2 / 18
200434101	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434102	■	↑	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	↑	3 / 18
200434103	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434104	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434105	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434106	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434107	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434108	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434109	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	2 / 18
200434110	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434111	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434112	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434113	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200434114																			
200434115																			
200434401	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434402	■	↑	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	↑	3 / 18
200434403	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434501	■	■	↑	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	2 / 18
200434502	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200434503	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200434504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434505																			
200434506	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200434507	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200434508	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-4 (Page 6): Summary of Hematology Data for Viagen Clone Progeny and Comparators (October 2004)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary		
200434509	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434510	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434511	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434512	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434601	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434603	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434604	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434605	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434606	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200434607	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200434608	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200435801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200435802	■	■	■	↑	↑	■	■	■	■	■	■	■	↑	■	↑	■	■	■	4	/	18
200435803	■	■	■	↑	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	3	/	18
200436001	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436002	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436003	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436004	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436005	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436006	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1	/	18
200436007	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436008	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1	/	18
200436009	■	■	■	↑	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	3	/	18
200436010	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1	/	18
200436011	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436012	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436013	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436014																					
200436401	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200436402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436403	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	18
200436404	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436405	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436601	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436605	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18
200436606	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	18

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-4 (Page 7): Summary of Hematology Data for Viagen Clone Progeny and Comparators (October 2004)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary
200436607	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436608	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436609	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200436610	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436611	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	↓	■	■	3 / 18
200436612	■	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	2 / 18
200436901	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436902	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436903	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200436904	■	↑	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	↑	3 / 18
200436905																			
200436906	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436907	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436908	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436909	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200436910	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436911	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436912	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436913	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437401	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437403	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437404	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	2 / 18
200437405																			
200437406	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437407																			
200437408	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437409																			
200437410																			
200437411																			
200437501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200431013																			
200431402																			
200431405																			
200437503																			
200437504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437505																			
200437506	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437507																			

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-4 (Page 8): Summary of Hematology Data for Viagen Clone Progeny and Comparators (October 2004)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary		
200437508																					
200437509	■	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	2 /	18
200437510	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200437801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200437802	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1 /	18
200437803	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200437804	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200438101	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200438102																					
200438103																					
200438104																					
200438105	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	1 /	18
200438106	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200438107	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	1 /	18
200438601																					
200438602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200438603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200438604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200438605	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	1 /	18
200438606	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200438607	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200438608	■	■	■	↓	■	■	■	↓	■	↓	■	■	■	■	■	■	↑	■	■	4 /	18
200438609	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200438610	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 /	18
200438611	■	■	■	■	■	■	■	↓	■	↓	■	■	■	■	■	■	■	■	■	2 /	18
200439501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1 /	18
200439502	■	■	■	↓	↓	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	3 /	18
200439503	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200439504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 /	18
200439505	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1 /	18
Summary	2 / 265	17 / 265	20 / 265	7 / 265	11 / 265	9 / 265	6 / 265	16 / 265	0 / 265	14 / 265	4 / 265	3 / 265	7 / 265	14 / 265	8 / 265	3 / 265	5 / 265	5 / 265			

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-5 (Page 4): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (January 2005)

Animal_ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary
200432001	■	■	■	■	■	■	■	■	■	■	↑	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35	
200432002	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200432003	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200432004	■	■	■	■	■	■	■	■	■	■	↑	↑	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	4 / 35	
200432005	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432006	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200432007	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	2 / 35	
200432008	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432101	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432102	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200432103	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200432104	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	1 / 35	
200432105	↓	■	↑	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	↓	■	■	■	↓	■	↑	■	7 / 35	
200432106																																				
200432107																																				
200432108	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	↑	■	4 / 35
200432109																																				
200432301																																				
200432302	■	■	↑	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35	
200432303																																				
200432304	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	↑	↓	■	■	■	■	■	■	■	■	↑	■	■	↑	■	■	■	■	■	■	6 / 35	
200432305																																				
200432306	↓	■	↑	■	■	■	■	■	■	↓	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	↓	■	↑	↓	8 / 35	
200432307																																				
200432308	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200432309																																				
200432310	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-5 (Page 5): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (January 2005)

Animal_ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	∑ Bilirubin	Total Protein	Urea	Summary																	
200432311																																																					
200432312	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35													
200432401	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35													
200432402	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	2	/	35												
200432403	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	↓	■	■	■	■	■	■	■	■	■	3	/	35												
200432404	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	3	/	35											
200432405	■	■	■	■	■	■	■	■	↑	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35											
200432406	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35										
200432407	■	■	■	■	■	■	■	■	■	■	■	↑	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	3	/	35										
200432408																																																					
200432501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35										
200432502																																																					
200432503	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	2	/	35									
200432504	■	■	■	■	■	■	↑	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	3	/	35								
200432505																																																					
200432506	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	2	/	35								
200432507	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35							
200432508																																																					
200432701																																																					
200432702	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35							
200432703																																																					
200432704	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35					
200432705	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35					
200432706	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	2	/	35					
200433001	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	4	/	35					
200433002	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/	35					
200433003	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35				

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-5 (Page 6): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (January 2005)

Animal_ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary			
200433004																																							
200433005																																							
200433006																																							
200433007	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	1 / 35		
200433008																																							
200433009																																							
200433010	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	2 / 35		
200433011	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35		
200433201	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200433202																																							
200433203	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	2 / 35		
200433204	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200433205	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	2 / 35	
200433206	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35		
200433207	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	↑	↓	■	■	■	■	■	■	■	■	↑	■	■	↑	■	■	■	■	■	■	■	↑	7 / 35		
200433208	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200433601	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200433602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200433603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200433604	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35		
200433605	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200433701																																							
200433702																																							
200433703																																							
200433704																																							
200433705																																							
200433706																																							

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-5 (Page 8): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (January 2005)

Animal_ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary
200434113	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35
200434114																																				
200434115																																				
200434401	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35
200434402	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35
200434403	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200434501	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	2 / 35
200434502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200434503	■	↑	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	↑	■	■	■	■	■	4 / 35
200434504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200434505																																				
200434506																																				
200434507	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	2 / 35
200434508																																				
200434509	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	1 / 35
200434510																																				
200434511	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	3 / 35
200434512	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200434601	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200434602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200434603	■	↑	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	↑	■	■	↑	■	■	■	6 / 35
200434604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	2 / 35
200434605	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200434606	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	3 / 35
200434607	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200434608	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35
200435801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-5 (Page 9): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (January 2005)

Animal ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary
200435802	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35
200435803	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200436001	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	1 / 35
200436002	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200436003	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	1 / 35
200436004	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200436005	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200436006	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200436007	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200436008	↑	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	3 / 35	
200436009	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35
200436010	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35
200436011	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	1 / 35
200436012	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	1 / 35	
200436013	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200436014																																				
200436401	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35
200436402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35
200436403	■	■	↑	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	↑	■	4 / 35
200436404	■	↑	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35
200436405	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	1 / 35
200436601	■	■	■	■	■	■	■	↓	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35
200436602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200436603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35
200436604	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	↑	■	■	■	■	■	3 / 35
200436605																																				
200436606	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-5 (Page 10): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (January 2005)

Animal_ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary	
200436607	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200436608	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35	
200436609																																					
200436610	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200436611																																					
200436612	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200436901	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200436902	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200436903	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	2 / 35	
200436904																																					
200436905	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	1 / 35	
200436906																																					
200436907																																					
200436908																																					
200436909	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	1 / 35	
200436910																																					
200436911	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200436912	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200436913	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	1 / 35
200437401	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	2 / 35
200437402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	1 / 35
200437403																																					
200437404	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200437405																																					
200437406	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35
200437407																																					
200437408	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-5 (Page 11): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (January 2005)

Animal_ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary			
200437409																																							
200437410																																							
200437411																																							
200437501	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35		
200437502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200437503																																							
200437504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35		
200437505																																							
200437506	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200437507																																							
200437508																																							
200437509	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	2 / 35		
200437510	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200437801	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	3 / 35	
200437802	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	↑	■	■	■	■	↑	■	■	■	■	■	■	■	↓	■	■	■	■	4 / 35	
200437803	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	↓	■	■	■	3 / 35	
200437804	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200438101	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35	
200438102	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200438103																																							
200438104	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35	
200438105	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200438106	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 35
200438107	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200438601																																							
200438602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 35	
200438603	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 35

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-5 (Page 12): Summary of Clinical Chemistry Data for Viagen Clone Progeny and Comparators (January 2005)

Animal_ID	A/G	Anion Gap	Globulin	Hemolysis	Icterus	Bilirubin-i	Lipemia	NaK	% Sat	Albumin-bulk	Alk Phos	ALT/P5P	Amylase	AST/P5P	hBA	Bicarbonate	Calcium	Chloride	Cholesterol	CK	Creatinine	Bilirubin-d	GGT	Glucose	Iron	Lipase	MG - XB	Phosphate	Potassium	SDH	Sodium	TIBC	Σ Bilirubin	Total Protein	Urea	Summary				
200438604	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	4	/	35	
200438605	■	■	■	■	■	■	■	■	■	■	■	↑	■	↑	■	■	■	■	■	■	■	↑	■	■	■	↑	↑	↑	■	↑	■	■	■	■	■	■	■	7	/	35
200438606	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200438607	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200438608	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
200438609	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35
200438610	■	■	■	■	■	■	■	■	■	■	■	↑	↑	↑	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	4	/	35
200438611	■	■	■	■	■	■	■	■	↓	↓	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	↓	■	↓	■	■	■	■	■	↓	■	■	■	■	6	/	35
200439501	■	■	■	■	■	■	■	■	■	■	■	↑	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35
200439502	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2	/	35
200439503	■	■	■	■	■	■	■	↓	■	■	■	↑	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	↑	↑	■	■	■	■	■	6	/	35	
200439504	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	↑	■	■	■	■	■	3	/	35	
200439505	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/	35
Summary	7	11	9	5	0	0	1	34	3	5	11	14	2	59	17	4	5	10	5	29	2	5	2	8	3	6	18	2	10	53	3	8	0	10	4					
	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/			
	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241	241			

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-6 (Page 1): Summary of Hematology Data for Viagen Clone Progeny and Comparators (January 2005)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary	
200430601																				
200430602	■	■	■	■	↑	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 18
200430603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200430604																				
200430605																				
200430606																				
200430607																				
200430608	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	↑	■	■	■	■	2 / 18
200430609																				
200430610	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	↑	■	↑	↑	■	4 / 18
200430611	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	↑	■	■	■	■	2 / 18
200430701	■	■	■	↑	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	3 / 18
200430702	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1 / 18
200430703																				
200430704																				
200430705																				
200430706																				
200430707																				
200430708																				
200430709																				
200430710																				
200430711																				
200430801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	1 / 18
200430802																				
200430803																				
200430804																				
200430805																				
200430901																				
200430902																				
200430903																				
200430904																				
200430905																				
200430906																				
200431001																				
200431002	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200431003																				
200431004	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200431005	■	■	■	↑	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	2 / 18
200431006																				
200431007	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	1 / 18

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-6 (Page 2): Summary of Hematology Data for Viagen Clone Progeny and Comparators (January 2005)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary	
200431008																				
200431009																				
200431010	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200431011																				
200431012	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200431013																				
200431401																				
200431402	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18	
200431403																				
200431404																				
200431405																				
200431406																				
200431407																				
200431408																				
200431409	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200431410	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1 / 18	
200431411																				
200431501																				
200431502																				
200431503	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200431504																				
200431505																				
200431506	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200431601	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200431602	■	■	■	■	■	↑	■	■	↓	■	■	■	■	■	■	■	↑	■	3 / 18	
200431603	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18	
200431604																				
200431605	↑	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	2 / 18	
200431606																				
200431607																				
200431608																				
200431609																				
200431610																				
200431701	↑	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	↑	■	■	3 / 18	
200431702	■	■	↑	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	2 / 18	
200431703																				
200431704																				
200431705																				
200431706																				
200431707																				

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-6 (Page 3): Summary of Hematology Data for Viagen Clone Progeny and Comparators (January 2005)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary
200431708	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432001	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432002																			
200432003	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	2 / 18
200432004																			
200432005	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432006																			
200432007	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200432008	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432101																			
200432102	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432103																			
200432104	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	↑	2 / 18
200432105																			
200432106																			
200432107																			
200432108																			
200432109																			
200432301																			
200432302																			
200432303																			
200432304	■	■	■	■	■	■	■	■	↓	■	■	■	↑	■	↓	■	■	■	3 / 18
200432305																			
200432306	■	■	■	■	■	■	■	■	↑	↓	■	■	↑	■	↓	■	■	■	4 / 18
200432307																			
200432308																			
200432309																			
200432310																			
200432311																			
200432312	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432401	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200432402																			
200432403	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432404																			
200432405																			
200432406																			
200432407	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200432408																			
200432501																			
200432502																			

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-6 (Page 4): Summary of Hematology Data for Viagen Clone Progeny and Comparators (January 2005)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary	
200432503																				
200432504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	1	/ 18
200432505																				
200432506	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	2	/ 18
200432507	■	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	2	/ 18
200432508																				
200432701																				
200432702																				
200432703																				
200432704																				
200432705																				
200432706																				
200433001	■	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	2	/ 18
200433002	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1	/ 18
200433003	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1	/ 18
200433004																				
200433005																				
200433006																				
200433007	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200433008																				
200433009																				
200433010	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	1	/ 18
200433011	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200433201																				
200433202	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200433203	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200433204	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1	/ 18
200433205																				
200433206																				
200433207																				
200433208	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200433601																				
200433602																				
200433603																				
200433604																				
200433605																				
200433701																				
200433702																				
200433703																				
200433704																				

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-6 (Page 5): Summary of Hematology Data for Viagen Clone Progeny and Comparators (January 2005)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary	
200433705																				
200433706																				
200433707																				
200433708	■	■	■	■	■	■	■	■	↑	↓	■	■	■	■	■	■	■	■	2 / 18	
200433709	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200433710																				
200433711																				
200433712	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200433801	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	↑	■	■	■	2 / 18	
200433802	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200433803																				
200433804																				
200433805																				
200433806	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	1 / 18	
200433807																				
200433808																				
200433809																				
200434101	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200434102	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	1 / 18	
200434103																				
200434104	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200434105	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200434106																				
200434107	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	↑	↑	3 / 18	
200434108																				
200434109	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200434110																				
200434111	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	1 / 18	
200434112																				
200434113	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200434114																				
200434115																				
200434401																				
200434402																				
200434403																				
200434501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200434502	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18	
200434503	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200434504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18	
200434505																				

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-6 (Page 6): Summary of Hematology Data for Viagen Clone Progeny and Comparators (January 2005)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary		
200434506																					
200434507																					
200434508																					
200434509	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	
200434510																					
200434511	■	■	■	■	■	■	■	■	↓	■	■	■	■	■	■	■	■	■	1	/ 18	
200434512																					
200434601																					
200434602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	
200434603	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/ 18	
200434604	■	■	■	■	■	↑	■	■	↑	■	■	■	■	■	■	■	■	■	2	/ 18	
200434605	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1	/ 18	
200434606	■	■	■	■	■	■	■	■	↓	■	■	■	■	↑	■	■	■	■	2	/ 18	
200434607	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	
200434608	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	
200435801	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	
200435802																					
200435803	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1	/ 18	
200436001	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	
200436002	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1	/ 18	
200436003	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	
200436004	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	
200436005	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	1	/ 18	
200436006	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	
200436007	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	
200436008	■	■	■	↑	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	3	/ 18	
200436009																					
200436010																					
200436011																					
200436012																					
200436013	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1	/ 18	
200436014																					
200436401	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	
200436402																					
200436403	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	↑	■	2	/ 18	
200436404	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	
200436405																					
200436601	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/ 18	
200436602	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	
200436603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18	

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-6 (Page 7): Summary of Hematology Data for Viagen Clone Progeny and Comparators (January 2005)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary
200436604	■	■	■	■	↑	↑	■	■	■	■	■	■	■	■	■	■	■	■	2 / 18
200436605																			
200436606	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436607	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	1 / 18
200436608	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	↑	■	3 / 18
200436609																			
200436610	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	↑	■	2 / 18
200436611																			
200436612	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	1 / 18
200436901	■	■	■	■	↑	■	■	■	■	■	■	■	■	↑	■	■	■	■	2 / 18
200436902	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436903	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200436904																			
200436905	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1 / 18
200436906																			
200436907																			
200436908																			
200436909																			
200436910																			
200436911																			
200436912	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1 / 18
200436913	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437401	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	1 / 18
200437402	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437403																			
200437404																			
200437405																			
200437406	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437407																			
200437408	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437409																			
200437410																			
200437411																			
200437501	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0 / 18
200437503																			
200437504																			
200437505																			
200437506																			
200437507																			

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-6 (Page 8): Summary of Hematology Data for Viagen Clone Progeny and Comparators (January 2005)

Animal ID	Retic abs	Basophils	Eosinophils	Hemoglobin	Hematocrit	LUC	Lymphocytes	MCH	MCHC	MCV	Monocytes	MPV	Platelets	RBC	RDW	Retic	Seg Neut	WBC	Summary	
200437508																				
200437509	■	■	■	↑	↑	■	■	■	■	■	■	■	■	↑	■	■	↑	■	4	/ 18
200437510	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	1	/ 18
200437801																				
200437802																				
200437803	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200437804	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	1	/ 18
200438101	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200438102	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200438103																				
200438104	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200438105	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200438106	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200438107																				
200438601																				
200438602	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	■	1	/ 18
200438603	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↓	■	■	■	1	/ 18
200438604	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200438605	■	■	■	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	1	/ 18
200438606	■	■	↑	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	1	/ 18
200438607	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	↑	■	2	/ 18
200438608	■	■	■	■	■	↑	■	■	■	■	■	■	■	■	■	■	↑	↑	3	/ 18
200438609	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200438610	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	1	/ 18
200438611	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	↑	↑	3	/ 18
200439501	■	■	■	■	■	↑	■	■	■	■	■	■	■	↑	↑	■	■	■	3	/ 18
200439502	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200439503	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200439504	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	0	/ 18
200439505	■	■	■	■	■	■	■	■	■	■	■	■	■	■	↑	■	■	■	1	/ 18
Summary	5 / 138	0 / 138	6 / 138	4 / 138	11 / 138	19 / 138	0 / 138	0 / 138	11 / 138	2 / 138	1 / 138	0 / 138	4 / 138	19 / 138	11 / 138	3 / 138	21 / 138	5 / 138		

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-7 (Page 1): Summary of Urinalysis Data for Viagen Clone Progeny and Comparators

Animal_ID	Treatment	Blood	pH	Protein	Blood	pH	Protein	Summary		
200430501	Control	0	8	0.3	■	■	■	0	/	3
200430502	Control	0	8	1	■	■	■	0	/	3
200430503	Control	0	7	0.1	■	↓	■	1	/	3
200430504	Control	0	7.5	0.1	■	■	■	0	/	3
200430506	Control	0	8	1	■	■	■	0	/	3
200430507	Control	0	7	1	■	↓	■	1	/	3
200430508	Control	0	6	0.3	■	↓	■	1	/	3
200430509	Control	80	8	1	↑	■	■	1	/	3
200430510	Control	0	8	3	■	■	■	0	/	3
200431801	Control	0	8	3	■	■	■	0	/	3
200431802	Control	0	8	0.1	■	■	■	0	/	3
200431803	Control	0	8	0.3	■	■	■	0	/	3
200431804	Control	0	7	3	■	↓	■	1	/	3
200431805	Control	0	8	0.1	■	■	■	0	/	3
200432601	Control	0	6.5	1	■	↓	■	1	/	3
200432602	Control	0	8	0.3	■	■	■	0	/	3
200432604	Control	10	6.5	0.3	↑	↓	■	2	/	3
200432607	Control	0	8	3	■	■	■	0	/	3
200432609	Control	0	8	0.1	■	■	■	0	/	3
200432610	Control	10	8	0.1	↑	■	■	1	/	3
200432901	Control	0	8	0.3	■	■	■	0	/	3
200432904	Control	0	8	3	■	■	■	0	/	3
200432906	Control	200	7	3	↑	↓	■	2	/	3
200432907	Control	0	8	1	■	■	■	0	/	3
200432909	Control	0	8	3	■	■	■	0	/	3
200432911	Control	0	8	0.1	■	■	■	0	/	3
200432913	Control	25	6	1	↑	↓	■	2	/	3
200433301	Control	0	8	0.1	■	■	■	0	/	3
200433302	Control	0	7.5	0.1	■	■	■	0	/	3
200433303	Control	0	8	0.3	■	■	■	0	/	3
200433306	Control	0	8	0.1	■	■	■	0	/	3
200433307	Control	0	7.5	0.1	■	■	■	0	/	3
200433401	Control	0	8	0.3	■	■	■	0	/	3
200433402	Control	0	7.5	1	■	■	■	0	/	3
200433403	Control	0	8	0.3	■	■	■	0	/	3
200433404	Control	0	8	0.3	■	■	■	0	/	3
200433405	Control	0	8	1	■	■	■	0	/	3
200433406	Control	0	7.5	1	■	■	■	0	/	3
200433408	Control	10	8	0.1	↑	■	■	1	/	3
200433409	Control	0	7.5	0.3	■	■	■	0	/	3

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-7 (Page 2): Summary of Urinalysis Data for Viagen Clone Progeny and Comparators

Animal_ID	Treatment	Blood	pH	Protein	Blood	pH	Protein	Summary		
200433501	Control	25	7.5	0.1	↑	■	■	1	/	3
200433502	Control	0	8	0.3	■	■	■	0	/	3
200433503	Control	0	7.5	0.3	■	■	■	0	/	3
200433504	Control	0	7.5	1	■	■	■	0	/	3
200433505	Control	0	8	0.3	■	■	■	0	/	3
200433506	Control	0	7.5	0.3	■	■	■	0	/	3
200433507	Control	200	7.5	1	↑	■	■	1	/	3
200433510	Control	0	7	0.1	■	↓	■	1	/	3
200433511	Control	0	8	0.1	■	■	■	0	/	3
200433512	Control	10	8	0.3	↑	■	■	1	/	3
200434001	Control	0	8	0.1	■	■	■	0	/	3
200434003	Control	0	8	3	■	■	■	0	/	3
200434004	Control	200	8	1	↑	■	■	1	/	3
200434005	Control	0	8	0	■	■	■	0	/	3
200434006	Control	200	6.5	1	↑	↓	■	2	/	3
200434007	Control	0	8	1	■	■	■	0	/	3
200434008	Control	0	7.5	3	■	■	■	0	/	3
200434009	Control	25	8	0.1	↑	■	■	1	/	3
200434201	Control	0	8.5	3	■	↑	■	1	/	3
200434202	Control	80	8	0.1	↑	■	■	1	/	3
200434203	Control	0	8	0.1	■	■	■	0	/	3
200434206	Control	10	7.5	3	↑	■	■	1	/	3
200434301	Control	0	7.5	0.1	■	■	■	0	/	3
200434302	Control	0	7.5	0.3	■	■	■	0	/	3
200434303	Control	0	8	1	■	■	■	0	/	3
200434304	Control	0	6.5	0.3	■	↓	■	1	/	3
200434306	Control	0	7.5	0.3	■	■	■	0	/	3
200434310	Control	10	8	3	↑	■	■	1	/	3
200434802	Control	0	8	1	■	■	■	0	/	3
200434803	Control	0	7.5	0.3	■	■	■	0	/	3
200434804	Control	0	8	1	■	■	■	0	/	3
200434805	Control	0	7.5	1	■	■	■	0	/	3
200434806	Control	0	8	0.1	■	■	■	0	/	3
200434807	Control	0	8	0.1	■	■	■	0	/	3
200434808	Control	0	8	1	■	■	■	0	/	3
200434809	Control	0	8	1	■	■	■	0	/	3
200434901	Control	10	8	0.3	↑	■	■	1	/	3
200434902	Control	0	8	3	■	■	■	0	/	3
200434903	Control	0	8	0.3	■	■	■	0	/	3
200434904	Control	0	8	3	■	■	■	0	/	3

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-7 (Page 3): Summary of Urinalysis Data for Viagen Clone Progeny and Comparators

Animal_ID	Treatment	Blood	pH	Protein	Blood	pH	Protein	Summary		
200434905	Control	25	7	3	↑	↓	■	2	/	3
200435301	Control	0	8	0.3	■	■	■	0	/	3
200435302	Control	0	8	0.1	■	■	■	0	/	3
200435303	Control	0	8	0.1	■	■	■	0	/	3
200435304	Control	0	8	3	■	■	■	0	/	3
200435305	Control	80	6	0.3	↑	↓	■	2	/	3
200435306	Control	0	8	1	■	■	■	0	/	3
200435307	Control	10	8	0.3	↑	■	■	1	/	3
200435308	Control	10	7.5	0.1	↑	■	■	1	/	3
200435309	Control	0	7	0.3	■	↓	■	1	/	3
200435310	Control	10	7.5	0.3	↑	■	■	1	/	3
200435311	Control	200	7.5	0.3	↑	■	■	1	/	3
200435401	Control	0	8	1	■	■	■	0	/	3
200435402	Control	0	8	1	■	■	■	0	/	3
200435403	Control	0	8	1	■	■	■	0	/	3
200435404	Control	0	8	0.1	■	■	■	0	/	3
200435405	Control	10	6.5	1	↑	↓	■	2	/	3
200435407	Control	0	8	0.3	■	■	■	0	/	3
200435409	Control	200	8	1	↑	■	■	1	/	3
200435410	Control	0	6	0.1	■	↓	■	1	/	3
200435411	Control	10	7	0.3	↑	↓	■	2	/	3
200435412	Control	10	8	3	↑	■	■	1	/	3
200435413	Control	10	8	3	↑	■	■	1	/	3
200436301	Control	0	7	0.1	■	↓	■	1	/	3
200436303	Control	0	8	0.3	■	■	■	0	/	3
200436305	Control	0	7.5	0.1	■	■	■	0	/	3
200436309	Control	0	8	3	■	■	■	0	/	3
200436310	Control	0	6	1	■	↓	■	1	/	3
200436312	Control	0	7.5	3	■	■	■	0	/	3
200437301	Control	0	7.5	0.3	■	■	■	0	/	3
200437302	Control	80	6.5	1	↑	↓	■	2	/	3
200437303	Control	0	8	0.1	■	■	■	0	/	3
200437305	Control	0	8	0.1	■	■	■	0	/	3
200437307	Control	0	8	0.3	■	■	■	0	/	3
200437309	Control	0	8	0.3	■	■	■	0	/	3
200438401	Control	0	8	1	■	■	■	0	/	3
200438402	Control	10	7.5	1	↑	■	■	1	/	3
200438403	Control	0	8	0.1	■	■	■	0	/	3
200438404	Control	10	8	0.3	↑	■	■	1	/	3
200438406	Control	10	8	1	↑	■	■	1	/	3

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-7 (Page 4): Summary of Urinalysis Data for Viagen Clone Progeny and Comparators

Animal_ID	Treatment	Blood	pH	Protein	Blood	pH	Protein	Summary	
200438408	Control	10	8	3	↑	■	■	1	/ 3
200438501	Control	0	7.5	0.3	■	■	■	0	/ 3
200438503	Control	0	7	3	■	↓	■	1	/ 3
200438505	Control	0	8	3	■	■	■	0	/ 3
200438511	Control	0	8	1	■	■	■	0	/ 3
200438513	Control	0	8	3	■	■	■	0	/ 3
200438701	Control	0	7.5	0.3	■	■	■	0	/ 3
200438703	Control	0	7	0.1	■	↓	■	1	/ 3
200438705	Control	0	8	3	■	■	■	0	/ 3
200438706	Control	0	8	0.3	■	■	■	0	/ 3
200438707	Control	0	8	0.1	■	■	■	0	/ 3
200438709	Control	0	7.5	0.1	■	■	■	0	/ 3
200439202	Control	0	8	0.3	■	■	■	0	/ 3
200439204	Control	0	7.5	3	■	■	■	0	/ 3
200439205	Control	0	8	0.3	■	■	■	0	/ 3
200439206	Control	10	7.5	3	↑	■	■	1	/ 3
200439208	Control	200	7.5	1	↑	■	■	1	/ 3
200439210	Control	0	8	0.3	■	■	■	0	/ 3
200439401	Control	0	6	1	■	↓	■	1	/ 3
200439404	Control	0	6.5	3	■	↓	■	1	/ 3
200439406	Control	0	8	3	■	■	■	0	/ 3
200439901	Control	10	8	0.1	↑	■	■	1	/ 3
200439902	Control	0	7	0.1	■	↓	■	1	/ 3
200439903	Control	200	7.5	0.1	↑	■	■	1	/ 3
200439904	Control	0	7.5	1	■	■	■	0	/ 3
200440601	Control	0	6.5	1	■	↓	■	1	/ 3
200440602	Control	10	6	1	↑	↓	■	2	/ 3
200440603	Control	0	7	1	■	↓	■	1	/ 3
200440605	Control	0	8	0.3	■	■	■	0	/ 3
200440901	Control	0	8	0.3	■	■	■	0	/ 3
200440903	Control	80	8	3	↑	■	■	1	/ 3
200440906	Control	0	7.5	0.3	■	■	■	0	/ 3
200440907	Control	80	6.5	3	↑	↓	■	2	/ 3
200440909	Control	0	6	1	■	↓	■	1	/ 3
200440911	Control	10	6	1	↑	↓	■	2	/ 3
200441201	Control	0	6.5	1	■	↓	■	1	/ 3
200441202	Control	0	7.5	1	■	■	■	0	/ 3
200441203	Control	0	6	0.3	■	↓	■	1	/ 3
200441205	Control	10	6	0.3	↑	↓	■	2	/ 3
200441207	Control	0	6	0.3	■	↓	■	1	/ 3

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-7 (Page 5): Summary of Urinalysis Data for Viagen Clone Progeny and Comparators

Animal_ID	Treatment	Blood	pH	Protein	Blood	pH	Protein	Summary		
200441209	Control	0	6	0.3	■	↓	■	1	/	3
200441211	Control	0	7.5	0.3	■	■	■	0	/	3
200430601										
200430602	Clone	0	8	1	■	■	■	0	/	3
200430603	Clone	0	8.5	1	■	↑	■	1	/	3
200430604	Clone	10	8	0.1	↑	■	■	1	/	3
200430605	Clone	0	7.5	0.1	■	■	■	0	/	3
200430606	Clone	10	7.5	0.3	↑	■	■	1	/	3
200430607	Clone	200	8	0.1	↑	■	■	1	/	3
200430608	Clone	10	8	0.3	↑	■	■	1	/	3
200430609	Clone	0	8	0.1	■	■	■	0	/	3
200430610	Clone	80	7.5	0.3	↑	■	■	1	/	3
200430611	Clone	10	6	0.1	↑	↓	■	2	/	3
200430701	Clone	0	7	1	■	↓	■	1	/	3
200430702	Clone	0	8	0.3	■	■	■	0	/	3
200430703	Clone	0	8	1	■	■	■	0	/	3
200430704	Clone	0	8	3	■	■	■	0	/	3
200430705	Clone	0	8	0.1	■	■	■	0	/	3
200430706	Clone	10	7	0.1	↑	↓	■	2	/	3
200430707	Clone	0	8	0.1	■	■	■	0	/	3
200430708	Clone	0	8	0.1	■	■	■	0	/	3
200430709	Clone	0	8	0.1	■	■	■	0	/	3
200430710	Clone	10	7.5	0.1	↑	■	■	1	/	3
200430711	Clone	0	8	0.3	■	■	■	0	/	3
200430801	Clone	0	7	3	■	↓	■	1	/	3
200430802	Clone	0	8	1	■	■	■	0	/	3
200430803	Clone	10	7.5	0.1	↑	■	■	1	/	3
200430804	Clone	10	7.5	0	↑	■	■	1	/	3
200430805	Clone	0	7.5	0.1	■	■	■	0	/	3
200430901										
200430902	Clone	0	6.5	0	■	↓	■	1	/	3
200430903	Clone	0	7	0.3	■	↓	■	1	/	3
200430904										
200430905	Clone	0	7.5	0.1	■	■	■	0	/	3
200430906	Clone	0	7	0.3	■	↓	■	1	/	3
200431001										
200431002	Clone	0	7.5	0.3	■	■	■	0	/	3
200431003	Clone	0	8	0.1	■	■	■	0	/	3
200431004	Clone	0	8	0.3	■	■	■	0	/	3
200431005	Clone	0	8	0.1	■	■	■	0	/	3

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-7 (Page 6): Summary of Urinalysis Data for Viagen Clone Progeny and Comparators

Animal_ID	Treatment	Blood	pH	Protein	Blood	pH	Protein	Summary		
200431006	Clone	0	6.5	0.1	■	↓	■	1	/	3
200431007	Clone	80	8	0	↑	■	■	1	/	3
200431008	Clone	0	7.5	0.1	■	■	■	0	/	3
200431009	Clone	10	7.5	0.1	↑	■	■	1	/	3
200431010	Clone	0	8	1	■	■	■	0	/	3
200431011	Clone	0	8	3	■	■	■	0	/	3
200431012	Clone	25	7.5	0.3	↑	■	■	1	/	3
200431013										
200431401	Clone	0	8	0.1	■	■	■	0	/	3
200431402	Clone	0	8	0.3	■	■	■	0	/	3
200431403	Clone	0	8	1	■	■	■	0	/	3
200431404	Clone	0	7.5	0.1	■	■	■	0	/	3
200431405	Clone	0	7.5	0.1	■	■	■	0	/	3
200431406	Clone	0	8	0.1	■	■	■	0	/	3
200431407	Clone	0	8	0.1	■	■	■	0	/	3
200431408	Clone	0	6.5	0.1	■	↓	■	1	/	3
200431409	Clone	0	8	0.1	■	■	■	0	/	3
200431410	Clone	10	7.5	0.1	↑	■	■	1	/	3
200431411	Clone	0	7.5	0.1	■	■	■	0	/	3
200431501	Clone	0	7.5	0.1	■	■	■	0	/	3
200431502	Clone	0	8	0	■	■	■	0	/	3
200431503	Clone	0	7	0.1	■	↓	■	1	/	3
200431504	Clone	0	8	0.1	■	■	■	0	/	3
200431505	Clone	0	6.5	0.1	■	↓	■	1	/	3
200431506	Clone	0	8	0.3	■	■	■	0	/	3
200431601	Clone	0	8	1	■	■	■	0	/	3
200431602	Clone	0	8	0.3	■	■	■	0	/	3
200431603	Clone	0	8	1	■	■	■	0	/	3
200431604	Clone	0	8	0.3	■	■	■	0	/	3
200431605	Clone	0	8	0.3	■	■	■	0	/	3
200431606	Clone	0	8	0.1	■	■	■	0	/	3
200431607	Clone	0	8	0.3	■	■	■	0	/	3
200431608	Clone	0	7.5	0	■	■	■	0	/	3
200431609										
200431610	Clone	10	8	0.3	↑	■	■	1	/	3
200431701	Clone	0	8	0.3	■	■	■	0	/	3
200431702	Clone	0	8	0.1	■	■	■	0	/	3
200431703	Clone	80	8	0.3	↑	■	■	1	/	3
200431704	Clone	80	8	0.1	↑	■	■	1	/	3
200431705										

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-7 (Page 7): Summary of Urinalysis Data for Viagen Clone Progeny and Comparators

Animal_ID	Treatment	Blood	pH	Protein	Blood	pH	Protein	Summary
200431706	Clone	0	8	3	■	■	■	0 / 3
200431707								
200431708	Clone	200	8	3	↑	■	■	1 / 3
200432001	Clone	0	8	1	■	■	■	0 / 3
200432002	Clone	0	8	3	■	■	■	0 / 3
200432003	Clone	0	7	0.3	■	↓	■	1 / 3
200432004	Clone	0	8	0.3	■	■	■	0 / 3
200432005	Clone	80	8	0.1	↑	■	■	1 / 3
200432006	Clone	10	8	0.1	↑	■	■	1 / 3
200432007	Clone	0	8	1	■	■	■	0 / 3
200432008	Clone	0	7.5	0.3	■	■	■	0 / 3
200432101	Clone	0	6	0	■	↓	■	1 / 3
200432102	Clone	0	8	1	■	■	■	0 / 3
200432103	Clone	0	7.5	0.3	■	■	■	0 / 3
200432104	Clone	10	8	0.3	↑	■	■	1 / 3
200432105	Clone	0	8	0.3	■	■	■	0 / 3
200432106								
200432107								
200432108	Clone	0	8	3	■	■	■	0 / 3
200432109								
200432301								
200432302	Clone	10	8	0.1	↑	■	■	1 / 3
200432303								
200432304	Clone	0	6.5	0.1	■	↓	■	1 / 3
200432305								
200432306	Clone	0	7.5	0.3	■	■	■	0 / 3
200432307								
200432308	Clone	200	8	1	↑	■	■	1 / 3
200432309								
200432310	Clone	0	8	1	■	■	■	0 / 3
200432311								
200432312	Clone	200	6	3	↑	↓	■	2 / 3
200432401	Clone	10	8	3	↑	■	■	1 / 3
200432402	Clone	0	8	0.3	■	■	■	0 / 3
200432403	Clone	0	8	1	■	■	■	0 / 3
200432404	Clone	0	7.5	0.3	■	■	■	0 / 3
200432405	Clone	0	7.5	0.1	■	■	■	0 / 3
200432406	Clone	0	8	0.3	■	■	■	0 / 3
200432407	Clone	0	8	1	■	■	■	0 / 3
200432408								

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-7 (Page 8): Summary of Urinalysis Data for Viagen Clone Progeny and Comparators

Animal_ID	Treatment	Blood	pH	Protein	Blood	pH	Protein	Summary
200432501	Clone	0	7.5	0.1	■	■	■	0 / 3
200432502								
200432503	Clone	0	7.5	0.1	■	■	■	0 / 3
200432504	Clone	0	8	0.3	■	■	■	0 / 3
200432505								
200432506	Clone	0	8	0.3	■	■	■	0 / 3
200432507	Clone	0	7.5	0.3	■	■	■	0 / 3
200432508								
200432701								
200432702	Clone	0	8	0.3	■	■	■	0 / 3
200432703								
200432704	Clone	10	8	0.3	↑	■	■	1 / 3
200432705	Clone	10	8	0.3	↑	■	■	1 / 3
200432706	Clone	0	8	0.1	■	■	■	0 / 3
200433001	Clone	0	8	0.3	■	■	■	0 / 3
200433002	Clone	0	8	0.3	■	■	■	0 / 3
200433003	Clone	0	7.5	0.3	■	■	■	0 / 3
200433004								
200433005								
200433006								
200433007	Clone	0	8	0.3	■	■	■	0 / 3
200433008								
200433009								
200433010	Clone	10	8	0.1	↑	■	■	1 / 3
200433011	Clone	10	6	0.3	↑	↓	■	2 / 3
200433201	Clone	0	8	0.1	■	■	■	0 / 3
200433202	Clone	0	8	0.1	■	■	■	0 / 3
200433203	Clone	0	6	0.1	■	↓	■	1 / 3
200433204	Clone	0	8	1	■	■	■	0 / 3
200433205	Clone	10	8	1	↑	■	■	1 / 3
200433206	Clone	0	7.5	0.1	■	■	■	0 / 3
200433207	Clone	10	8	0.1	↑	■	■	1 / 3
200433208	Clone	10	8	1	↑	■	■	1 / 3
200433601	Clone	0	8	0.1	■	■	■	0 / 3
200433602	Clone	0	8	1	■	■	■	0 / 3
200433603	Clone	0	8	0.1	■	■	■	0 / 3
200433604	Clone	0	8	0.1	■	■	■	0 / 3
200433605	Clone	0	8	0.1	■	■	■	0 / 3
200433701								
200433702								

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-7 (Page 9): Summary of Urinalysis Data for Viagen Clone Progeny and Comparators

Animal_ID	Treatment	Blood	pH	Protein	Blood	pH	Protein	Summary
200433703								
200433704								
200433705								
200433706								
200433707								
200433708	Clone	0	8.5	0.3	■	↑	■	1 / 3
200433709	Clone	0	6	0.1	■	↓	■	1 / 3
200433710	Clone	10	7.5	0.1	↑	■	■	1 / 3
200433711	Clone	80	8	0.3	↑	■	■	1 / 3
200433712	Clone	0	8	0.1	■	■	■	0 / 3
200433801	Clone	0	7	0.3	■	↓	■	1 / 3
200433802	Clone	10	7	3	↑	↓	■	2 / 3
200433803	Clone	0	8	3	■	■	■	0 / 3
200433804	Clone	0	8.5	0.3	■	↑	■	1 / 3
200433805								
200433806	Clone	10	6.5	0.3	↑	↓	■	2 / 3
200433807	Clone	0	7.5	0.1	■	■	■	0 / 3
200433808	Clone	10	8	0.1	↑	■	■	1 / 3
200433809	Clone	0	8	1	■	■	■	0 / 3
200434101	Clone	0	7.5	0.1	■	■	■	0 / 3
200434102	Clone	0	8	0.1	■	■	■	0 / 3
200434103								
200434104	Clone	0	8	0.3	■	■	■	0 / 3
200434105	Clone	0	8	0.1	■	■	■	0 / 3
200434106								
200434107	Clone	0	8	0.1	■	■	■	0 / 3
200434108								
200434109	Clone	0	8	0.3	■	■	■	0 / 3
200434110								
200434111	Clone	0	7.5	0.3	■	■	■	0 / 3
200434112								
200434113	Clone	0	8	20	■	■	↑	1 / 3
200434114								
200434115								
200434401	Clone	0	8	1	■	■	■	0 / 3
200434402	Clone	0	8	1	■	■	■	0 / 3
200434403	Clone	0	8	0.3	■	■	■	0 / 3
200434501	Clone	0	8	1	■	■	■	0 / 3
200434502	Clone	0	8	0.1	■	■	■	0 / 3
200434503	Clone	0	6	0.1	■	↓	■	1 / 3

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-7 (Page 10): Summary of Urinalysis Data for Viagen Clone Progeny and Comparators

Animal_ID	Treatment	Blood	pH	Protein	Blood	pH	Protein	Summary		
200434504	Clone	0	8	0.3	■	■	■	0	/	3
200434505										
200434506										
200434507	Clone	80	6	0.1	↑	↓	■	2	/	3
200434508										
200434509	Clone	25	7	0.3	↑	↓	■	2	/	3
200434510										
200434511	Clone	0	7	0.1	■	↓	■	1	/	3
200434512	Clone	0	8	1	■	■	■	0	/	3
200434601	Clone	0	8	0.3	■	■	■	0	/	3
200434602	Clone	0	8	1	■	■	■	0	/	3
200434603	Clone	0	8	3	■	■	■	0	/	3
200434604	Clone	0	6.5	0.3	■	↓	■	1	/	3
200434605	Clone	0	8	1	■	■	■	0	/	3
200434606	Clone	0	8	0.3	■	■	■	0	/	3
200434607	Clone	0	7	0.3	■	↓	■	1	/	3
200434608	Clone	10	8	1	↑	■	■	1	/	3
200435801	Clone	0	7.5	0.1	■	■	■	0	/	3
200435802	Clone	10	8.5	0.1	↑	↑	■	2	/	3
200435803	Clone	200	8	1	↑	■	■	1	/	3
200436001	Clone	0	8	0.3	■	■	■	0	/	3
200436002	Clone	0	7.5	0.3	■	■	■	0	/	3
200436003	Clone	0	8	3	■	■	■	0	/	3
200436004	Clone	0	7.5	0.1	■	■	■	0	/	3
200436005	Clone	80	8	0.3	↑	■	■	1	/	3
200436006	Clone	25	8	3	↑	■	■	1	/	3
200436007	Clone	0	8	0.3	■	■	■	0	/	3
200436008	Clone	0	8	0.3	■	■	■	0	/	3
200436009	Clone	0	7.5	0.1	■	■	■	0	/	3
200436010	Clone	0	8	0.1	■	■	■	0	/	3
200436011	Clone	0	8	0.1	■	■	■	0	/	3
200436012	Clone	0	7.5	0.1	■	■	■	0	/	3
200436013	Clone	0	6.5	0.3	■	↓	■	1	/	3
200436014										
200436401	Clone	0	8	3	■	■	■	0	/	3
200436402	Clone	0	7.5	0.3	■	■	■	0	/	3
200436403	Clone	0	8	1	■	■	■	0	/	3
200436404	Clone	0	8	0.1	■	■	■	0	/	3
200436405	Clone	0	8	0.3	■	■	■	0	/	3
200436601	Clone	0	8	0.3	■	■	■	0	/	3

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-7 (Page 11): Summary of Urinalysis Data for Viagen Clone Progeny and Comparators

Animal_ID	Treatment	Blood	pH	Protein	Blood	pH	Protein	Summary	
200436602	Clone	0	8	3	■	■	■	0	/ 3
200436603	Clone	0	7.5	0.1	■	■	■	0	/ 3
200436604	Clone	0	7.5	3	■	■	■	0	/ 3
200436605									
200436606	Clone	0	7.5	0.3	■	■	■	0	/ 3
200436607	Clone	200	7.5	1	↑	■	■	1	/ 3
200436608	Clone	0	6.5	0.1	■	↓	■	1	/ 3
200436609									
200436610	Clone	0	7	0.1	■	↓	■	1	/ 3
200436611									
200436612	Clone	0	8	1	■	■	■	0	/ 3
200436901	Clone	0	8	0.1	■	■	■	0	/ 3
200436902	Clone	0	7.5	0.1	■	■	■	0	/ 3
200436903	Clone	0	7.5	3	■	■	■	0	/ 3
200436904									
200436905	Clone	0	8	0.3	■	■	■	0	/ 3
200436906									
200436907									
200436908									
200436909	Clone	0	8	0.1	■	■	■	0	/ 3
200436910									
200436911	Clone	0	8	0.1	■	■	■	0	/ 3
200436912	Clone	0	6	0.1	■	↓	■	1	/ 3
200436913	Clone	10	8	1	↑	■	■	1	/ 3
200437401	Clone	0	7.5	0.1	■	■	■	0	/ 3
200437402	Clone	0	8	1	■	■	■	0	/ 3
200437403									
200437404	Clone	0	8	1	■	■	■	0	/ 3
200437405									
200437406	Clone	0	8	0.3	■	■	■	0	/ 3
200437407									
200437408	Clone	0	7	1	■	↓	■	1	/ 3
200437409									
200437410									
200437411									
200437501	Clone	80	8	1	↑	■	■	1	/ 3
200437502	Clone	0	8	1	■	■	■	0	/ 3
200437503									
200437504	Clone	0	8	1	■	■	■	0	/ 3
200437505									

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

Chart F-7 (Page 12): Summary of Urinalysis Data for Viagen Clone Progeny and Comparators

Animal_ID	Treatment	Blood	pH	Protein	Blood	pH	Protein	Summary		
200437506	Clone	10	7.5	0.1	↑	■	■	1	/	3
200437507										
200437508										
200437509	Clone	200	8	1	↑	■	■	1	/	3
200437510	Clone	25	7.5	0.3	↑	■	■	1	/	3
200437801	Clone	0	8	3	■	■	■	0	/	3
200437802	Clone	0	6.5	0.1	■	↓	■	1	/	3
200437803	Clone	0	7	0.3	■	↓	■	1	/	3
200437804	Clone	10	8	0.3	↑	■	■	1	/	3
200438101	Clone	0	8	0.3	■	■	■	0	/	3
200438102	Clone	0	8	0.3	■	■	■	0	/	3
200438103										
200438104	Clone	0	8	1	■	■	■	0	/	3
200438105	Clone	0	8	1	■	■	■	0	/	3
200438106	Clone	0	8	0.1	■	■	■	0	/	3
200438107	Clone	0	7.5	0.1	■	■	■	0	/	3
200438601										
200438602	Clone	0	7.5	0.3	■	■	■	0	/	3
200438603	Clone	0	7	0.1	■	↓	■	1	/	3
200438604	Clone	0	8	1	■	■	■	0	/	3
200438605	Clone	0	8	1	■	■	■	0	/	3
200438606	Clone	0	7.5	0.1	■	■	■	0	/	3
200438607	Clone	0	8	0.1	■	■	■	0	/	3
200438608	Clone	0	7.5	1	■	■	■	0	/	3
200438609	Clone	0	6.5	0.1	■	↓	■	1	/	3
200438610	Clone	0	6.5	0.1	■	↓	■	1	/	3
200438611	Clone	0	8	0.3	■	■	■	0	/	3
200439501	Clone	0	7.5	0.3	■	■	■	0	/	3
200439502	Clone	10	6	1	↑	↓	■	2	/	3
200439503	Clone	0	8	0.1	■	■	■	0	/	3
200439504	Clone	0	7.5	0.3	■	■	■	0	/	3
200439505	Clone	0	7.5	0.3	■	■	■	0	/	3

■ In Comparison Range
 ↑/↓ Animal is above/below reference range
 Blank rows indicate values that were missing

ViaGen Raw Data

Data in white space indicates Trial 1; Data in gray space indicates Trial 2

Sunday	Monday	Tuesday	Wednesday	Thursday	Friday	Saturday
9-Nov-03	10-Nov-03	11-Nov-03	12-Nov-03	13-Nov-03	14-Nov-03	15-Nov-03
16-Nov-03	17-Nov-03	18-Nov-03	19-Nov-03	20-Nov-03	21-Nov-03 Bi-weekly weighing Castration of clones	22-Nov-03
23-Nov-03	24-Nov-03	25-Nov-03	26-Nov-03	27-Nov-03	28-Nov-03	29-Nov-03
30-Nov-03 Bi-weekly weighing	1-Dec-03 Clones vaccinated	2-Dec-03	3-Dec-03	4-Dec-03 Initial urine collection Controls vaccinated	5-Dec-03 Initial blood collection Monthly vet exam	6-Dec-03
7-Dec-03	8-Dec-03	9-Dec-03	10-Dec-03	11-Dec-03	12-Dec-03 Bi-weekly weighing	13-Dec-03

Data in white space indicates Trial 1; Data in gray space indicates Trial 2

Sunday	Monday	Tuesday	Wednesday	Thursday	Friday	Saturday
14-Dec-03	15-Dec-03	16-Dec-03	17-Dec-03	18-Dec-03	19-Dec-03	20-Dec-03
21-Dec-03	22-Dec-03	23-Dec-03	24-Dec-03	25-Dec-03	26-Dec-03	27-Dec-03
28-Dec-03	29-Dec-03	30-Dec-03	31-Dec-03	1-Jan-04	2-Jan-04	3-Jan-04
4-Jan-04	5-Jan-04	6-Jan-04	7-Jan-04	8-Jan-04	9-Jan-04 Bi-weekly weighing	10-Jan-04
11-Jan-04	12-Jan-04	13-Jan-04	14-Jan-04	15-Jan-04	16-Jan-04	17-Jan-04
					Initial urine collection Initial blood collection Bi-weekly weighing	

Data in white space indicates Trial 1; Data in gray space indicates Trial 2

Sunday	Monday	Tuesday	Wednesday	Thursday	Friday	Saturday
18-Jan-04	19-Jan-04	20-Jan-04	21-Jan-04	22-Jan-04	23-Jan-04 Middle urine collection Middle blood collection Monthly vet exam Bi-weekly weighing Monthly vet exam	24-Jan-04
25-Jan-04	26-Jan-04	27-Jan-04	28-Jan-04	29-Jan-04	30-Jan-04 Bi-weekly weighing	31-Jan-04
1-Feb-04	2-Feb-04	3-Feb-04	4-Feb-04	5-Feb-04	6-Feb-04	7-Feb-04
8-Feb-04	9-Feb-04	10-Feb-04	11-Feb-04	12-Feb-04	13-Feb-04 Bi-weekly weighing	14-Feb-04
15-Feb-04	16-Feb-04	17-Feb-04	18-Feb-04	19-Feb-04	20-Feb-04 Bi-weekly weighing	21-Feb-04

Data in white space indicates Trial 1; Data in gray space indicates Trial 2

Sunday	Monday	Tuesday	Wednesday	Thursday	Friday	Saturday
22-Feb-04	23-Feb-04	24-Feb-04 Monthly behavioral observation	25-Feb-04	26-Feb-04	27-Feb-04	28-Feb-04
		Monthly behavioral observations			Bi-weekly weighing	
29-Feb-04	1-Mar-04	2-Mar-04	3-Mar-04	4-Mar-04	5-Mar-04 Bi-weekly weighing	6-Mar-04
7-Mar-04	8-Mar-04	9-Mar-04	10-Mar-04	11-Mar-04	12-Mar-04	13-Mar-04
					Bi-weekly weighing	
14-Mar-04	15-Mar-04	16-Mar-04 Monthly behavioral observations Monthly Vet Exam	17-Mar-04	18-Mar-04	19-Mar-04 Bi-weekly weighing	20-Mar-04
		Middle urine collection Middle blood collection Monthly Vet Exam				

Data in white space indicates Trial 1; Data in gray space indicates Trial 2

Sunday	Monday	Tuesday	Wednesday	Thursday	Friday	Saturday
21-Mar-04	22-Mar-04	23-Mar-04 Final weighing (n=7 animals for slaughter on 25-Mar-04)	24-Mar-04	25-Mar-04 Final urine collection (n=7) Final blood collection (n=7) Slaughter of 7 animals (025413, 025461, 025436, 025462, 025415, 025457, and 025416)	26-Mar-04	27-Mar-04
					Bi-weekly weighing	
28-Mar-04	29-Mar-04	30-Mar-04	31-Mar-04	1-Apr-04	2-Apr-04	3-Apr-04
		Monthly behavioral observation	Monthly Vet Exam		Bi-weekly weighing (n=8)	
4-Apr-04	5-Apr-04	6-Apr-04	7-Apr-04	8-Apr-04	9-Apr-04	10-Apr-04
		Final weighing (n=6 animals for slaughter on 8-Apr-04) Bi-weekly weighing (n=2)		Final urine collection (n=8) Final blood collection (n=8) Slaughter of 6 animals (025437, 025456, 025463, 018, 019, 020)	Bi-weekly weighing	

Data in white space indicates Trial 1; Data in gray space indicates Trial 2

Sunday 11-Apr-04	Monday 12-Apr-04	Tuesday 13-Apr-04	Wednesday 14-Apr-04	Thursday 15-Apr-04	Friday 16-Apr-04	Saturday 17-Apr-04
					Monthly Vet Exam (n=2)	
					Monthly Vet Exam	
18-Apr-04	19-Apr-04 Bi-weekly weighing (n=2)	20-Apr-04	21-Apr-04	22-Apr-04	23-Apr-04 Bi-weekly weighing (n=2)	24-Apr-04
	Final weighing (n=2 animals for slaughter on 20-Apr-04) Bi-weekly weighing (n=5)	Slaughter of 2 animals (044615, 044662) Final blood collection (n=2) Final urine collection (n=2)			Bi-weekly weighing (n=5)	
25-Apr-04	26-Apr-04	27-Apr-04	28-Apr-04	29-Apr-04	30-Apr-04	1-May-04
		observations				
2-May-04	3-May-04	4-May-04	5-May-04	6-May-04	7-May-04 Bi-weekly weighing (n=2)	8-May-04
					Bi-weekly weighing (n=5)	

Data in white space indicates Trial 1; Data in gray space indicates Trial 2

Sunday	Monday	Tuesday	Wednesday	Thursday	Friday	Saturday
9-May-04	10-May-04 Bi-weekly weighing (n=2)	11-May-04	12-May-04	13-May-04	14-May-04	15-May-04
	Final weighing (n=1 animal for slaughter on 11-May-04) Bi-weekly weighing (n=4)	Slaughter of 1 animal (044661) Final blood collection (n=1) Final urine collection (n=1)				
16-May-04	17-May-04 Monthly Vet Exam (n=2)	18-May-04	19-May-04	20-May-04	21-May-04 Bi-weekly weighing (n=2)	22-May-04
	Monthly vet exam (n=4)				Bi-weekly weighing (n=4)	
23-May-04	24-May-04	25-May-04	26-May-04	27-May-04	28-May-04	29-May-04
			Final weighing (n=2 animals for euthanasia on 27-May-04)	Euthanize (n=2 remaining animals on trial 1 [021,022]) Blood collection (n=2) Urine collection (n=2)		
			Final weighing (n=4 animals for slaughter on 27-May-04)	Slaughter (n=4 remaining animals on trial 2 [023, 024, 027446,044584]) Final blood collection (n=4) Final urine collection (n=4)		

Barrow ID - Daily Health Record															
Date	018	019	020	022	025456	025463	025413	025461	025435	025436	025462	025415	025457	025416	025437
18-Nov-03									Weak, scours, lame on left hind leg - received 0.4ml Naxcel IM, 0.5ml Dexamethasone IM, 1 bowl pedalyte, 39mg Neomycin						
19-Nov-03									Alert, scours, placing more weight on leg - received 0.4ml Naxcel IM, 0.25ml Banamine IM, 39mg Neomycin in water 2 times daily						
20-Nov-03									Scours improving, eating and drinking, still sore on left hind leg - received 0.4ml Naxcel IM, 0.25ml Banamine IM, 39 mg Neomycin in water 2 times daily						
21-Nov-03	castrated - 6:30 pm bleeding and exposure/partial strangulation of intestines noted - applied Procaine and Pen G to exposed intestine - preformed surgery to repair damage - received 3ml Penicillin IM and 0.5ml Banamine IM	castrated - received 3ml Penicillin IM	castrated - received 3ml Penicillin IM	castrated - received 1.5ml Penicillin					Scours improving, eating and drinking, still sore on left hind leg - received 0.4ml Naxcel IM, 39 mg Neomycin in water 2 times daily						
22-Nov-03	moving around, urinating, drinking and eating - received 0.5ml Banamine IM and 0.7ml Naxcel IM	received 0.6ml Naxcel	received 0.6ml Naxcel	received 0.58ml Naxcel IM					Scours gone, eating - received Neomycin in water						
23-Nov-03	BAR: still weak but is eating more and drinking - received 0.5ml Banamine IM and 0.7ml Naxcel IM	BAR - received 0.6ml Naxcel	BAR - received 0.6ml Naxcel	BAR - received 0.58ml Naxcel IM					Scours returned, eating a little - received 39mg Neomycin in water in AM and PM						
24-Nov-03	temp: 10.9 - scours - received 0.5ml Banamine IM and 0.7ml Naxcel IM	BAR	BAR	BAR - received 0.5ml Naxcel IM					Scours gone, improving - received 39mg Neomycin in water AM and PM						
25-Nov-03	BAR: some swelling but Dr. Reeves checked incision and feels swelling is just fluid - received 0.7ml Naxcel - blue lotion wound spray applied to site								Scours returned (Dr. Reeves thinks it may be a protozoa) - received 39mg Neomycin in water AM and PM and 2 cans Ensure in PM						
26-Nov-03	BAR: eating, drinking, active - received 0.7ml Naxcel IM - blue lotion wound spray applied to site								Scours, eating and drinking, up walking around - received 39mg Neomycin in water and 2 cans Ensure						
27-Nov-03	eating and drinking - very active - received 0.7ml Naxcel IM - blue lotion wound spray applied to site								Bright, walking around and drinking - received Neomycin in water and 2 cans Ensure						
28-Nov-03	BAR - received 0.7ml Naxcel IM								Scours gone, bright and alert - received Neomycin in water and 2 cans Ensure						
29-Nov-03									Ate some pellets, coat looks better - received Neomycin in water						
30-Nov-03	BAR								Scours gone, bright						
1-Dec-03	BAR - received 1ml myco silencer BPME vaccine IM	BAR - received 1ml myco silencer BPME vaccine IM	BAR - received 1ml myco silencer BPME vaccine IM	BAR - received 1ml myco silencer BPME vaccine IM	BAR	BAR	BAR	BAR	Weak, breathing heavily, 1:30am open mouth breathing - cleared discharge from nostrils, received Ensure every 4 hours	BAR	BAR	BAR	BAR	BAR	BAR
2-Dec-03	BAR: scours - received kapectate PO	BAR: scours - received kapectate PO	BAR: scours - received kapectate PO	BAR: scours - received kapectate PO	BAR	BAR	BAR	BAR	Bright and alert, breathing improved some, sneezing - received Ensure, kapectate PO, 0.2ml Naxcel IM, nostrils cleared	BAR	BAR	BAR	BAR	BAR	BAR
3-Dec-03	BAR: scours - received kapectate PO	BAR: scours - received kapectate PO	BAR: scours - received kapectate PO	BAR: scours - received kapectate PO	BAR	BAR	BAR	BAR	Breathing hard, discharge from nose, not putting weight on hind end - received Ensure, nostrils cleared, needs to be euthanized	BAR	BAR	BAR	BAR	BAR	BAR

	018	019	020	022	025456	025463	025413	025461	025435	025436	025462	025415	025457	025416	025437	
4-Dec-03	BAR: scours - received kapectate PO	BAR: scours - received kapectate PO	BAR: scours - received kapectate PO	BAR: scours - received kapectate PO	BAR - received 1ml myco silencer BPME vaccine IM	BAR - received 1ml myco silencer BPME vaccine IM	BAR - received 1ml myco silencer BPME vaccine IM	BAR - received 1ml myco silencer BPME vaccine IM	Condition not improving enough pig euthanized and taken to UGAVTH for necropsy	BAR	BAR - received 1ml myco silencer BPME vaccine IM	BAR - received 1ml myco silencer BPME vaccine IM	BAR - received 1ml myco silencer BPME vaccine IM	BAR - received 1ml myco silencer BPME vaccine IM	BAR - received 1ml myco silencer BPME vaccine IM	
5-Dec-03	BAR: still scouring - received kapectate PO [Dr. Reeves Health Exam: Pig has previous history of intestinal herniation post castration w/ surgical correction. Has loose stool however is currently eating and drinking and is alert.]	BAR: scours - received kapectate PO [Dr. Reeves Health Exam: Pig appears to be in a good state of health however has had diarrhea for 3 days. Treating pig currently with kapectate.]	BAR: scours - received kapectate PO [Dr. Reeves Health Exam: Pig appears to be in a good state of health however has had diarrhea for 3 days. Treating pig currently with kapectate.]	BAR - scours gone [Dr. Reeves Health Exam: This pig currently appears to be very healthy. Had diarrhea for two days. Treated with kapectate for two days. Currently has normal stool.]	BAR [Dr. Reeves Health Exam: Pig appears to be in a good state of health.]	BAR [Dr. Reeves Health Exam: Pig appears to be in a good state of health.]	BAR [Dr. Reeves Health Exam: Pig appears to be in a good state of health.]	BAR [Dr. Reeves Health Exam: Pig appears to be in a good state of health.]			BAR [Dr. Reeves Health Exam: Pig appears to be in a good state of health.]	BAR [Dr. Reeves Health Exam: Pig appears to be in a good state of health.]	BAR [Dr. Reeves Health Exam: Pig appears to be in a good state of health.]	BAR [Dr. Reeves Health Exam: Pig appears to be in a good state of health.]	BAR [Dr. Reeves Health Exam: Pig appears to be in a good state of health.]	BAR [Dr. Reeves Health Exam: Pig appears to be in a good state of health.]
6-Dec-03	BAR: scours improved	BAR: scours improved	BAR: scours improved	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
7-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
8-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
9-Dec-03	BAR: scours - received 6mg/kg Neomycin in water per Dr. Reeves	BAR: scours - received 6mg/kg Neomycin in water per Dr. Reeves	BAR: scours - received 6mg/kg Neomycin in water per Dr. Reeves	BAR: scours - received 6mg/kg Neomycin in water per Dr. Reeves	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
10-Dec-03	BAR: scours improving - received 6mg/kg Neomycin in water	BAR: scours improving - received 6mg/kg Neomycin in water	BAR: scours improving - received 6mg/kg Neomycin in water	BAR: scours improving - received 6mg/kg Neomycin in water	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
11-Dec-03	BAR: scours improved - received 6mg/kg Neomycin in water	BAR: scours improved - received 6mg/kg Neomycin in water	BAR: scours improved - received 6mg/kg Neomycin in water	BAR: scours improved - received 6mg/kg Neomycin in water	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
12-Dec-03	BAR: scours gone	BAR: scours gone	BAR: scours gone	BAR: scours gone	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
13-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
14-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
15-Dec-03	BAR: swelling and heat on right side of abdomen, called Dr. Reeves to examine	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
16-Dec-03	BAR: swelling going down, less heat	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
17-Dec-03	BAR: swelling almost gone no heat	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
18-Dec-03	BAR: swelling gone	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
19-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
20-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
21-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
22-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
23-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
24-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
25-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
26-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
27-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
28-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
29-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
30-Dec-03	BAR - feed changed from starter to growers ration	BAR - feed changed from starter to growers ration	BAR - feed changed from starter to growers ration	BAR - feed changed from starter to growers ration	BAR - feed changed from starter to growers ration	BAR - feed changed from starter to growers ration	BAR - feed changed from starter to growers ration	BAR - feed changed from starter to growers ration		BAR - feed changed from starter to growers ration	BAR - feed changed from starter to growers ration	BAR - feed changed from starter to growers ration	BAR - feed changed from starter to growers ration	BAR - feed changed from starter to growers ration	BAR - feed changed from starter to growers ration	
31-Dec-03	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
1-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
2-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
3-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
4-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
5-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
6-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
7-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
8-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
9-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
10-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
11-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
12-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
13-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
14-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
15-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
16-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
17-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
18-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
19-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
20-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
21-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	
22-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR	

	018	019	020	022	025456	025463	025413	025461	025435	025436	025462	025415	025457	025416	025437
23-Jan-04	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]		BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]
24-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
25-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
26-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
27-Jan-04	BAR	BAR - put in individual pen due to large size	BAR - put in individual pen due to large size	BAR	BAR - put in individual pen due to large size	BAR - put in individual pen due to large size	BAR - put in individual pen due to large size	BAR - put in individual pen due to large size		BAR - put in individual pen due to large size	BAR - put in individual pen due to large size	BAR - put in individual pen due to large size	BAR - put in individual pen due to large size	BAR - put in individual pen due to large size	BAR - put in individual pen due to large size
28-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
29-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
30-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
31-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
1-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
2-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
3-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
4-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
5-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
6-Feb-04															
7-Feb-04															
8-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
9-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
10-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
11-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
12-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
13-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
14-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
15-Feb-04	BAR	BAR	BAR	Lethargic and not eating much, some loose stool - received 2mg/kg Naxcel IM, Pedialyte PO, and Kaopectate PO	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
16-Feb-04	BAR	BAR	BAR	Lethargic, loose stool, bright yellow urine - received Naxcel IM, Pedialyte PO, and Kaopectate PO	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
17-Feb-04	BAR	BAR	BAR	More alert but not eating, very little stool - received Naxcel IM, Pedialyte PO, Ensure PO	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
18-Feb-04	BAR	BAR	BAR	Condition improving - received Naxcel IM, Pedialyte PO, Ensure PO	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
19-Feb-04	BAR	BAR	BAR	Condition still improving in AM, by PM looks weak and uncomfortable, temp is 104 F - received Naxcel IM, Pedialyte PO, Ensure PO, 8ml Pen G IM in PM, 1ml Banamine IM in PM	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
20-Feb-04	BAR	BAR	BAR	Weak, temp is 102.9 F in AM, developing sores on eyes and face, will assist to stand 3x daily - received 8ml Pen G IM, 2 bottles Ensure PO, Saw Dr. Reeves in PM: septic? Received 2ml Dexamethasone IM and will receive 8ml Pen G Procaine IM 2x daily for 7 days	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
21-Feb-04	BAR	BAR	BAR	Still looks poor, assisted o stand - received 8ml Pen G Procaine IM in AM and PM, eye ointment 3x daily, 2ml Dexamethasone	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
22-Feb-04	BAR	BAR	BAR	Still weak but eating and drinking, sores look better - received 8ml Pen G Procaine IM in AM and PM, 2ml Dexamethasone, eye ointment 3x daily, Ensure PO, and Pedialyte PO	BAR	BAR	BAR	BAR		BAR	BAR	BAR	BAR	BAR	BAR
23-Feb-04	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	Condition improving, sores look better - received 8ml Pen G Procaine IM in AM and PM, 2ml Dexamethasone, eye ointment 3x daily [Dr. Reeves Health Exam: Diagnosed with influenza and secondary bacterial septicemia - treated with antibiotics and appears to be responding]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]		BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]

	018	019	020	022	025456	025463	025413	025461	025435	025436	025462	025415	025457	025416	025437
24-Feb-04	BAR	BAR	BAR	Condition improving, sores almost gone, stool is slightly loose - received 8ml Pen G Procaine IM in AM and PM, 2ml Dexamethasone eye ointment, ~10ml Kaopectate PO	BAR	BAR	BAR	BAR							
25-Feb-04	BAR	BAR	BAR	Condition improving, firm stool, sores gone - received 8ml Pen G Procaine IM in AM and PM	BAR	BAR	BAR	BAR							
26-Feb-04	BAR	BAR	BAR	Condition continuing to improve - received 8ml Pen G Procaine IM in AM and PM	BAR	BAR	BAR	BAR							
27-Feb-04	BAR	BAR	BAR	Condition continuing to improve, eating, drinking, brighter and more responsive	BAR	BAR	BAR	BAR							
28-Feb-04	BAR	BAR	BAR	Condition continuing to improve, eating and drinking regularly	BAR	BAR	BAR	BAR							
29-Feb-04	BAR	BAR	BAR	Alert and responsive, getting stronger	BAR	BAR	BAR	BAR							
1-Mar-04	BAR	BAR	BAR	Alert and responsive, eating and drinking, eyes are clear	BAR	BAR	BAR	BAR							
2-Mar-04	BAR	BAR	BAR	Alert and responsive, eating and drinking, gaining weight	BAR	BAR	BAR	BAR							
3-Mar-04	BAR	BAR	BAR	Alert and responsive; seems to have recovered	BAR	BAR	BAR	BAR							
4-Mar-04	BAR	BAR	BAR	Alert and responsive	BAR	BAR	BAR	BAR							
5-Mar-04	BAR	BAR	BAR	Alert and responsive	BAR	BAR	BAR	BAR							
6-Mar-04	BAR	BAR	BAR	Alert and responsive	BAR	BAR	BAR	BAR							
7-Mar-04	BAR	BAR	BAR	Alert and responsive	BAR	BAR	BAR	BAR							
8-Mar-04	BAR	BAR	BAR	Alert and responsive	BAR	BAR	BAR	BAR							
9-Mar-04	BAR	BAR	BAR	Alert and responsive	BAR	BAR	BAR	BAR							
10-Mar-04	BAR	BAR	BAR	Alert and responsive	BAR	BAR	BAR	BAR							
11-Mar-04	BAR	BAR	BAR	Alert and responsive	BAR	BAR	BAR	BAR							
12-Mar-04	BAR	BAR	BAR	Alert and responsive	BAR	BAR	BAR	BAR							
13-Mar-04	BAR	BAR	BAR	Alert and responsive	BAR	BAR	BAR	BAR							
14-Mar-04	BAR	BAR	BAR	Alert and responsive	BAR	BAR	BAR	BAR							
15-Mar-04	BAR	BAR	BAR	Alert and responsive	BAR	BAR	BAR	BAR							
16-Mar-04	BAR [Dr. Reeves Health Exam: Pig appears healthy]	BAR [Dr. Reeves Health Exam: Pig appears healthy]	BAR [Dr. Reeves Health Exam: Pig appears healthy]	BAR [Dr. Reeves Health Exam: Diagnosed with influenza with secondary bacterial septicemia (Feb 20th). Responded to antibiotic therapy, but appears to be visually impaired presently. Able to eat, drink and move around.]	BAR [Dr. Reeves Health Exam: Pig appears healthy]	BAR [Dr. Reeves Health Exam: Pig appears healthy]	BAR [Dr. Reeves Health Exam: Pig appears healthy]	BAR [Dr. Reeves Health Exam: Pig appears healthy]		BAR [Dr. Reeves Health Exam: Pig appears healthy]	BAR [Dr. Reeves Health Exam: Pig appears healthy]	BAR [Dr. Reeves Health Exam: Pig appears healthy]	BAR [Dr. Reeves Health Exam: Pig appears healthy]	BAR [Dr. Reeves Health Exam: Pig appears healthy]	BAR [Dr. Reeves Health Exam: Pig appears healthy]
17-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR							
18-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR							
19-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR							
20-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR							
21-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR							
22-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR	BAR							
23-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR; weighed off study	BAR; weighed off study		BAR; weighed off study	BAR; weighed off study	BAR; weighed off study	BAR; weighed off study	BAR; weighed off study	BAR; weighed off study
24-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR; feed removed in am, moved to meats lab in pm	BAR; feed removed in am, moved to meats lab in pm		BAR; feed removed in am, moved to meats lab in pm	BAR; feed removed in am, moved to meats lab in pm	BAR; feed removed in am, moved to meats lab in pm	BAR; feed removed in am, moved to meats lab in pm	BAR; feed removed in am, moved to meats lab in pm	BAR; feed removed in am, moved to meats lab in pm
25-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR	BAR; processed for study	BAR; processed for study		BAR; processed for study	BAR; processed for study	BAR; processed for study	BAR; processed for study	BAR; processed for study	BAR; processed for study
26-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR									
27-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR									
28-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR									
29-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR									
30-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR									
31-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR									
1-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR									
2-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR									
3-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR									
4-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR									
5-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR									
6-Apr-04	BAR - weighed off study	BAR - weighed off study	BAR - weighed off study	BAR - weighed with others but will not process with them due to light weight; will process with trial 2	BAR - weighed off study	BAR - weighed off study									BAR - weighed off study
7-Apr-04	BAR - feed removed AM, moved to meats lab PM	BAR - feed removed AM, moved to meats lab PM	BAR - feed removed AM, moved to meats lab PM	BAR	BAR - feed removed AM, moved to meats lab PM	BAR - feed removed AM, moved to meats lab PM									BAR - feed removed AM, moved to meats lab PM
8-Apr-04	Processed for study	Processed for study	Processed for study	BAR	Processed for study	Processed for study									Processed for study
9-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR									
10-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR									

	018	019	020	022	025456	025463	025413	025461	025435	025436	025462	025415	025457	025416	025437
11-Apr-04				BAR											
12-Apr-04				BAR											
13-Apr-04				BAR											
14-Apr-04				BAR											
15-Apr-04				BAR											
16-Apr-04				BAR [Dr. Reeves Health Exam: This pig was previously diagnosed as having influenza with a secondary bacterial septicemia. Appears to be fully recovered although eyes still appear cloudy. Eating and drinking well and appears to be gaining weight.]											
17-Apr-04				BAR											
18-Apr-04				BAR											
19-Apr-04				BAR											
20-Apr-04				BAR											
21-Apr-04				BAR											
22-Apr-04				BAR											
23-Apr-04				BAR											
24-Apr-04				BAR											
25-Apr-04				BAR											
26-Apr-04				BAR											
27-Apr-04				BAR											
28-Apr-04				BAR											
29-Apr-04				BAR											
30-Apr-04				BAR											
1-May-04				BAR											
2-May-04				BAR											
3-May-04				BAR											
4-May-04				BAR											
5-May-04				BAR											
6-May-04				BAR											
7-May-04				BAR											
8-May-04				BAR											
9-May-04				BAR											
10-May-04				BAR											
11-May-04				BAR											
12-May-04				BAR											
13-May-04				BAR											
14-May-04				BAR											
15-May-04				BAR											
16-May-04				BAR											
17-May-04				BAR [Dr. Reeves Health Exam: Pig was previously diagnosed with influenza and secondary bacterial septicemia but has recovered though eyes are still cloudy. Eating and drinking well.]											
18-May-04				BAR											
19-May-04				BAR											
20-May-04				BAR											
21-May-04				BAR											
22-May-04				BAR											
23-May-04				BAR											
24-May-04				BAR											
25-May-04				BAR											
26-May-04				BAR - weighed off study. Feed removed in AM but still has access to water. Will not be processed due to low weight.											
27-May-04				BAR - feed put back in AM after blood and urine sample obtained. Euthanized											

	024	027446	044584	044615	044661	044662
11-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
12-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
13-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
14-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
15-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
16-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
17-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
18-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
19-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
20-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
21-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
22-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
23-Jan-04	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]
24-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
25-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
26-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
27-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
28-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
29-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
30-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
31-Jan-04	BAR	BAR	BAR	BAR	BAR	BAR
1-Feb-04	BAR	BAR	BAR			
2-Feb-04	BAR	BAR	BAR	BAR		
3-Feb-04	BAR	BAR	BAR	BAR		
4-Feb-04	BAR	BAR	BAR	BAR		
5-Feb-04	BAR - feed changed from starter to grower	BAR - feed changed from starter to grower	BAR - feed changed from starter to grower	BAR - feed changed from starter to grower		
6-Feb-04						
7-Feb-04						
8-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
9-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
10-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
11-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
12-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
13-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
14-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
15-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
16-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
17-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
18-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
19-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
20-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
21-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
22-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
23-Feb-04	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR - moved to separate pen due to size [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig is slow grower with rough hair coat]
24-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
25-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
26-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
27-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
28-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
29-Feb-04	BAR	BAR	BAR	BAR	BAR	BAR
1-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
2-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
3-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
4-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
5-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
6-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
7-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
8-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
9-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
10-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
11-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
12-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
13-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
14-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
15-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
16-Mar-04	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]	BAR [Dr. Reeves Health Exam: Pig appears healthy.]
17-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
18-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
19-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR

	024	027446	044584	044615	044661	044662
20-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
21-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
22-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
23-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
24-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
25-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
26-Mar-04	BAR	BAR - swollen right rear dew claw - 5ml Pen G Procaine 2x daily IM	BAR	BAR	BAR	BAR
27-Mar-04	BAR	BAR - leg looks better - 5ml Pen G Procaine 2x daily IM	BAR	BAR	BAR	BAR
28-Mar-04	BAR	BAR - 5ml Pen G Procaine 2x daily IM	BAR	BAR	BAR	BAR
29-Mar-04	BAR	BAR - 5ml Pen G Procaine 2x daily IM	BAR	BAR	BAR	BAR
30-Mar-04	BAR	BAR - 5ml Pen G Procaine 2x daily IM	BAR	BAR	BAR	BAR
31-Mar-04	BAR	BAR	BAR	BAR	BAR	BAR
1-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
2-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
3-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
4-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
5-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
6-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
7-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
8-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
9-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
10-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
11-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
12-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
13-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
14-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
15-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
16-Apr-04	BAR [Dr. Reeves Health Exam - Pig appears healthy]	BAR [Dr. Reeves Health Exam - Pig appears healthy]	BAR [Dr. Reeves Health Exam - Pig appears healthy]	BAR [Dr. Reeves Health Exam - Pig appears healthy]	BAR [Dr. Reeves Health Exam - Pig appears healthy]	BAR [Dr. Reeves Health Exam - Pig appears healthy]
17-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
18-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
19-Apr-04	BAR	BAR	BAR	BAR - weighed off study. Feed removed in AM but still has access to water. Moved to meats lab in PM.	BAR	BAR - weighed off study. Feed removed in AM but still has access to water. Moved to meats lab in PM.
20-Apr-04	BAR	BAR	BAR	Processed for study	BAR	Processed for study
21-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
22-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
23-Apr-04	Has dropped weight but no difference in attitude or appetite noticed	BAR	BAR	BAR	BAR	BAR
24-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
25-Apr-04	BAR	BAR	BAR	BAR	BAR	BAR
26-Apr-04	Dull in AM, lethargic and breathing hard by 1PM, temp is 100.6 - received 6ml Procaine Pen G IM BID per Dr. Reeves recommendation	BAR	BAR	BAR	BAR	BAR
27-Apr-04	Very labored respiration, weak on hind end, not eating or defecating but is urinating, temp is 104.3 - received 6ml Procaine Pen G IM BID, 2ml Dexamethasone IM, 1ml Banamine IM, Pedialyte and Ensure PO - assisted to stand every three hours	BAR	BAR	BAR	BAR	BAR
28-Apr-04	Standing more easily, coughing and open mouth breathing, drinking, temp is 101.6 - assisted to stand throughout day and night - received 6ml Procaine Pen G IM BID, Pedialyte and Ensure PO, 2ml Dexamethasone IM	BAR	BAR	BAR	BAR	BAR

	024	027446	044584	044615	044661	044662
29-Apr-04	Standing more easily, drinking, urinating, has deficated some, more alert, temp is 100.5 - received 6ml Procaine Pen G IM BID, Pedialyte and Ensure PO	BAR	BAR		BAR	
30-Apr-04	Showing intrest in feed, drinking water, stronger on feet and legs - received 6ml Procaine Pen G IM BID, Pedialyte and Ensure PO	BAR	BAR		BAR	
1-May-04	Standing on his own, eating some, urinating and deficating - received pedialyte and ensure PO	BAR	BAR		BAR	
2-May-04	Standing and eating, urinating and deficating, alert and responsive - received Pedialyte and Ensure PO	BAR	BAR		BAR	
3-May-04	Alert and responsive, eating regularly	BAR	BAR		BAR	
4-May-04	Alert, vocal and responsive, eating regularly	BAR	BAR		BAR	
5-May-04	Alert and responsive, improving steadily	BAR	BAR		BAR	
6-May-04	Alert and responsive, improving steadily	BAR	BAR		BAR	
7-May-04	Alert and responsive, has lost considerable weight due to illness	BAR	BAR		BAR	
8-May-04	Alert and responsive, improving	BAR	BAR		BAR	
9-May-04	BAR - recovered	BAR	BAR			
10-May-04	BAR	BAR	BAR		Weighed off study. Feed removed in AM but still has access to water. Moved to meats lab in PM.	
11-May-04	BAR	BAR	BAR		Processed for study	
12-May-04	BAR	BAR	BAR			
13-May-04	BAR	BAR	BAR			
14-May-04	BAR	BAR	BAR			
15-May-04	BAR	BAR	BAR			
16-May-04	BAR	BAR	BAR			
17-May-04	BAR [Dr. Reeves Health Exam - Diagnosed recently (April 26th) with pnemonia, fever, lethargy, and poor appetite. Treated for five days with Procaine Penicillin G. Responded well to treatment and is now eating, drinking and gaining weight.	BAR [Dr. Reeves Health Exam - Structurally unsound due to poor feet/leg conformation, but otherwise healthy]	BAR [Dr. Reeves Health Exam - Pig appears healthy]			
18-May-04	BAR	BAR	BAR			
19-May-04	BAR	BAR	BAR			
20-May-04	BAR	BAR	BAR			
21-May-04	BAR	BAR	BAR			
22-May-04	BAR	BAR	BAR			
23-May-04	BAR	BAR	BAR			
24-May-04	BAR	BAR	BAR			
25-May-04	BAR	BAR	BAR			
26-May-04	BAR - weighed off study, feed removed in AM but still has access to water, moved to meats lab in PM	BAR - weighed off study, feed removed in AM but still has access to water, moved to meats lab in PM	BAR - weighed off study, feed removed in AM but still has access to water, moved to meats lab in PM			
27-May-04	Processed for study	Processed for study	Processed for study			

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Temperature (oF)
W195	H498	1	O18	1	9:35	AM	99.7
W195	H498	1	O18	1	2:45	PM	99.1
W195	H498	1	O18	1	7:00	PM	99.8
W195	H498	1	O18	2	12:00	AM	101
W195	H498	1	O18	2	6:00	AM	101.1
W195	H498	1	O18	2	12:00	PM	100.5
W195	H498	1	O18	2	6:00	PM	101.6
W195	H498	1	O18	3	12:00	AM	102.2
W195	H498	1	O18	3	6:00	AM	101.1
W195	H498	1	O18	3	12:00	PM	101.5
W195	H498	1	O18	3	5:30	PM	100.7
W195	H498	1	O18	4	12:00	AM	101.5
W195	H498	1	O18	4	2:00	AM	100.9
W195	H498	1	O18	4	6:15	AM	100.7
W195	H498	1	O18	4	12:00	PM	101.3
W195	H498	1	O18	4	6:15	PM	101.4
W195	H498	1	O18	5	2:00	AM	101.7
W195	H498	1	O18	5	9:45	AM	101.6
W195	H498	1	O18	5	7:00	PM	101.7
W195	H498	1	O18	6	2:00	AM	101.8
W195	H498	1	O18	6	10:00	AM	101.3
W195	H498	1	O18	6	6:00	PM	101.2
W195	H498	1	O18	7	1:30	AM	101.3
W195	H498	1	O18	7	10:00	AM	101.7
W195	H498	1	O18	8	2:00	AM	101.9
W195	H498	1	O18	8	10:00	AM	101.8
W195	H498	1	O18	8	6:00	PM	101.2
W195	H498	1	O19	1	9:40	AM	99.1
W195	H498	1	O19	1	2:45	PM	98.3
W195	H498	1	O19	1	7:00	PM	101.2
W195	H498	1	O19	2	12:00	AM	100.6
W195	H498	1	O19	2	6:00	AM	100.7
W195	H498	1	O19	2	12:00	PM	100.5
W195	H498	1	O19	2	6:00	PM	100.8
W195	H498	1	O19	3	12:00	AM	101.8
W195	H498	1	O19	3	6:00	AM	100.9
W195	H498	1	O19	3	12:00	PM	101.4
W195	H498	1	O19	3	5:30	PM	101.7
W195	H498	1	O19	4	2:00	AM	101.9
W195	H498	1	O19	4	9:45	AM	101.8
W195	H498	1	O19	4	7:00	PM	101.9
W195	H498	1	O19	5	2:00	AM	101.6
W195	H498	1	O19	5	10:00	AM	102
W195	H498	1	O19	5	6:00	PM	100.7
W195	H498	1	O19	6	2:00	AM	100.9
W195	H498	1	O19	6	10:00	AM	101.2
W195	H498	1	O19	7	2:00	AM	101.4
W195	H498	1	O19	7	10:00	AM	101.4
W195	H498	1	O19	7	6:00	PM	101
W195	H498	1	O19	8	2:00	AM	102.2
W195	H498	1	O19	8	10:00	AM	101.4
W195	H498	1	O19	8	6:00	PM	101.9
W195	H498	1	O20	1	9:45	AM	98
W195	H498	1	O20	1	2:45	PM	99.6

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Temperature (oF)
W195	H498	1	O20	1	7:00	PM	100.1
W195	H498	1	O20	2	12:00	AM	100.9
W195	H498	1	O20	2	6:00	AM	101.1
W195	H498	1	O20	2	12:00	PM	100.8
W195	H498	1	O20	2	6:00	PM	102.4
W195	H498	1	O20	3	12:00	AM	101.9
W195	H498	1	O20	3	6:00	AM	101.5
W195	H498	1	O20	3	12:00	PM	101.6
W195	H498	1	O20	3	5:30	PM	101.5
W195	H498	1	O20	4	12:00	AM	101.3
W195	H498	1	O20	4	2:00	AM	101.5
W195	H498	1	O20	4	9:45	AM	101.9
W195	H498	1	O20	4	7:00	PM	101.8
W195	H498	1	O20	5	2:00	AM	102
W195	H498	1	O20	5	10:00	AM	101.4
W195	H498	1	O20	5	6:00	PM	100.8
W195	H498	1	O20	6	1:30	AM	100.6
W195	H498	1	O20	6	10:00	AM	102
W195	H498	1	O20	7	2:00	AM	102.1
W195	H498	1	O20	7	10:00	AM	101
W195	H498	1	O20	7	6:00	PM	101.6
W195	H498	1	O20	8	2:00	AM	101.9
W195	H498	1	O20	8	10:00	AM	101.9
W195	H498	1	O20	8	6:00	PM	102.2
W170	H498	1	O22	1	10:00	AM	98.4
W170	H498	1	O22	1	11:15	AM	102.4
W170	H498	1	O22	1	12:30	PM	99
W170	H498	1	O22	1	1:20	PM	98.9
W170	H498	1	O22	1	2:30	PM	100
W170	H498	1	O22	1	4:25	PM	100.6
W170	H498	1	O22	1	7:20	PM	100.2
W170	H498	1	O22	1	8:00	PM	100.2
W170	H498	1	O22	1	11:45	PM	100.5
W170	H498	1	O22	2	4:00	AM	101
W170	H498	1	O22	2	10:00	AM	101.3
W170	H498	1	O22	2	12:00	PM	101
W170	H498	1	O22	2	1:30	PM	97.7
W170	H498	1	O22	2	3:45	PM	97.7
W170	H498	1	O22	2	5:30	PM	98.6
W170	H498	1	O22	3	2:00	AM	101.6
W170	H498	1	O22	3	6:00	AM	101.2
W170	H498	1	O22	3	10:00	AM	101.4
W170	H498	1	O22	3	6:00	PM	100.2
W170	H498	1	O22	4	12:00	AM	100.5
W170	H498	1	O22	4	2:00	am	100.9
W170	H498	1	O22	4	6:00	AM	101
W170	H498	1	O22	4	10:00	AM	101.6
W170	H498	1	O22	4	2:00	PM	101.7
W170	H498	1	O22	4	10:00	PM	101.2
W170	H498	1	O22	5	6:00	AM	101.3
W170	H498	1	O22	5	10:00	AM	101.5
W170	H498	1	O22	5	2:00	PM	101.5
W170	H498	1	O22	5	6:00	PM	101.2
W170	H498	1	O22	5	10:00	PM	101.3

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Temperature (oF)
W170	H498	1	O22	6	2:00	AM	101.5
W170	H498	1	O22	6	6:00	AM	101.3
W170	H498	1	O22	6	10:00	AM	100.9
W170	H498	1	O22	6	6:00	PM	101.3
W170	H498	1	O22	6	10:00	PM	101.5
W170	H498	1	O22	7	2:00	AM	101.2
W170	H498	1	O22	7	6:00	AM	101.2
W170	H498	1	O22	7	10:00	AM	101.5
W170	H498	1	O22	7	2:00	PM	101.4
W170	H498	1	O22	7	6:00	PM	102.4
W170	H498	1	O22	7	10:00	PM	102.1
W170	H498	1	O22	8	2:00	AM	101.9
W170	H498	1	O22	8	6:00	AM	102
W170	H498	1	O22	8	10:00	AM	102.2
W170	H498	1	O22	8	2:00	PM	101.7
W170	H498	1	O22	8	6:00	PM	101.2
W170	H498	1	O22	8	10:00	PM	101.4
W220	435-2	1	O24	1	9:00	AM	97.7
W220	435-2	2	O24	1	12:00	PM	104.6
W220	435-2	2	O24	1	12:20	PM	103.8
W220	435-2	2	O24	1	2:00	PM	102
W220	435-2	2	O24	1	6:00	PM	101.7
W220	435-2	2	O24	1	8:00	PM	102
W220	435-2	2	O24	1	10:00	PM	102.6
W220	435-2	2	O24	2	12:00	AM	102.2
W220	435-2	2	O24	2	2:00	AM	101.3
W220	435-2	2	O24	2	4:00	AM	100.6
W220	435-2	2	O24	2	6:00	AM	101.2
W220	435-2	2	O24	2	8:00	AM	101.1
W220	435-2	2	O24	2	10:00	AM	102.1
W220	435-2	2	O24	2	2:00	PM	100.7
W220	435-2	2	O24	2	6:00	PM	101.6
W220	435-2	2	O24	2	10:00	PM	101.5
W220	435-2	2	O24	3	12:00	AM	101.7
W220	435-2	2	O24	3	2:00	AM	101.6
W220	435-2	2	O24	3	6:00	AM	101.1
W220	435-2	2	O24	3	10:00	AM	100.9
W220	435-2	2	O24	3	1:45	PM	101.1
W220	435-2	2	O24	3	6:00	PM	101.9
W220	435-2	2	O24	3	10:00	PM	101.6
W220	435-2	2	O24	4	2:00	AM	101.7
W220	435-2	2	O24	4	6:00	AM	102.5
W220	435-2	2	O24	4	10:00	AM	102.1
W220	435-2	2	O24	4	2:00	PM	100.8
W220	435-2	2	O24	4	6:00	PM	101.1
W220	435-2	2	O24	5	12:00	AM	101.7
W220	435-2	2	O24	5	6:00	AM	101.5
W220	435-2	2	O24	5	12:00	PM	101.9
W220	435-2	2	O24	5	6:00	PM	102.1
W220	435-2	2	O24	5	10:00	PM	101.6
W220	435-2	2	O24	6	12:00	AM	101.5
W220	435-2	2	O24	6	6:00	AM	101.8
W220	435-2	2	O24	6	11:40	AM	101.9
W220	435-2	2	O24	6	6:00	PM	101.8

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Temperature (oF)
W220	435-2	2	O24	7	1:00	AM	101.4
W220	435-2	2	O24	7	12:00	PM	102.1
W220	435-2	2	O24	7	6:30	PM	102.3
W220	435-2	2	O24	8	6:00	AM	102.2
W220	435-2	2	O24	8	8:00	PM	103
W220	435-2	2	O24	8	9:30	PM	102

Day	# of Observations	Temperature (°F)
1	25	100.38+0.18a
2	27	100.8+0.17a
3	23	101.40+0.19b
4	23	101.43+0.19b
5	19	101.53+0.21b
6	16	101.41+0.23b
7	17	101.59+0.22b
8	18	101.89+0.21b

Control piglets were obtained over 3 weeks of age from a commercial producer and no data was available. Day 1 is day of birth and measurements were obtained over the first 8 days of life.

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Heart rate (beats per min)
W195	H498	1	O18	1	9:35	AM	220
W195	H498	1	O18	1	2:45	PM	198
W195	H498	1	O18	1	7:00	PM	204
W195	H498	1	O18	1	8:30	PM	198
W195	H498	1	O18	1	10:30	PM	196
W195	H498	1	O18	2	12:00	AM	196
W195	H498	1	O18	2	2:00	AM	192
W195	H498	1	O18	2	4:00	AM	196
W195	H498	1	O18	2	6:00	AM	188
W195	H498	1	O18	2	8:15	AM	180
W195	H498	1	O18	2	10:30	AM	188
W195	H498	1	O18	2	12:00	PM	180
W195	H498	1	O18	2	2:00	PM	184
W195	H498	1	O18	2	4:00	PM	188
W195	H498	1	O18	2	6:00	PM	180
W195	H498	1	O18	2	8:00	PM	188
W195	H498	1	O18	2	10:00	PM	184
W195	H498	1	O18	3	12:00	AM	184
W195	H498	1	O18	3	2:00	AM	180
W195	H498	1	O18	3	4:00	AM	184
W195	H498	1	O18	3	6:00	AM	180
W195	H498	1	O18	3	8:00	AM	176
W195	H498	1	O18	3	10:00	AM	176
W195	H498	1	O18	3	12:00	PM	180
W195	H498	1	O18	3	2:00	PM	184
W195	H498	1	O18	3	4:15	PM	186
W195	H498	1	O18	3	5:30	PM	180
W195	H498	1	O18	3	7:30	PM	178
W195	H498	1	O18	3	9:30	PM	176
W195	H498	1	O18	4	12:00	AM	180
W195	H498	1	O18	4	2:00	AM	180
W195	H498	1	O18	4	4:00	AM	176
W195	H498	1	O18	4	6:15	AM	180
W195	H498	1	O18	4	8:15	AM	176
W195	H498	1	O18	4	10:00	AM	176
W195	H498	1	O18	4	12:00	PM	178
W195	H498	1	O18	4	2:00	PM	176
W195	H498	1	O18	4	4:00	PM	178
W195	H498	1	O18	4	6:15	PM	180
W195	H498	1	O18	4	8:15	PM	176
W195	H498	1	O18	4	10:00	PM	178
W195	H498	1	O18	5	2:00	AM	176
W195	H498	1	O18	5	6:00	AM	178
W195	H498	1	O18	5	9:45	AM	179
W195	H498	1	O18	5	2:00	PM	176
W195	H498	1	O18	5	7:00	PM	178
W195	H498	1	O18	5	10:00	PM	176
W195	H498	1	O18	6	2:00	AM	176
W195	H498	1	O18	6	6:00	AM	180
W195	H498	1	O18	6	10:00	AM	180
W195	H498	1	O18	6	6:00	PM	180
W195	H498	1	O18	6	10:00	PM	180
W195	H498	1	O18	7	1:30	AM	176
W195	H498	1	O18	7	5:45	AM	178

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Heart rate (beats per min)
W195	H498	1	O18	7	10:00	AM	180
W195	H498	1	O18	7	2:00	PM	180
W195	H498	1	O18	7	10:00	PM	180
W195	H498	1	O18	8	2:00	AM	176
W195	H498	1	O18	8	6:00	AM	180
W195	H498	1	O18	8	10:00	AM	180
W195	H498	1	O18	8	2:00	PM	178
W195	H498	1	O18	8	6:00	PM	180
W195	H498	1	O18	8	10:00	PM	180
W195	H498	1	O19	1	9:40	AM	208
W195	H498	1	O19	1	2:45	PM	196
W195	H498	1	O19	1	7:00	PM	204
W195	H498	1	O19	1	8:30	PM	196
W195	H498	1	O19	1	10:30	PM	198
W195	H498	1	O19	2	12:00	AM	196
W195	H498	1	O19	2	2:00	AM	192
W195	H498	1	O19	2	4:00	AM	196
W195	H498	1	O19	2	6:00	AM	196
W195	H498	1	O19	2	8:15	AM	188
W195	H498	1	O19	2	10:30	AM	188
W195	H498	1	O19	2	12:00	PM	184
W195	H498	1	O19	2	2:00	PM	184
W195	H498	1	O19	2	4:00	PM	180
W195	H498	1	O19	2	6:00	PM	184
W195	H498	1	O19	2	8:00	PM	184
W195	H498	1	O19	2	10:00	PM	186
W195	H498	1	O19	3	12:00	AM	188
W195	H498	1	O19	3	2:00	AM	184
W195	H498	1	O19	3	4:00	AM	184
W195	H498	1	O19	3	6:00	AM	184
W195	H498	1	O19	3	8:00	AM	180
W195	H498	1	O19	3	10:00	AM	184
W195	H498	1	O19	3	12:00	PM	180
W195	H498	1	O19	3	2:00	PM	184
W195	H498	1	O19	3	4:15	PM	186
W195	H498	1	O19	3	5:30	PM	180
W195	H498	1	O19	3	7:30	PM	180
W195	H498	1	O19	3	9:30	PM	178
W195	H498	1	O19	4	2:00	AM	174
W195	H498	1	O19	4	6:00	AM	176
W195	H498	1	O19	4	9:45	AM	176
W195	H498	1	O19	4	2:00	PM	176
W195	H498	1	O19	4	7:00	PM	180
W195	H498	1	O19	4	10:00	PM	174
W195	H498	1	O19	5	2:00	AM	172
W195	H498	1	O19	5	6:00	AM	172
W195	H498	1	O19	5	10:00	AM	180
W195	H498	1	O19	5	6:00	PM	172
W195	H498	1	O19	5	10:00	PM	180
W195	H498	1	O19	6	2:00	AM	176
W195	H498	1	O19	6	6:00	AM	176
W195	H498	1	O19	6	10:00	AM	178
W195	H498	1	O19	6	2:00	PM	176
W195	H498	1	O19	6	10:00	PM	180

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Heart rate (beats per min)
W195	H498	1	O19	7	2:00	AM	180
W195	H498	1	O19	7	6:00	AM	180
W195	H498	1	O19	7	10:00	AM	178
W195	H498	1	O19	7	2:00	PM	178
W195	H498	1	O19	7	6:00	PM	182
W195	H498	1	O19	7	10:00	PM	180
W195	H498	1	O19	8	2:00	AM	182
W195	H498	1	O19	8	6:00	AM	180
W195	H498	1	O19	8	10:00	AM	176
W195	H498	1	O19	8	2:00	PM	176
W195	H498	1	O19	8	6:00	PM	172
W195	H498	1	O19	8	10:00	PM	176
W195	H498	1	O20	1	9:45	AM	216
W195	H498	1	O20	1	2:45	PM	190
W195	H498	1	O20	1	4:20	PM	196
W195	H498	1	O20	1	7:00	PM	198
W195	H498	1	O20	1	8:30	PM	196
W195	H498	1	O20	1	10:30	PM	196
W195	H498	1	O20	2	12:00	AM	190
W195	H498	1	O20	2	2:00	AM	188
W195	H498	1	O20	2	4:00	AM	192
W195	H498	1	O20	2	6:00	AM	190
W195	H498	1	O20	2	8:15	AM	190
W195	H498	1	O20	2	10:30	AM	188
W195	H498	1	O20	2	12:00	PM	184
W195	H498	1	O20	2	4:00	PM	176
W195	H498	1	O20	2	6:00	PM	176
W195	H498	1	O20	2	8:00	PM	188
W195	H498	1	O20	2	10:00	PM	184
W195	H498	1	O20	3	12:00	AM	188
W195	H498	1	O20	3	2:00	AM	172
W195	H498	1	O20	3	4:00	AM	188
W195	H498	1	O20	3	6:00	AM	180
W195	H498	1	O20	3	8:00	AM	176
W195	H498	1	O20	3	10:00	AM	180
W195	H498	1	O20	3	12:00	PM	180
W195	H498	1	O20	3	2:00	PM	180
W195	H498	1	O20	3	4:15	PM	180
W195	H498	1	O20	3	5:30	PM	180
W195	H498	1	O20	3	7:30	PM	178
W195	H498	1	O20	3	9:30	PM	176
W195	H498	1	O20	4	12:00	AM	180
W195	H498	1	O20	4	2:00	AM	174
W195	H498	1	O20	4	6:00	AM	176
W195	H498	1	O20	4	9:45	AM	176
W195	H498	1	O20	4	2:00	PM	178
W195	H498	1	O20	4	7:00	PM	176
W195	H498	1	O20	4	10:00	PM	178
W195	H498	1	O20	5	2:00	AM	180
W195	H498	1	O20	5	6:00	AM	172
W195	H498	1	O20	5	10:00	AM	172
W195	H498	1	O20	5	6:00	PM	180
W195	H498	1	O20	5	10:00	PM	176
W195	H498	1	O20	6	1:30	AM	178

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Heart rate (beats per min)
W195	H498	1	O20	6	5:45	AM	178
W195	H498	1	O20	6	10:00	AM	180
W195	H498	1	O20	6	2:00	PM	180
W195	H498	1	O20	6	10:00	PM	180
W195	H498	1	O20	7	2:00	AM	180
W195	H498	1	O20	7	6:00	AM	176
W195	H498	1	O20	7	10:00	AM	180
W195	H498	1	O20	7	2:00	PM	180
W195	H498	1	O20	7	6:00	PM	178
W195	H498	1	O20	7	10:00	PM	180
W195	H498	1	O20	8	2:00	AM	180
W195	H498	1	O20	8	6:00	AM	180
W195	H498	1	O20	8	10:00	AM	180
W195	H498	1	O20	8	2:00	PM	174
W195	H498	1	O20	8	6:00	PM	180
W195	H498	1	O20	8	10:00	PM	180
W170	H498	1	O22	1	10:00	AM	100
W170	H498	1	O22	1	12:30	PM	196
W170	H498	1	O22	1	2:30	PM	198
W170	H498	1	O22	1	4:25	PM	196
W170	H498	1	O22	1	6:00	PM	196
W170	H498	1	O22	1	8:00	PM	196
W170	H498	1	O22	1	10:00	PM	186
W170	H498	1	O22	1	11:45	PM	186
W170	H498	1	O22	2	2:00	AM	184
W170	H498	1	O22	2	4:00	AM	180
W170	H498	1	O22	2	6:00	AM	180
W170	H498	1	O22	2	8:00	AM	184
W170	H498	1	O22	2	10:00	AM	180
W170	H498	1	O22	2	12:00	PM	180
W170	H498	1	O22	2	1:30	PM	176
W170	H498	1	O22	2	2:30	PM	180
W170	H498	1	O22	2	3:45	PM	170
W170	H498	1	O22	2	5:30	PM	184
W170	H498	1	O22	2	7:30	PM	184
W170	H498	1	O22	3	12:30	AM	180
W170	H498	1	O22	3	2:00	AM	184
W170	H498	1	O22	3	4:00	AM	184
W170	H498	1	O22	3	6:00	AM	184
W170	H498	1	O22	3	7:45	AM	180
W170	H498	1	O22	3	10:00	AM	176
W170	H498	1	O22	3	12:00	PM	178
W170	H498	1	O22	3	2:00	PM	178
W170	H498	1	O22	3	6:00	PM	184
W170	H498	1	O22	4	12:00	AM	180
W170	H498	1	O22	4	2:00	am	178
W170	H498	1	O22	4	4:00	AM	178
W170	H498	1	O22	4	6:00	AM	178
W170	H498	1	O22	4	8:00	AM	180
W170	H498	1	O22	4	10:00	AM	180
W170	H498	1	O22	4	12:00	PM	180
W170	H498	1	O22	4	2:00	PM	180
W170	H498	1	O22	4	4:00	PM	178
W170	H498	1	O22	4	8:00	PM	180

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Heart rate (beats per min)
W170	H498	1	O22	4	10:00	PM	180
W170	H498	1	O22	5	12:00	AM	176
W170	H498	1	O22	5	4:00	AM	180
W170	H498	1	O22	5	6:00	AM	180
W170	H498	1	O22	5	8:00	AM	180
W170	H498	1	O22	5	10:00	AM	180
W170	H498	1	O22	5	12:00	PM	178
W170	H498	1	O22	5	2:00	PM	178
W170	H498	1	O22	5	6:00	PM	176
W170	H498	1	O22	5	10:00	PM	180
W170	H498	1	O22	6	12:00	AM	174
W170	H498	1	O22	6	2:00	AM	180
W170	H498	1	O22	6	4:00	AM	176
W170	H498	1	O22	6	6:00	AM	180
W170	H498	1	O22	6	8:00	AM	176
W170	H498	1	O22	6	10:00	AM	178
W170	H498	1	O22	6	12:00	PM	180
W170	H498	1	O22	6	4:00	PM	178
W170	H498	1	O22	6	6:00	PM	178
W170	H498	1	O22	6	8:00	PM	178
W170	H498	1	O22	6	10:00	PM	176
W170	H498	1	O22	7	12:00	AM	180
W170	H498	1	O22	7	2:00	AM	180
W170	H498	1	O22	7	4:00	AM	180
W170	H498	1	O22	7	6:00	AM	178
W170	H498	1	O22	7	8:00	AM	176
W170	H498	1	O22	7	10:00	AM	180
W170	H498	1	O22	7	12:00	PM	180
W170	H498	1	O22	7	2:00	PM	176
W170	H498	1	O22	7	6:00	PM	176
W170	H498	1	O22	7	8:00	PM	178
W170	H498	1	O22	7	10:00	PM	178
W170	H498	1	O22	8	12:00	AM	176
W170	H498	1	O22	8	2:00	AM	176
W170	H498	1	O22	8	4:00	AM	180
W170	H498	1	O22	8	6:00	AM	176
W170	H498	1	O22	8	8:00	AM	180
W170	H498	1	O22	8	10:00	AM	180
W170	H498	1	O22	8	12:00	PM	172
W170	H498	1	O22	8	2:00	PM	176
W170	H498	1	O22	8	4:00	PM	176
W170	H498	1	O22	8	6:00	PM	176
W170	H498	1	O22	8	8:00	PM	180
W170	H498	1	O22	8	10:00	PM	180
W220	435-2	2	O24	2	4:00	AM	180
W220	435-2	2	O24	2	6:00	AM	180
W220	435-2	2	O24	2	8:00	AM	180
W220	435-2	2	O24	2	10:00	AM	176
W220	435-2	2	O24	2	12:00	PM	172
W220	435-2	2	O24	2	6:00	PM	170
W220	435-2	2	O24	2	8:00	PM	176
W220	435-2	2	O24	3	12:00	AM	176
W220	435-2	2	O24	3	2:00	AM	172
W220	435-2	2	O24	3	4:00	AM	172

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Heart rate (beats per min)
W220	435-2	2	O24	3	6:00	AM	176
W220	435-2	2	O24	3	8:00	AM	176
W220	435-2	2	O24	3	10:00	AM	180
W220	435-2	2	O24	3	12:00	PM	176
W220	435-2	2	O24	3	1:45	PM	176
W220	435-2	2	O24	3	4:00	PM	180
W220	435-2	2	O24	3	6:00	PM	178
W220	435-2	2	O24	3	8:00	PM	180
W220	435-2	2	O24	3	10:00	PM	174
W220	435-2	2	O24	4	12:00	AM	180
W220	435-2	2	O24	4	2:00	AM	180
W220	435-2	2	O24	4	4:00	AM	172
W220	435-2	2	O24	4	6:00	AM	176
W220	435-2	2	O24	4	8:00	AM	176
W220	435-2	2	O24	4	10:00	AM	172
W220	435-2	2	O24	4	12:00	PM	168
W220	435-2	2	O24	4	2:00	PM	172
W220	435-2	2	O24	4	6:00	PM	180
W220	435-2	2	O24	4	10:00	PM	180
W220	435-2	2	O24	5	12:00	AM	176
W220	435-2	2	O24	5	2:00	AM	176
W220	435-2	2	O24	5	6:00	AM	178
W220	435-2	2	O24	5	9:45	AM	176
W220	435-2	2	O24	5	2:00	PM	180
W220	435-2	2	O24	5	6:00	PM	180
W220	435-2	2	O24	5	10:00	PM	180
W220	435-2	2	O24	6	2:00	AM	180
W220	435-2	2	O24	6	6:00	AM	180
W220	435-2	2	O24	6	10:00	AM	178
W220	435-2	2	O24	6	2:00	PM	172
W220	435-2	2	O24	6	6:00	PM	178
W220	435-2	2	O24	6	10:00	PM	174
W220	435-2	2	O24	7	1:00	AM	180
W220	435-2	2	O24	7	5:00	AM	180
W220	435-2	2	O24	7	10:00	AM	180
W220	435-2	2	O24	7	2:00	PM	180
W220	435-2	2	O24	7	6:30	PM	180
W220	435-2	2	O24	7	10:00	PM	178
W220	435-2	2	O24	8	3:00	AM	180
W220	435-2	2	O24	8	6:00	AM	176
W220	435-2	2	O24	8	10:00	AM	180
W220	435-2	2	O24	8	2:00	PM	180
W220	435-2	2	O24	8	9:30	PM	180

Day	# Observations	Heart rate (beats per min)
1	24	194.33+1.42a
2	53	184.22+0.96b
3	57	179.96+0.92c
4	46	177.30+1.03c
5	32	177.28+1.23c
6	32	177.94+1.23c
7	34	179.00+1.19c
8	35	178.11+1.18c

Control piglets were obtained over 3 weeks of age from a commercial producer and no data was available.
Day 1 is day of birth and measurements were obtained over the first 8 days of life.

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Respiration (breaths per min)
W195	H498	1	O18	1	9:35	AM	96
W195	H498	1	O18	1	2:45	PM	90
W195	H498	1	O18	1	4:20	PM	90
W195	H498	1	O18	1	7:00	PM	76
W195	H498	1	O18	1	8:30	PM	84
W195	H498	1	O18	1	10:30	PM	84
W195	H498	1	O18	2	12:00	AM	90
W195	H498	1	O18	2	2:00	AM	84
W195	H498	1	O18	2	4:00	AM	88
W195	H498	1	O18	2	6:00	AM	84
W195	H498	1	O18	2	8:15	AM	76
W195	H498	1	O18	2	10:30	AM	76
W195	H498	1	O18	2	12:00	PM	76
W195	H498	1	O18	2	2:00	PM	80
W195	H498	1	O18	2	4:00	PM	80
W195	H498	1	O18	2	6:00	PM	72
W195	H498	1	O18	2	8:00	PM	80
W195	H498	1	O18	2	10:00	PM	78
W195	H498	1	O18	3	12:00	AM	78
W195	H498	1	O18	3	2:00	AM	78
W195	H498	1	O18	3	4:00	AM	80
W195	H498	1	O18	3	6:00	AM	76
W195	H498	1	O18	3	8:00	AM	72
W195	H498	1	O18	3	10:00	AM	76
W195	H498	1	O18	3	12:00	PM	76
W195	H498	1	O18	3	2:00	PM	76
W195	H498	1	O18	3	4:15	PM	76
W195	H498	1	O18	3	5:30	PM	80
W195	H498	1	O18	3	7:30	PM	76
W195	H498	1	O18	3	9:30	PM	72
W195	H498	1	O18	4	12:00	AM	76
W195	H498	1	O18	4	2:00	AM	72
W195	H498	1	O18	4	4:00	AM	76
W195	H498	1	O18	4	6:15	AM	72
W195	H498	1	O18	4	8:15	AM	74
W195	H498	1	O18	4	10:00	AM	76
W195	H498	1	O18	4	12:00	PM	74
W195	H498	1	O18	4	2:00	PM	72
W195	H498	1	O18	4	4:00	PM	72
W195	H498	1	O18	4	6:15	PM	72
W195	H498	1	O18	4	8:15	PM	74
W195	H498	1	O18	4	10:00	PM	70
W195	H498	1	O18	5	2:00	AM	66
W195	H498	1	O18	5	6:00	AM	70
W195	H498	1	O18	5	9:45	AM	72
W195	H498	1	O18	5	2:00	PM	70
W195	H498	1	O18	5	7:00	PM	72
W195	H498	1	O18	5	10:00	PM	68
W195	H498	1	O18	6	2:00	AM	72
W195	H498	1	O18	6	6:00	AM	72
W195	H498	1	O18	6	10:00	AM	70
W195	H498	1	O18	6	6:00	PM	72
W195	H498	1	O18	6	10:00	PM	72
W195	H498	1	O18	7	1:30	AM	70

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Respiration (breaths per min)
W195	H498	1	O18	7	5:45	AM	70
W195	H498	1	O18	7	10:00	AM	68
W195	H498	1	O18	7	2:00	PM	68
W195	H498	1	O18	7	10:00	PM	72
W195	H498	1	O18	8	2:00	AM	72
W195	H498	1	O18	8	6:00	AM	72
W195	H498	1	O18	8	10:00	AM	66
W195	H498	1	O18	8	2:00	PM	68
W195	H498	1	O18	8	6:00	PM	68
W195	H498	1	O18	8	10:00	PM	72
W195	H498	1	O19	1	9:40	AM	90
W195	H498	1	O19	1	2:45	PM	92
W195	H498	1	O19	1	4:20	PM	96
W195	H498	1	O19	1	7:00	PM	90
W195	H498	1	O19	1	8:30	PM	90
W195	H498	1	O19	1	10:30	PM	84
W195	H498	1	O19	2	12:00	AM	90
W195	H498	1	O19	2	2:00	AM	88
W195	H498	1	O19	2	4:00	AM	92
W195	H498	1	O19	2	6:00	AM	90
W195	H498	1	O19	2	8:15	AM	84
W195	H498	1	O19	2	10:30	AM	84
W195	H498	1	O19	2	12:00	PM	72
W195	H498	1	O19	2	2:00	PM	76
W195	H498	1	O19	2	4:00	PM	80
W195	H498	1	O19	2	6:00	PM	80
W195	H498	1	O19	2	8:00	PM	80
W195	H498	1	O19	2	10:00	PM	78
W195	H498	1	O19	3	12:00	AM	72
W195	H498	1	O19	3	2:00	AM	78
W195	H498	1	O19	3	4:00	AM	84
W195	H498	1	O19	3	6:00	AM	80
W195	H498	1	O19	3	8:00	AM	76
W195	H498	1	O19	3	10:00	AM	76
W195	H498	1	O19	3	12:00	PM	80
W195	H498	1	O19	3	2:00	PM	76
W195	H498	1	O19	3	4:15	PM	76
W195	H498	1	O19	3	5:30	PM	72
W195	H498	1	O19	3	7:30	PM	72
W195	H498	1	O19	3	9:30	PM	72
W195	H498	1	O19	4	2:00	AM	70
W195	H498	1	O19	4	6:00	AM	70
W195	H498	1	O19	4	9:45	AM	70
W195	H498	1	O19	4	2:00	PM	74
W195	H498	1	O19	4	7:00	PM	74
W195	H498	1	O19	4	10:00	PM	70
W195	H498	1	O19	5	2:00	AM	76
W195	H498	1	O19	5	6:00	AM	72
W195	H498	1	O19	5	10:00	AM	72
W195	H498	1	O19	5	6:00	PM	68
W195	H498	1	O19	5	10:00	PM	68
W195	H498	1	O19	6	2:00	AM	68
W195	H498	1	O19	6	6:00	AM	72
W195	H498	1	O19	6	10:00	AM	70

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Respiration (breaths per min)
W195	H498	1	O19	6	2:00	PM	68
W195	H498	1	O19	6	10:00	PM	68
W195	H498	1	O19	7	2:00	AM	72
W195	H498	1	O19	7	6:00	AM	72
W195	H498	1	O19	7	10:00	AM	68
W195	H498	1	O19	7	2:00	PM	68
W195	H498	1	O19	7	6:00	PM	70
W195	H498	1	O19	7	10:00	PM	72
W195	H498	1	O19	8	2:00	AM	78
W195	H498	1	O19	8	6:00	AM	68
W195	H498	1	O19	8	10:00	AM	68
W195	H498	1	O19	8	2:00	PM	74
W195	H498	1	O19	8	6:00	PM	72
W195	H498	1	O19	8	10:00	PM	68
W195	H498	1	O20	1	9:45	AM	98
W195	H498	1	O20	1	2:45	PM	84
W195	H498	1	O20	1	4:20	PM	96
W195	H498	1	O20	1	7:00	PM	90
W195	H498	1	O20	1	8:30	PM	84
W195	H498	1	O20	1	10:30	PM	78
W195	H498	1	O20	2	12:00	AM	86
W195	H498	1	O20	2	2:00	AM	84
W195	H498	1	O20	2	4:00	AM	88
W195	H498	1	O20	2	6:00	AM	80
W195	H498	1	O20	2	8:15	AM	84
W195	H498	1	O20	2	10:30	AM	80
W195	H498	1	O20	2	12:00	PM	80
W195	H498	1	O20	2	4:00	PM	80
W195	H498	1	O20	2	6:00	PM	76
W195	H498	1	O20	2	8:00	PM	80
W195	H498	1	O20	2	10:00	PM	78
W195	H498	1	O20	3	12:00	AM	72
W195	H498	1	O20	3	2:00	AM	76
W195	H498	1	O20	3	4:00	AM	78
W195	H498	1	O20	3	6:00	AM	76
W195	H498	1	O20	3	8:00	AM	72
W195	H498	1	O20	3	10:00	AM	76
W195	H498	1	O20	3	12:00	PM	80
W195	H498	1	O20	3	2:00	PM	78
W195	H498	1	O20	3	4:15	PM	72
W195	H498	1	O20	3	5:30	PM	72
W195	H498	1	O20	3	7:30	PM	72
W195	H498	1	O20	3	9:30	PM	70
W195	H498	1	O20	4	12:00	AM	80
W195	H498	1	O20	4	2:00	AM	70
W195	H498	1	O20	4	6:00	AM	70
W195	H498	1	O20	4	9:45	AM	74
W195	H498	1	O20	4	2:00	PM	74
W195	H498	1	O20	4	7:00	PM	72
W195	H498	1	O20	4	10:00	PM	70
W195	H498	1	O20	5	2:00	AM	76
W195	H498	1	O20	5	6:00	AM	68
W195	H498	1	O20	5	10:00	AM	66
W195	H498	1	O20	5	6:00	PM	72

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Respiration (breaths per min)
W195	H498	1	O20	5	10:00	PM	68
W195	H498	1	O20	6	1:30	AM	68
W195	H498	1	O20	6	5:45	AM	72
W195	H498	1	O20	6	10:00	AM	72
W195	H498	1	O20	6	2:00	PM	78
W195	H498	1	O20	6	10:00	PM	64
W195	H498	1	O20	7	2:00	AM	68
W195	H498	1	O20	7	6:00	AM	68
W195	H498	1	O20	7	10:00	AM	68
W195	H498	1	O20	7	2:00	PM	74
W195	H498	1	O20	7	6:00	PM	68
W195	H498	1	O20	7	10:00	PM	68
W195	H498	1	O20	8	2:00	AM	78
W195	H498	1	O20	8	6:00	AM	72
W195	H498	1	O20	8	10:00	AM	72
W195	H498	1	O20	8	2:00	PM	72
W195	H498	1	O20	8	6:00	PM	76
W195	H498	1	O20	8	10:00	PM	68
W170	H498	1	O22	1	12:30	PM	92
W170	H498	1	O22	1	2:30	PM	88
W170	H498	1	O22	1	4:25	PM	90
W170	H498	1	O22	1	6:00	PM	90
W170	H498	1	O22	1	8:00	PM	90
W170	H498	1	O22	1	10:00	PM	92
W170	H498	1	O22	1	11:45	PM	80
W170	H498	1	O22	2	2:00	AM	84
W170	H498	1	O22	2	4:00	AM	80
W170	H498	1	O22	2	6:00	AM	76
W170	H498	1	O22	2	8:00	AM	76
W170	H498	1	O22	2	10:00	AM	72
W170	H498	1	O22	2	12:00	PM	76
W170	H498	1	O22	2	1:30	PM	72
W170	H498	1	O22	2	2:30	PM	80
W170	H498	1	O22	2	3:45	PM	80
W170	H498	1	O22	2	5:30	PM	72
W170	H498	1	O22	2	7:30	PM	84
W170	H498	1	O22	3	12:30	AM	76
W170	H498	1	O22	3	2:00	AM	72
W170	H498	1	O22	3	4:00	AM	76
W170	H498	1	O22	3	6:00	AM	76
W170	H498	1	O22	3	7:45	AM	76
W170	H498	1	O22	3	10:00	AM	76
W170	H498	1	O22	3	12:00	PM	78
W170	H498	1	O22	3	2:00	PM	74
W170	H498	1	O22	3	6:00	PM	76
W170	H498	1	O22	4	12:00	AM	72
W170	H498	1	O22	4	2:00	am	72
W170	H498	1	O22	4	4:00	AM	72
W170	H498	1	O22	4	6:00	AM	74
W170	H498	1	O22	4	8:00	AM	78
W170	H498	1	O22	4	10:00	AM	76
W170	H498	1	O22	4	12:00	PM	72
W170	H498	1	O22	4	2:00	PM	72
W170	H498	1	O22	4	4:00	PM	72

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Respiration (breaths per min)
W170	H498	1	O22	4	8:00	PM	76
W170	H498	1	O22	4	10:00	PM	72
W170	H498	1	O22	5	12:00	AM	72
W170	H498	1	O22	5	4:00	AM	76
W170	H498	1	O22	5	6:00	AM	72
W170	H498	1	O22	5	8:00	AM	72
W170	H498	1	O22	5	10:00	AM	72
W170	H498	1	O22	5	12:00	PM	72
W170	H498	1	O22	5	2:00	PM	72
W170	H498	1	O22	5	6:00	PM	70
W170	H498	1	O22	5	10:00	PM	72
W170	H498	1	O22	6	12:00	AM	68
W170	H498	1	O22	6	2:00	AM	72
W170	H498	1	O22	6	4:00	AM	68
W170	H498	1	O22	6	6:00	AM	76
W170	H498	1	O22	6	8:00	AM	68
W170	H498	1	O22	6	10:00	AM	68
W170	H498	1	O22	6	12:00	PM	70
W170	H498	1	O22	6	4:00	PM	70
W170	H498	1	O22	6	6:00	PM	70
W170	H498	1	O22	6	8:00	PM	70
W170	H498	1	O22	6	10:00	PM	68
W170	H498	1	O22	7	12:00	AM	72
W170	H498	1	O22	7	2:00	AM	68
W170	H498	1	O22	7	4:00	AM	72
W170	H498	1	O22	7	6:00	AM	72
W170	H498	1	O22	7	8:00	AM	72
W170	H498	1	O22	7	10:00	AM	72
W170	H498	1	O22	7	12:00	PM	68
W170	H498	1	O22	7	2:00	PM	72
W170	H498	1	O22	7	6:00	PM	68
W170	H498	1	O22	7	8:00	PM	70
W170	H498	1	O22	7	10:00	PM	70
W170	H498	1	O22	8	12:00	AM	68
W170	H498	1	O22	8	2:00	AM	72
W170	H498	1	O22	8	4:00	AM	72
W170	H498	1	O22	8	6:00	AM	68
W170	H498	1	O22	8	8:00	AM	76
W170	H498	1	O22	8	10:00	AM	76
W170	H498	1	O22	8	12:00	PM	72
W170	H498	1	O22	8	2:00	PM	72
W170	H498	1	O22	8	4:00	PM	72
W170	H498	1	O22	8	6:00	PM	76
W170	H498	1	O22	8	8:00	PM	76
W170	H498	1	O22	8	10:00	PM	72
W220	435-2	2	O24	1	9:00	AM	88
W220	435-2	2	O24	1	12:00	PM	88
W220	435-2	2	O24	1	4:00	PM	88
W220	435-2	2	O24	1	6:00	PM	84
W220	435-2	2	O24	1	7:00	PM	88
W220	435-2	2	O24	1	8:00	PM	72
W220	435-2	2	O24	1	10:00	PM	80
W220	435-2	2	O24	2	12:00	AM	76
W220	435-2	2	O24	2	2:00	AM	76

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Respiration (breaths per min)
W220	435-2	2	O24	2	4:00	AM	78
W220	435-2	2	O24	2	6:00	AM	76
W220	435-2	2	O24	2	8:00	AM	78
W220	435-2	2	O24	2	10:00	AM	78
W220	435-2	2	O24	2	12:00	PM	76
W220	435-2	2	O24	2	2:00	PM	76
W220	435-2	2	O24	2	6:00	PM	74
W220	435-2	2	O24	2	8:00	PM	72
W220	435-2	2	O24	2	10:00	PM	72
W220	435-2	2	O24	3	12:00	AM	76
W220	435-2	2	O24	3	2:00	AM	72
W220	435-2	2	O24	3	4:00	AM	68
W220	435-2	2	O24	3	6:00	AM	72
W220	435-2	2	O24	3	8:00	AM	76
W220	435-2	2	O24	3	10:00	AM	72
W220	435-2	2	O24	3	12:00	PM	68
W220	435-2	2	O24	3	1:45	PM	78
W220	435-2	2	O24	3	4:00	PM	80
W220	435-2	2	O24	3	6:00	PM	88
W220	435-2	2	O24	3	8:00	PM	78
W220	435-2	2	O24	3	10:00	PM	84
W220	435-2	2	O24	4	12:00	AM	72
W220	435-2	2	O24	4	2:00	AM	76
W220	435-2	2	O24	4	4:00	AM	76
W220	435-2	2	O24	4	6:00	AM	68
W220	435-2	2	O24	4	8:00	AM	72
W220	435-2	2	O24	4	10:00	AM	68
W220	435-2	2	O24	4	12:00	PM	72
W220	435-2	2	O24	4	2:00	PM	68
W220	435-2	2	O24	4	6:00	PM	66
W220	435-2	2	O24	4	10:00	PM	72
W220	435-2	2	O24	5	12:00	AM	68
W220	435-2	2	O24	5	2:00	AM	68
W220	435-2	2	O24	5	6:00	AM	70
W220	435-2	2	O24	5	9:45	AM	80
W220	435-2	2	O24	5	2:00	PM	78
W220	435-2	2	O24	5	6:00	PM	80
W220	435-2	2	O24	5	10:00	PM	80
W220	435-2	2	O24	6	2:00	AM	76
W220	435-2	2	O24	6	6:00	AM	72
W220	435-2	2	O24	6	10:00	AM	76
W220	435-2	2	O24	6	2:00	PM	80
W220	435-2	2	O24	6	6:00	PM	72
W220	435-2	2	O24	6	10:00	PM	72
W220	435-2	2	O24	7	1:00	AM	70
W220	435-2	2	O24	7	5:00	AM	72
W220	435-2	2	O24	7	10:00	AM	70
W220	435-2	2	O24	7	2:00	PM	72
W220	435-2	2	O24	7	6:30	PM	72
W220	435-2	2	O24	7	10:00	PM	74
W220	435-2	2	O24	8	3:00	AM	76
W220	435-2	2	O24	8	6:00	AM	68
W220	435-2	2	O24	8	10:00	AM	78
W220	435-2	2	O24	8	2:00	PM	76

Gilt Number	Cell Line	Trial	Piglet ID	Day	Time	AM/PM	Respiration (breaths per min)
W220	435-2	2	O24	8	9:30	PM	78

Day	# Observations	Respiration (breaths per min)
1	32	87.56+0.71a
2	57	79.61+0.53b
3	57	75.72+0.53c
4	46	72.52+0.59d
5	32	71.81+0.71de
6	32	71.06+0.71de
7	34	70.29+0.79e
8	35	72.34+0.68d

Control piglets were obtained over 3 weeks of age from a commercial producer and no data was available. Day 1 is day of birth and measurements were obtained over the first 8 days of life.

Body Weight													ADG	WDA
Treatment	Barrow ID	Trial #	Line	DOB	BWT	On Test Date	On Test WT	Age on Test	Off Test Date	Off Test WT	Off Test Age	ADG	WDA	
Clone	Clone O18	1	Ham	3-Oct-03	2.80	21-Nov-03	42.6	49	6-Apr-04	265	186	1.62	1.41	
Clone	Clone O19	1	Ham	3-Oct-03	2.40	21-Nov-03	38.8	49	6-Apr-04	244	186	1.50	1.30	
Clone	Clone O20	1	Ham	3-Oct-03	2.40	21-Nov-03	41.7	49	6-Apr-04	280	186	1.74	1.49	
Clone	Clone O21	1	Ham	12-Oct-03	2.30	21-Nov-03	17.2	40	21-May-04	190	222	0.95	0.85	
Clone	Clone O22	1	Ham	12-Oct-03	2.40	21-Nov-03	26.4	40	21-May-04	289	222	1.44	1.29	
Control	O25456	1	Ham	11-Oct-03	3.54	21-Nov-03	20.2	41	6-Apr-04	303	178	2.06	1.68	
Control	O25463	1	Ham	11-Oct-03	4.60	21-Nov-03	17.4	41	6-Apr-04	283	178	1.94	1.56	
Control	O25413	1	Ham	8-Oct-03	2.94	21-Nov-03	27.5	44	23-Mar-04	280	167	2.05	1.66	
Control	O25461	1	Ham	11-Oct-03	4.56	21-Nov-03	30.7	41	23-Mar-04	304	164	2.22	1.83	
Control	O25436	1	Ham	10-Oct-03	3.68	21-Nov-03	20.7	42	23-Mar-04	268	165	2.01	1.60	
Control	O25462	1	Ham	11-Oct-03	3.26	21-Nov-03	24.8	41	23-Mar-04	266	164	1.96	1.60	
Control	O25415	1	Ham	8-Oct-03	4.46	21-Nov-03	36.5	44	23-Mar-04	293	167	2.09	1.73	
Control	O25457	1	Ham	11-Oct-03	3.56	21-Nov-03	25.1	41	23-Mar-04	259	164	1.90	1.56	
Control	O25416	1	Ham	8-Oct-03	2.84	21-Nov-03	27.9	44	23-Mar-04	278	167	2.03	1.65	
Control	O25437	1	Ham	10-Oct-03	3.92	21-Nov-03	26.6	42	6-Apr-04	280	179	1.85	1.54	
Clone	023	2	Ham	13-Nov-03	2.00	16-Jan-04	19	64	26-May-04	198	195	1.37	1.01	
Clone	024	2	Duroc	14-Nov-03	3.00	16-Jan-04	67	63	26-May-04	246	194	1.37	1.25	
Control	027446	2	Ham	10-Nov-03	3.50	16-Jan-04	46	67	26-May-04	286	198	1.83	1.43	
Control	044584	2	Ham	13-Nov-03	3.40	16-Jan-04	39	64	26-May-04	310	195	2.07	1.57	
Control	044615	2	Ham	14-Nov-03	3.80	16-Jan-04	60	63	19-Apr-04	278	157	2.32	1.75	
Control	044661	2	Ham	15-Nov-03	4.80	16-Jan-04	53.5	62	10-May-04	306	177	2.20	1.70	
Control	044662	2	Ham	15-Nov-03	4.20	16-Jan-04	49.5	62	19-Apr-04	268	156	2.32	1.69	

Treatment	Trial	#	BWT	On Test Date	On Test WT	Age on Test	Off Test Date	Off Test WT	Off Test Age	ADG	WDA
Clone		1	5	2.46	33.34	45.4		253.6	200.4	1.45	1.268
Clone		2	2	2.5	43	63.5		222	194.5	1.37	1.13
Control		1	10	3.736	25.74	42.1		281.4	169.3	2.011	1.641
Control		2	5	3.94	49.6	63.6		289.6	176.6	2.148	1.628
Averages		clone		2.47	36.10	50.57		244.57	198.71	1.43	1.23
		control		3.80	33.69	49.27		284.13	171.73	2.06	1.64

Treatment	#	BWT	On Test Date	On Test WT	Age on Test	Off Test Date	Off Test WT	Off Test Age	ADG	WDA
Control	Ham	15	mean	3.80	33.69	49.27	284.13	171.73	2.06	1.64
			stdev	0.61	13.15	10.61	15.97	12.38	0.15	0.10
			stderr	0.16	3.40	2.74	4.12	3.20	0.04	0.03
Clone	Ham	6	mean	2.30	28.62	48.40	240.20	202.20	1.40	1.19
			stdev	0.17	11.21	9.81	45.50	18.44	0.29	0.26
			stderr	0.07	4.58	4.01	18.58	7.53	0.12	0.11
Clone	Duroc	1		3.00	67.00	63.00	246.00	194.00	1.37	1.25

Collection Date	Trial #	treatment	Barrow ID	Line	WBC								
					WBC	SEG	BAND	LYMPH	MONO	EOS	BASO	Other	FIBR
					x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	mg/dl
5-Dec-03	1	Clone	018	Ham	9.8	3.332	0.000	5.782	0.294	0.294	0.098	0.000	300
5-Dec-03	1	Clone	019	Ham	15.2	6.688	0.152	5.472	1.672	0.912	0.304	0.000	200
5-Dec-03	1	Clone	020	Ham	32.5	13.000	0.325	17.550	1.300	0.325	0.000	0.000	300
5-Dec-03	1	Clone	021	Ham	23.2	7.888	3.248	10.672	0.928	0.464	0.000	0.000	300
5-Dec-03	1	Clone	022	Ham	17.0	8.160	0.000	8.500	0.170	0.170	0.000	0.000	200
5-Dec-03	1	Control	025456	Ham	17.8	5.340	0.178	10.680	1.424	0.178	0.000	0.000	200
5-Dec-03	1	Control	025463	Ham	28.9	9.537	0.867	17.340	0.867	0.289	0.000	0.000	200
5-Dec-03	1	Control	025413	Ham	34.8	10.440	0.000	22.272	1.740	0.348	0.000	0.000	300
5-Dec-03	1	Control	025461	Ham	19.9	11.343	0.199	7.363	0.796	0.199	0.000	0.000	300
5-Dec-03	1	Control	025436	Ham	27.1	14.634	0.271	10.027	1.626	0.271	0.271	0.000	300
5-Dec-03	1	Control	025462	Ham	20.6	12.566	0.206	6.592	0.824	0.412	0.000	0.000	200
5-Dec-03	1	Control	025415	Ham	24.0	9.120	0.000	13.200	1.200	0.480	0.000	0.000	200
5-Dec-03	1	Control	025457	Ham	21.4	13.482	0.214	7.490	0.000	0.214	0.000	0.000	200
5-Dec-03	1	Control	025416	Ham	32.3	12.597	0.000	17.765	1.615	0.323	0.000	0.000	300
5-Dec-03	1	Control	025437	Ham	18.6	8.184	0.186	7.254	1.116	1.302	0.558	0.000	400
23-Jan-04	1	Clone	018	Ham	22.4	9.856	0.448	10.528	1.120	0.448	0.000	0.000	300
23-Jan-04	1	Clone	019	Ham	16.8	9.744	0.168	6.216	0.336	0.336	0.000	0.000	400
23-Jan-04	1	Clone	020	Ham	16.4	6.560	0.328	8.364	0.820	0.164	0.164	0.000	300
23-Jan-04	1	Clone	021	Ham	20.4	10.200	0.204	8.364	1.224	0.204	0.204	0.000	200
23-Jan-04	1	Clone	022	Ham	12.8	5.120	0.256	6.144	0.768	0.384	0.128	0.000	300
23-Jan-04	1	Control	025456	Ham	19.3	7.527	0.193	9.843	1.158	0.579	0.000	0.000	200
23-Jan-04	1	Control	025463	Ham	24.5	9.555	0.000	13.475	0.735	0.735	0.000	0.000	100
23-Jan-04	1	Control	025413	Ham	21.1	8.229	0.211	9.495	1.266	1.688	0.211	0.000	300
23-Jan-04	1	Control	025461	Ham	18.6	7.626	0.186	9.858	0.744	0.186	0.000	0.000	100
23-Jan-04	1	Control	025436	Ham	24.9	13.197	0.249	8.466	2.241	0.747	0.000	0.000	400
23-Jan-04	1	Control	025462	Ham	16.4	7.708	0.000	7.708	0.656	0.328	0.000	0.000	200
23-Jan-04	1	Control	025415	Ham	21.1	10.550	0.211	8.862	1.055	0.422	0.000	0.000	200
23-Jan-04	1	Control	025457	Ham	17.6	8.448	0.176	7.920	0.880	0.176	0.000	0.000	200
23-Jan-04	1	Control	025416	Ham	28.2	14.100	0.000	12.972	0.846	0.282	0.000	0.000	200
23-Jan-04	1	Control	025437	Ham	23.0	7.360	0.230	12.420	1.610	0.920	0.460	0.000	100
8-Apr-04	1	Clone	018	Ham	9.0	1.980	0.000	6.480	0.360	0.180	0.000	0.000	100
8-Apr-04	1	Clone	019	Ham	14.2	5.254	0.000	8.520	0.284	0.142	0.000	0.000	400
8-Apr-04	1	Clone	020	Ham	12.1	5.324	0.000	6.050	0.484	0.242	0.000	0.000	400
8-Apr-04	1	Control	025456	Ham	16.9	6.929	0.000	9.126	0.338	0.507	0.000	0.000	500
8-Apr-04	1	Control	025463	Ham	14.3	6.149	0.000	7.150	0.429	0.572	0.000	0.000	200
8-Apr-04	1	Control	025413	Ham	17.5	8.050	0.000	8.750	0.700	0.000	0.000	0.000	100
8-Apr-04	1	Control	025461	Ham	12.7	5.334	0.000	6.477	0.762	0.127	0.000	0.000	300
8-Apr-04	1	Control	025436	Ham	17.0	2.550	0.000	13.770	0.680	0.000	0.000	0.000	300
8-Apr-04	1	Control	025462	Ham	13.1	3.275	0.000	9.170	0.393	0.262	0.000	0.000	400
8-Apr-04	1	Control	025415	Ham	9.6	3.520	0.000	5.856	0.192	0.000	0.000	0.000	200
8-Apr-04	1	Control	025457	Ham	12.9	5.934	0.129	6.450	0.258	0.129	0.000	0.000	300

Collection Date	Trial #	treatment	Barrow ID	Line	WBC								
					WBC	SEG	BAND	LYMPH	MONO	EOS	BASO	Other	FIBR
					x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	mg/dl
8-Apr-04	1	Control	025416	Ham	15.3	7.344	0.000	7.038	0.765	0.000	0.153	0.000	300
8-Apr-04	1	Control	025437	Ham	24.4	11.468	0.244	9.516	0.976	2.196	0.000	0.000	400
8-Apr-04	1	Clone	021	Ham	19.1	6.876	0.000	9.932	0.955	1.337	0.000	0.000	100
8-Apr-04	1	Clone	022	Ham	14.2	6.532	0.000	6.390	0.426	0.852	0.000	0.000	400
16-Jan-04	2	Clone	023	Ham	12.6	7.182	0.504	4.284	0.630	0.000	0.000	0.000	200
16-Jan-04	2	Clone	024	Duroc	13.2	7.260	0.396	5.016	0.264	0.264	0.000	0.000	500
16-Jan-04	2	Control	027446	Ham	26.6	10.374	0.798	13.832	0.798	0.532	0.266	0.000	200
16-Jan-04	2	Control	044584	Ham	18.9	8.316	0.000	9.261	0.945	0.378	0.000	0.000	200
16-Jan-04	2	Control	044615	Ham	18.5	9.990	0.185	7.400	0.740	0.185	0.000	0.000	100
16-Jan-04	2	Control	044661	Ham	15.6	8.892	0.000	5.928	0.468	0.156	0.156	0.000	300
16-Jan-04	2	Control	044662	Ham	12.7	5.969	0.254	5.588	0.508	0.381	0.000	0.000	200
16-Mar-04	2	Clone	023	Ham	21.8	10.464	0.218	8.720	0.872	0.654	0.872	0.000	200
16-Mar-04	2	Clone	024	Duroc	21.3	6.816	0.426	12.993	0.852	0.213	0.000	0.000	200
16-Mar-04	2	Control	027446	Ham	29.3	9.962	0.293	16.408	1.758	0.879	0.000	0.000	200
16-Mar-04	2	Control	044584	Ham	20.0	7.800	0.000	11.800	0.400	0.000	0.000	0.000	300
16-Mar-04	2	Control	044615	Ham	26.6	11.438	0.000	13.300	0.798	0.798	0.266	0.000	200
16-Mar-04	2	Control	044661	Ham	19.1	8.595	0.000	9.932	0.382	0.000	0.191	0.000	100
16-Mar-04	2	Control	044662	Ham	20.2	6.262	0.000	12.120	0.808	1.010	0.000	0.000	200
27-May-04	2	Clone	023	Ham	17.5	4.550	0.000	11.725	0.350	0.700	0.175	0.000	100
27-May-04	2	Clone	024	Duroc	19.6	9.996	0.000	6.860	2.156	0.392	0.196	0.000	300
27-May-04	2	Control	027446	Ham	22.8	9.120	0.000	12.540	0.912	0.228	0.000	0.000	400
27-May-04	2	Control	044584	Ham	9.4	3.008	0.000	6.298	0.000	0.094	0.000	0.000	300
27-May-04	2	Control	044661	Ham	14.5	7.395	0.290	6.090	0.580	0.000	0.000	0.145	100
27-May-04	2	Control	044615	Ham	15.4	5.082	0.154	9.394	0.616	0.154	0.000	0.000	300
27-May-04	2	Control	044662	Ham	18.2	4.914	0.182	10.738	0.546	1.638	0.182	0.000	300

Date	Trial	TRT	#/TRT	WBC	SEG	BAND	LYMPH	MONO	EOS	BASO	Other	FIBR
				x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	x 10 ³ /ul	mg/dl
5-Dec-03	1	Clone	5	19.54	7.81	0.75	9.60	0.87	0.43	0.08	0.00	260.00
5-Dec-03	1	Control	10	24.54	10.72	0.21	12.00	1.12	0.40	0.08	0.00	260.00
23-Jan-04	1	Clone	5	17.76	8.30	0.28	7.92	0.85	0.31	0.10	0.00	300.00
23-Jan-04	1	Control	10	21.47	9.43	0.15	10.10	1.12	0.61	0.07	0.00	200.00
8-Apr-04	1	Clone	5	13.72	5.19	0.00	7.47	0.50	0.55	0.00	0.00	280.00
8-Apr-04	1	Control	10	15.37	6.06	0.04	8.33	0.55	0.38	0.02	0.00	300.00
16-Jan-04	2	Clone	2	12.90	7.22	0.45	4.65	0.45	0.13	0.00	0.00	350.00
16-Jan-04	2	Control	5	18.46	8.71	0.25	8.40	0.69	0.33	0.08	0.00	200.00
16-Mar-04	2	Clone	2	21.55	8.64	0.32	10.86	0.86	0.43	0.44	0.00	200.00
16-Mar-04	2	Control	5	23.04	8.81	0.06	12.71	0.83	0.54	0.09	0.00	200.00
27-May-04	2	Clone	2	18.55	7.27	0.00	9.29	1.25	0.55	0.19	0.00	200.00
27-May-04	2	Control	5	16.06	5.90	0.13	9.01	0.53	0.42	0.04	0.03	280.00
Literature Ranges				11 to 22	2 to 15	0 to 0.8	3.8 to 16.5	0 to 1.0	0 to 1.5	0 to 3		200 to 400

Collection Date	Trial #	Treatment	Barrow ID	Line	RBC/PLT													
					HCT		RBC	HGB	MCV	MCH	MCHC	Platelets	MPV	PLT EST	ABS retic	Retic	NRBC	
					%	x 10 ⁶ /ul	g/dl	fl	pg	g/dl	x 10 ³ /ul	fl		x 10 ³ /ul	%			
5-Dec-03	1	Clone	018	Ham	29.0	4.77	7.5	60.7	15.7	25.8	397	8.8	adequate	5	0.1	0		
5-Dec-03	1	Clone	019	Ham	32.0	6.96	10.8	45.9	15.5	33.7	439	9.5	adequate	0	0.0	0		
5-Dec-03	1	Clone	020	Ham	28.0	6.52	10.2	42.9	15.6	36.4	445	8.4	adequate	7	0.1	0		
5-Dec-03	1	Clone	021	Ham	30.0	5.51	8.6	54.4	15.6	28.6	417	8.7	adequate	22	0.4	0		
5-Dec-03	1	Clone	022	Ham	31.0	6.89	10.3	44.9	14.9	33.2	539	9.2	adequate	7	0.1	0		
5-Dec-03	1	Control	025456	Ham	26.0	5.80	8.7	44.8	15.0	33.4	647	9.7	adequate	58	1.0	1		
5-Dec-03	1	Control	025463	Ham	29.0	5.27	8.7	55.4	16.5	29.8	634	13.9	adequate	47	0.9	0		
5-Dec-03	1	Control	025413	Ham	28.5	6.38	9.0	44.6	14.1	31.5	622	9.0	adequate	115	1.8	0		
5-Dec-03	1	Control	025461	Ham	29.0	5.54	10.6	52.3	19.1	36.5	705	9.0	adequate	139	2.5	0		
5-Dec-03	1	Control	025436	Ham	28.5	5.19	9.7	54.9	18.7	34.0	593	8.6	adequate	130	2.5	0		
5-Dec-03	1	Control	025462	Ham	24.0	5.53	7.8	43.4	14.1	32.5	692	9.8	adequate	55	1.0	1		
5-Dec-03	1	Control	025415	Ham	32.0	6.54	10.7	48.9	16.4	33.4	713	9.6	adequate	105	1.6	0		
5-Dec-03	1	Control	025457	Ham	29.0	6.13	9.8	47.3	16.0	33.7	681	9.6	adequate	147	2.4	1		
5-Dec-03	1	Control	025416	Ham	26.5	6.09	8.7	43.5	14.3	32.8	782	8.8	adequate	85	1.4	0		
5-Dec-03	1	Control	025437	Ham	28.5	5.13	9.9	55.5	19.3	34.7	387	8.2	adequate	103	2.0	0		
23-Jan-04	1	Clone	018	Ham	36.0	7.90	11.1	45.6	14.1	30.8	559	8.4		16	0.2	0		
23-Jan-04	1	Clone	019	Ham	32.5	7.72	10.9	42.1	14.1	33.5	610	8.4	adequate	0	0.0	0		
23-Jan-04	1	Clone	020	Ham	32.5	7.84	10.9	41.4	13.9	33.5	453	8.7	adequate	0	0.0	0		
23-Jan-04	1	Clone	021	Ham	31.5	7.14	11.1	44.1	15.5	35.2	609	9.1		0	0.0	0		
23-Jan-04	1	Clone	022	Ham	34.0	7.57	11.3	44.9	14.9	33.2	426	8.7	adequate	8	0.1	0		
23-Jan-04	1	Control	025456	Ham	35.5	7.42	12.2	47.8	16.4	34.4	458	8.8	adequate	7	0.1	0		
23-Jan-04	1	Control	025463	Ham	33.0	7.25	11.6	45.5	16.0	35.1	415	9.1	adequate	15	0.2	0		
23-Jan-04	1	Control	025413	Ham	34.0	8.27	11.5	41.1	13.9	33.8	395	6.8	adequate	0	0.0	0		
23-Jan-04	1	Control	025461	Ham	33.0	6.57	11.9	50.2	18.1	36.1	430	7.8	adequate	72	1.1	0		
23-Jan-04	1	Control	025436	Ham	30.5	5.91	10.6	51.6	17.9	34.8	671	7.6	adequate	12	0.2	0		
23-Jan-04	1	Control	025462	Ham	30.5	6.96	10.3	43.8	14.8	33.8	351	7.0	adequate	56	0.8	0		
23-Jan-04	1	Control	025415	Ham	33.0	7.55	11.8	43.7	15.6	35.8	443	8.0		0	0.0	0		
23-Jan-04	1	Control	025457	Ham	32.0	7.12	11.6	44.9	16.3	36.2	475	7.9	adequate	7	0.1	0		
23-Jan-04	1	Control	025416	Ham	33.5	8.43	11.5	39.7	13.6	34.3	449	6.4	adequate	17	0.2	0		
23-Jan-04	1	Control	025437	Ham	35.5	6.70	11.9	53.0	17.8	33.5	363	8.5	adequate	0	0.0	0		
8-Apr-04	1	Clone	018	Ham	32.5	7.41	11.0	43.9	14.8	33.8	42	8.6	decr	15	0.2	0		
8-Apr-04	1	Clone	019	Ham	38.5	8.84	13.2	43.6	14.9	34.3	142	8.1	clumps	0	0.0	0		
8-Apr-04	1	Clone	020	Ham	38.5	8.81	12.7	43.7	14.4	33.0	162	7.9	sl. decr	18	0.2	0		
8-Apr-04	1	Control	025456	Ham	44.5	8.64	14.7	51.5	17.0	33.0	231	8.7	adequate	17	0.2	0		
8-Apr-04	1	Control	025463	Ham	42.0	8.77	13.8	47.9	15.7	32.9	317	8.8	adequate	18	0.2	0		
8-Apr-04	1	Control	025413	Ham	41.5	8.51	14.0	48.8	16.5	33.7	259	7.0	adequate	9	0.1	0		
8-Apr-04	1	Control	025461	Ham	38.0	7.18	13.6	52.9	18.9	35.8	311	7.8	adequate	0	0.0	0		
8-Apr-04	1	Control	025436	Ham	37.0	6.26	11.1	59.1	17.7	30.0	434	8.3	adequate	13	0.2	0		
8-Apr-04	1	Control	025462	Ham	37.0	6.29	11.3	58.4	18.0	30.5	326	8.5	adequate	6	0.1	0		
8-Apr-04	1	Control	025415	Ham	37.0	6.53	10.7	56.7	16.4	28.9	287	7.6	adequate	7	0.1	0		
8-Apr-04	1	Control	025457	Ham	39.0	7.01	12.8	55.6	18.3	32.8	65	7.8	clumps	0	0.0	0		
8-Apr-04	1	Control	025416	Ham	41.5	8.59	13.8	48.3	16.1	33.2	212	9.9	adequate	9	0.1	0		
8-Apr-04	1	Control	025437	Ham	40.0	6.97	12.1	57.4	17.4	30.2	332	8.2	adequate	35	0.5	0		
8-Apr-04	1	Clone	021	Ham	36.8	7.07	11.6	52.0	16.4	31.5	126	9.5	clumps	14	0.2	0		
8-Apr-04	1	Clone	022	Ham	42.0	8.80	13.1	47.7	14.9	31.2	319	8.8	adequate	18	0.2	2		
16-Jan-04	2	Clone	023	Ham	37.0	8.65	12.5	42.8	14.5	33.3	242	9.5	adequate	26	0.3	0		
16-Jan-04	2	Clone	024	Duroc	31.0	6.56	9.9	46.3	15.1	31.9	625	8.2	adequate	13	0.2	0		
16-Jan-04	2	Control	027446	Ham	32.0	5.95	10.6	53.8	17.8	33.1	386	8.9	adequate	131	2.2	0		
16-Jan-04	2	Control	044584	Ham	31.5	5.71	10.0	55.7	17.5	31.7	425	7.3	adequate	114	2.0	0		
16-Jan-04	2	Control	044615	Ham	29.0	5.39	9.8	53.8	18.2	33.7	365	8.7	adequate	11	0.2	0		
16-Jan-04	2	Control	044661	Ham	32.5	6.80	10.4	47.8	15.3	32.0	366	8.7	adequate	75	1.1	0		
16-Jan-04	2	Control	044662	Ham	32.0	5.96	10.3	54.0	17.3	32.2	328	8.4	adequate	54	0.9	0		
16-Mar-04	2	Clone	023	Ham	37.0	8.17	11.4	45.3	14.0	30.8	616	8.9	adequate	33	0.4	0		
16-Mar-04	2	Clone	024	Duroc	39.0	8.84	12.6	44.1	14.3	32.3	416	8.4	adequate	44	0.5	0		
16-Mar-04	2	Control	027446	Ham	29.0	6.51	11.3	44.5	17.4	38.9	291	8.9	adequate	7	0.1	0		
16-Mar-04	2	Control	044584	Ham	34.0	7.05	11.5	48.2	16.3	33.8	358	7.4	adequate	14	0.2	1		
16-Mar-04	2	Control	044615	Ham	31.5	5.84	11.0	53.9	18.8	34.9	336	8.4	adequate	12	0.2	0		
16-Mar-04	2	Control	044661	Ham	34.5	8.06	12.0	42.8	14.9	34.8	327	9.1	adequate	32	0.4	0		
16-Mar-04	2	Control	044662	Ham	34.0	6.79	11.1	50.1	16.3	32.6	320	8.6	adequate	20	0.3	0		
27-May-04	2	Clone	023	Ham	40.0	8.69	12.4	46.0	14.3	31.0	130	7.0	clumps	35	0.4	0		
27-May-04	2	Clone	024	Duroc	41.5	9.23	13.6	45.0	14.7	32.8	511	8.7	clumps	28	0.3	0		
27-May-04	2	Control	027446	Ham	47.5	9.51	15.7	49.9	16.5	33.0	344	9.0	clumps	48	0.5	1		
27-May-04	2	Control	044584	Ham	36.5	6.92	12.2	52.7	17.6	33.4	284	7.9	clumps	48	0.7	1		
27-May-04	2	Control	044661	Ham	39.5	8.70	13.6	45.4	15.6	34.4	267	9.2	adequate	26	0.3	0		
27-May-04	2	Control	044615	Ham	39.0	7.36	13.8	53.0	18.8	35.3	219	8.4	clumps	22	0.3	0		
27-May-04	2	Control	044662	Ham	37.5	7.37	12.4	50.9	16.8	33.1	326	8.5	clumps	74	1.0	0		

Trial	Date	TRT	#/TRT	HCT	RBC	HGB	MCV	MCH	MCHC	Platelets	MPV	PLT EST	ABS retic	Retic	NRBC
				%	x 10 ⁶ /ul	g/dl	fl	pg	g/dl	x 10 ³ /ul	fl		x 10 ³ /ul	%	
1	5-Dec-03	Clone	5	30.00	6.13	9.48	49.76	15.46	31.54	447.40	8.92		8.20	0.14	0.00
1	5-Dec-03	Control	10	28.10	5.76	9.36	49.06	16.35	33.23	645.60	9.62		98.40	1.71	0.30
1	23-Jan-04	Clone	5	33.30	7.63	11.06	43.62	14.50	33.24	531.40	8.66		4.80	0.06	0.00
1	23-Jan-04	Control	10	33.05	7.22	11.49	46.13	16.04	34.78	445.00	7.79		18.60	0.27	0.00
1	8-Apr-04	Clone	5	37.66	8.19	12.32	46.18	15.08	32.76	158.20	8.58		13.00	0.16	0.40
1	8-Apr-04	Control	10	39.75	7.48	12.79	53.66	17.20	32.10	277.40	8.26		11.40	0.15	0.00
2	16-Jan-04	Clone	2	34.00	7.61	11.20	44.55	14.80	32.60	433.50	8.85		19.50	0.25	0.00
2	16-Jan-04	Control	5	31.40	5.96	10.22	53.02	17.22	32.54	374.00	8.40		77.00	1.28	0.00
2	16-Mar-04	Clone	2	38.00	8.51	12.00	44.70	14.15	31.55	516.00	8.65		38.50	0.45	0.00
2	16-Mar-04	Control	5	32.60	6.85	11.38	47.90	16.74	35.00	326.40	8.48		17.00	0.24	0.20
2	27-May-04	Clone	2	40.75	8.96	13.00	45.50	14.50	31.90	320.50	7.85		31.50	0.35	0.00
2	27-May-04	Control	5	40.00	7.97	13.54	50.38	17.06	33.84	288.00	8.60		43.60	0.56	0.40
		Literature Ranges			5 to 7	9 to 13	52 to 56		29 to 34	200-500				0 to 12	

Collection Date	Trial #	Trt	Barrow ID	Line	Measurement/Units																				
					Serum Chemistry																				
					Urea Nitrogen	Total Protein	Albumin	Alk Phos	ALT	Glucose	Calcium	AST	Creatine Kinase	Amylase	Lipase	Cholesterol	Bile Acids	GGTP	SDH	Creatinine	Sodium	Potassium	Chloride	Bicarb.	Anion GAP
mg/dl	g/dl	g/dl	U/L	U/L	mg/dl	mg/dl	U/L	U/L	U/L	U/L	U/L	mg/dl	umol/L	U/L	U/L	mg/dl	mmol/L	mmol/L	mmol/L	mmol/L	mmol/L				
5-Dec-03	1	Clone	018	Ham	15	4.5	2.9	179	28	96	9.3	30	322	974	17	98	16.9	82	0.0	1.7	143	4.9	108	15	25
5-Dec-03	1	Clone	019	Ham	14	4.4	2.9	261	54	100	9.5	55	1094	1172	11	79	14.1	50	0.0	1.7	141	6.3	100	26	21
5-Dec-03	1	Clone	020	Ham	14	5.0	3.5	191	44	82	9.6	47	815	1093	21	83	19.4	86	0.0	1.7	144	5.2	104	22	23
5-Dec-03	1	Clone	021	Ham	23	4.9	3.1	156	36	73	9.8	58	376	1161	31	91	20.5	115	0.0	1.4	150	6.5	108	23	26
5-Dec-03	1	Clone	022	Ham	11	4.4	2.9	190	32	106	9.9	32	402	1167	31	74	16.8	82	0.0	1.2	142	5.6	101	26	21
5-Dec-03	1	Control	025456	Ham	9	4.8	3.7	281	98	117	10.6	51	1170	4119	12	42	14.6	66	0.0	1.5	144	6.4	103	26	21
5-Dec-03	1	Control	025463	Ham	10	4.9	3.4	324	123	118	10.6	107	5213	4120	19	64	40.9	61	0.0	1.3	147	5.8	103	20	30
5-Dec-03	1	Control	025413	Ham	12	5.6	4.2	286	53	127	11.9	27	342	4219	19	65	22.7	51	0.0	1.3	153	7.5	107	18	36
5-Dec-03	1	Control	025461	Ham	13	5.2	4.1	228	84	112	10.9	31	857	2338	14	54	19.9	50	0.0	1.1	144	6.2	100	28	22
5-Dec-03	1	Control	025436	Ham	13	5.0	3.5	264	53	114	10.4	32	395	1899	12	62	8.4	32	0.0	0.9	146	5.7	104	24	24
5-Dec-03	1	Control	025462	Ham	9	5.9	4.3	184	54	112	10.9	43	770	2978	14	51	10.7	82	0.0	1.1	145	6.0	102	23	26
5-Dec-03	1	Control	025415	Ham	16	5.2	3.9	201	82	119	11.1	35	727	5100	10	68	14.8	71	0.0	1.2	144	5.2	102	25	22
5-Dec-03	1	Control	025457	Ham	13	5.2	4.1	228	84	122	10.9	31	857	2338	14	54	19.9	50	0.0	1.1	146	5.6	103	26	23
5-Dec-03	1	Control	025416	Ham	14	5.5	4.1	264	73	131	11.5	30	1160	3269	12	63	25.0	56	0.0	1.2	148	6.3	101	23	30
5-Dec-03	1	Control	025437	Ham	11	5.5	4.2	224	53	112	10.8	27	519	1418	14	70	14.1	25	0.0	1.0	146	5.5	105	26	21
23-Jan-04	1	Clone	018	Ham	11	5.8	3.3	169	38	99	10.0	30	1396	1199	9	83	27.2	42	0.0	2.0	146	5.1	102	30	19
23-Jan-04	1	Clone	019	Ham	14	6.1	2.9	187	39	109	9.9	19	262	1201	8	129	54.7	51	0.0	1.6	146	5.7	101	33	18
23-Jan-04	1	Clone	020	Ham	15	5.8	3.3	184	37	105	9.7	23	660	1160	8	115	39.7	47	0.0	2.1	146	5.6	102	30	20
23-Jan-04	1	Clone	021	Ham	26	6.0	2.8	93	34	91	10.6	44	554	1131	11	93	47.8	48	4.4	1.7	149	5.7	105	30	20
23-Jan-04	1	Clone	022	Ham	15	5.6	3.3	181	45	114	9.9	24	927	1190	9	91	40.0	56	0.0	1.8	143	5.3	101	30	17
23-Jan-04	1	Control	025456	Ham	11	5.5	3.8	140	46	97	10.8	17	298	4031	10	79	40.5	51	0.0	1.4	149	5.4	104	31	19
23-Jan-04	1	Control	025463	Ham	13	5.0	3.5	172	58	113	9.9	25	270	3470	13	79	33.4	36	0.4	1.5	148	5.0	105	30	18
23-Jan-04	1	Control	025413	Ham	13	5.4	3.4	116	31	98	10.2	12	177	3664	15	78	14.4	39	0.4	1.4	147	5.5	104	31	18
23-Jan-04	1	Control	025461	Ham	12	5.9	3.8	139	40	88	10.7	18	359	2375	11	78	27.2	56	0.1	1.5	148	5.1	103	31	19
23-Jan-04	1	Control	025436	Ham	11	5.7	3.4	128	28	89	10.1	16	188	1323	7	83	17.7	27	0.6	1.1	147	4.7	102	31	19
23-Jan-04	1	Control	025462	Ham	11	5.8	3.8	120	42	105	10.6	19	388	2854	10	76	25.0	36	0.0	1.3	147	5.4	104	30	18
23-Jan-04	1	Control	025415	Ham	13	5.3	3.8	103	44	111	10.5	19	340	5368	10	86	20.2	59	0.0	1.8	149	5.9	102	34	19
23-Jan-04	1	Control	025457	Ham	11	5.2	3.6	130	49	102	10.0	14	493	2036	11	82	30.1	35	0.0	1.4	150	5.1	103	33	19
23-Jan-04	1	Control	025416	Ham	16	5.8	4.1	140	56	103	10.2	13	842	3072	13	71	25.7	45	0.0	1.8	149	5.3	104	30	20
23-Jan-04	1	Control	025437	Ham	13	5.9	4.1	143	49	93	10.7	18	272	1089	11	91	80.0	27	0.0	1.6	149	4.9	106	29	19
8-Apr-04	1	Clone	018	Ham	19	6.8	4.3	137	57	93	9.6	59	4926	1263	9	100	30.3	34	0.0	2.4	146	6.2	103	29	20
8-Apr-04	1	Clone	019	Ham	18	6.4	4.2	157	57	94	9.4	68	3061	1322	13	94	63.8	37	13.9	2.0	144	6.1	101	29	20
8-Apr-04	1	Clone	020	Ham	17	6.2	4.0	115	51	105	9.5	65	4539	991	11	96	62.7	35	19.2	2.3	144	7.4	101	29	21
8-Apr-04	1	Control	025456	Ham	18	7.5	5.1	125	67	87	10.7	34	2250	4274	20	129	49.4	52	0.0	2.2	144	8.0	101	28	23
8-Apr-04	1	Control	025463	Ham	19	6.7	4.6	167	84	91	9.6	68	2793	3577	22	112	55.5	34	19.8	1.8	144	8.3	102	29	21
8-Apr-04	1	Control	025413	Ham	20	7.5	4.5	163	61	73	10.6	53	3585	4314	15	94	11.9	42	0.2	1.9	144	6.5	100	28	23
8-Apr-04	1	Control	025461	Ham	14	7.6	4.8	174	53	96	10.2	44	3108	3082	42	104	22.7	47	3.9	1.9	138	8.2	98	30	18
8-Apr-04	1	Control	025436	Ham	17	7.1	4.9	130	44	84	10.6	39	2008	1705	7	112	26.3	28	4.1	1.8	145	6.9	104	26	22
8-Apr-04	1	Control	025462	Ham	19	8.0	4.9	136	60	97	10.9	55	3202	3214	9	109	25.5	39	0.0	2.0	142	6.7	98	26	25
8-Apr-04	1	Control	025415	Ham	22	7.1	4.9	125	74	93	11.0	63	4282	6252	10	99	14.2	46	0.2	2.1	150	6.3	105	29	22
8-Apr-04	1	Control	025457	Ham	15	7.5	4.8	106	56	100	10.7	57	7678	2065	12	91	24.3	35	0.0	1.9	144	7.2	102	29	20
8-Apr-04	1	Control	025416	Ham	21	7.7	5.1	170	76	96	11.1	48	3478	3643	10	112	39.3	48	0.6	2.6	148	6.1	102	28	24
8-Apr-04	1	Control	025437	Ham	17	7.3	4.9	144	60	80	10.5	48	2458	1557	24	115	30.1	25	2.1	1.7	143	5.8	101	25	23
8-Apr-04	1	Clone	021	Ham	20	6.4	3.6	129	103	93	10.1	210	12876	1120	14	92	30.7	96	2.2	4.2	145	6.9	98	30	24
8-Apr-04	1	Clone	022	Ham	19	7.1	3.9	101	47	72	10.1	60	3578	1136	5	106	38.2	47	22.5	2.5	148	7.5	104	27	23

Collection Date	Trial #	Trt	Borrow ID	Line	Measurement/Units																				
					Serum Chemistry																				
					Urea Nitrogen	Total Protein	Albumin	Alk Phos	ALT	Glucose	Calcium	AST	Creatine Kinase	Amylase	Lipase	Cholesterol	Bile Acids	GGTP	SDH	Creatinine	Sodium	Potassium	Chloride	Bicarb.	Anion GAP
mg/dl	g/dl	g/dl	U/L	U/L	mg/dl	mg/dl	U/L	U/L	U/L	U/L	U/L	U/L	U/L	U/L	U/L	mg/dl	mmol/L	mmol/L	mmol/L	mmol/L	mmol/L				
16-Jan-04	2	Clone	023	Ham	31	6.5	3.6	78	72	139	11.2	68	786	568	4	103	21.9	58	0.0	2.5	127	5.9	93	13	27
16-Jan-04	2	Clone	024	Duroc	14	5.8	4.2	260	44	130	11.2	21	1200	2643	8	61	20.9	14	0.4	1.0	145	5.4	101	29	20
16-Jan-04	2	Control	027446	Ham	10	5.2	4.0	249	53	121	10.5	46	1624	2182	7	74	19.9	48	0.0	1.4	144	5.4	103	22	24
16-Jan-04	2	Control	044584	Ham	15	5.0	3.7	212	43	121	10.9	86	3831	3148	6	58	27.5	50	0.0	1.6	144	5.9	102	27	21
16-Jan-04	2	Control	044615	Ham	13	4.9	3.4	226	81	115	11.0	33	1071	2853	10	68	19.5	44	0.0	1.3	145	5.3	103	27	20
16-Jan-04	2	Control	044661	Ham	10	5.4	3.8	243	92	109	11.4	54	682	4159	16	61	26.3	36	0.0	1.3	144	5.7	101	29	20
16-Jan-04	2	Control	044662	Ham	9	5.2	3.7	171	55	113	10.4	37	799	2635	11	73	16.6	35	0.0	1.1	144	5.1	104	26	19
16-Mar-04	2	Clone	023	Ham	14	6.3	2.9	181	52	98	9.9	22	314	1130	4	91	132.6	42	2.8	1.6	143	5.1	101	28	19
16-Mar-04	2	Clone	024	Duroc	10	6.6	4.1	111	68	107	10.8	38	686	2793	13	79	19.7	12	0.0	2.4	146	6.2	103	29	20
16-Mar-04	2	Control	027446	Ham	11	5.6	3.7	139	47	96	10.1	23	790	1990	10	84	46.2	39	2.0	1.7	146	4.8	104	27	20
16-Mar-04	2	Control	044584	Ham	13	6.3	3.7	106	56	100	10.5	24	718	3110	10	74	53.2	33	1.7	1.5	144	5.3	103	28	18
16-Mar-04	2	Control	044615	Ham	13	6.2	3.9	122	60	103	10.3	68	4559	2336	12	75	28.4	31	0.1	2.3	144	5.9	103	28	19
16-Mar-04	2	Control	044661	Ham	13	6.2	3.7	159	70	103	10.7	33	749	4207	17	74	40.2	32	1.0	1.8	148	5.9	103	28	23
16-Mar-04	2	Control	044662	Ham	9	6.2	3.8	109	48	100	10.1	23	332	2689	12	79	29.0	34	2.3	1.7	143	4.2	102	26	19
27-May-04	2	Clone	023	Ham	13	5.9	3.3	141	40	70	9.8	27	883	1070	5	106	29.2	35	0.0	2.2	142	4.2	97	30	20
27-May-04	2	Clone	024	Duroc	20	9.0	4.3	92	72	62	10.9	64	1334	2623	12	101	21.1	15	5.5	1.9	145	7.7	100	27	26
27-May-04	2	Control	027446	Ham	29	7.6	4.7	158	161	102	11.7	459	36792	2272	27	147	29.3	55	3.6	2.6	154	8.1	104	17	41
27-May-04	2	Control	044584	Ham	21	6.8	3.9	74	57	85	10.3	58	7604	2773	8	97	28.5	55	0.5	2.1	147	8.0	104	28	23
27-May-04	2	Control	044661	Ham	18	7.4	4.6	173	64	114	11.1	45	2677	4432	28	86	33.4	46	0.0	2.0	146	7.5	103	23	28
27-May-04	2	Control	044615	Ham	16	7.1	5.0	149	73	112	11.4	35	2302	2640	15	98	24.5	43	0.0	2.1	147	8.3	102	20	33
27-May-04	2	Control	044662	Ham	16	7.2	4.6	123	44	102	10.8	33	1375	2844	26	96	23.8	40	1.8	1.8	147	6.9	103	24	27
Trial Date	Trt	#/TRT	Urea Nitrogen	Total Protein	Albumin	Alk Phos	ALT	Glucose	Calcium	AST	Creatine Kinase	Amylase	Lipase	Cholesterol	Bile Acids	GGTP	SDH	Creatinine	Sodium	Potassium	Chloride	Bicarb.	Anion GAP		
1	5-Dec-03	Clone	5	15.40	4.64	3.06	195.40	38.80	91.40	9.62	44.40	601.80	1113.40	22.20	85.00	17.54	83.00	0.00	1.54	144.00	5.70	104.20	22.40	23.20	
1	5-Dec-03	Control	10	12.00	5.28	3.95	248.40	75.70	118.40	10.96	41.40	1201.00	3179.80	14.00	59.30	19.10	54.40	0.00	1.17	146.30	6.02	103.00	23.90	25.50	
1	23-Jan-04	Clone	5	16.20	5.86	3.12	162.80	38.60	103.60	10.02	28.00	759.80	1176.20	9.00	102.20	41.88	48.80	0.88	1.84	146.00	5.48	102.20	30.60	18.80	
1	23-Jan-04	Control	10	12.40	5.55	3.73	133.10	44.30	99.90	10.37	17.10	362.70	2928.20	11.10	80.30	31.42	41.10	0.15	1.48	148.30	5.23	103.70	31.00	18.80	
1	8-Apr-04	Clone	5	18.60	6.58	4.00	127.80	63.00	91.40	9.74	92.40	5796.00	1166.40	10.40	97.60	45.14	49.80	11.56	2.68	145.40	6.82	101.40	28.80	21.60	
1	8-Apr-04	Control	10	18.20	7.40	4.85	144.00	63.50	89.70	10.59	50.90	3484.20	3368.30	17.10	107.70	29.92	39.60	3.09	1.99	144.20	7.00	101.30	27.80	22.10	
2	16-Jan-04	Clone	2	22.50	6.15	3.90	169.00	58.00	134.50	11.20	44.50	993.00	1605.50	6.00	82.00	21.40	36.00	0.20	1.75	136.00	5.65	97.00	21.00	23.50	
2	16-Jan-04	Control	5	11.40	5.14	3.72	220.20	64.80	115.80	10.84	51.20	1601.40	2995.40	10.00	66.80	21.96	42.60	0.00	1.34	144.20	5.48	102.60	26.20	20.80	
2	16-Mar-04	Clone	2	12.00	6.45	3.50	146.00	60.00	102.50	10.35	30.00	500.00	1961.50	8.50	85.00	76.15	27.00	1.40	2.00	144.50	5.65	102.00	28.50	19.50	
2	16-Mar-04	Control	5	11.80	6.10	3.76	127.00	56.20	100.40	10.34	34.20	1429.60	2866.40	12.20	77.20	39.40	33.80	1.42	1.80	145.00	5.22	103.00	27.40	19.80	
2	27-May-04	Clone	2	16.50	7.45	3.80	116.50	56.00	66.00	10.35	45.50	1108.50	1846.50	8.50	103.50	25.15	25.00	2.75	2.05	143.50	5.95	98.50	28.50	23.00	
2	27-May-04	Control	5	20.00	7.22	4.56	135.40	79.80	103.00	11.06	126.00	10150.00	2992.20	20.80	104.80	27.90	47.80	1.18	2.12	148.20	7.76	103.20	22.40	30.40	
	Literature Ra			8.2-24.6	5.8-8.3	2.3-4.0	41.0-176.1	21.7-46.5	66.4-116.1	9.3-11.5	15.3-55.3		43.5-88.0		81.4-134.1		31.0-52.0	0.5-4.9	0.8-2.3	139.2-152.5	4.4-6.5	97.1-106.4	18.0-27.0		

Collection Date	Trial #	Trt	Barrow ID	Line	Measurement Units	
					Estradiol	IGF-1
					pg/ml	nmol/L
5-Dec-03	1	Clone	018	Ham	6.82	6
5-Dec-03	1	Clone	019	Ham	10.91	15
5-Dec-03	1	Clone	020	Ham	10.21	9
5-Dec-03	1	Clone	021	Ham	6.27	3
5-Dec-03	1	Clone	022	Ham	9.48	18
5-Dec-03	1	Control	025456	Ham	11.27	21
5-Dec-03	1	Control	025463	Ham	14.03	9
5-Dec-03	1	Control	025413	Ham	6.02	15
5-Dec-03	1	Control	025461	Ham	57.59	41
5-Dec-03	1	Control	025436	Ham	12.97	30
5-Dec-03	1	Control	025462	Ham	11.89	30
5-Dec-03	1	Control	025415	Ham	11.31	24
5-Dec-03	1	Control	025457	Ham	8.47	30
5-Dec-03	1	Control	025416	Ham	12.59	24
5-Dec-03	1	Control	025437	Ham	14.81	56
8-Apr-04	1	Clone	018	Ham	17.84	12
8-Apr-04	1	Clone	019	Ham	18.71	3
8-Apr-04	1	Clone	020	Ham	16.06	3
8-Apr-04	1	Control	025456	Ham	28.87	15
8-Apr-04	1	Control	025463	Ham	21.45	9
8-Apr-04	1	Control	025413	Ham	16.81	9
8-Apr-04	1	Control	025461	Ham	96.00	24
8-Apr-04	1	Control	025436	Ham	19.16	12
8-Apr-04	1	Control	025462	Ham	19.00	12
8-Apr-04	1	Control	025415	Ham	11.72	9
8-Apr-04	1	Control	025457	Ham	22.18	9
8-Apr-04	1	Control	025416	Ham	17.26	3
8-Apr-04	1	Control	025437	Ham	14.24	15
8-Apr-04	1	Clone	021	Ham	13.76	9
8-Apr-04	1	Clone	022	Ham	11.05	18
16-Jan-04	2	Clone	023	Ham	11.43	0.0
16-Jan-04	2	Clone	024	Duroc	9.74	44.3
16-Jan-04	2	Control	027446	Ham	8.45	32.5
16-Jan-04	2	Control	044584	Ham	6.71	23.6
16-Jan-04	2	Control	044615	Ham	8.12	26.6
16-Jan-04	2	Control	044661	Ham	8.19	47.2
16-Jan-04	2	Control	044662	Ham	8.29	50.2
27-May-04	2	Clone	023	Ham	9.80	12
27-May-04	2	Clone	024	Duroc	12.87	21
27-May-04	2	Control	027446	Ham	11.30	12
27-May-04	2	Control	044584	Ham	20.43	12
27-May-04	2	Control	044661	Ham	10.79	17.7
27-May-04	2	Control	044615	Ham		
27-May-04	2	Control	044662	Ham		
Date	Trial	TRT	#/TRT	Estradiol	IGF-1	
				pg/ml	nmol/L	
5-Dec-03	1	Clone	5	8.74	10.20	
5-Dec-03	1	Control	10	16.10	28.00	
8-Apr-04	1	Clone	5	15.48	9.00	
8-Apr-04	1	Control	10	26.67	11.70	
16-Jan-04	2	Clone	2	10.59	22.15	
16-Jan-04	2	Control	5	7.95	36.02	
27-May-04	2	Clone	2	11.34	16.50	
27-May-04	2	Control	3	14.17	13.90	

Collection Date	Trial	Trt	Barrow ID	Line	Measurement											
					Urine source	Color	Turbidity	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Nitrite	Leukocytes
4-Dec-03	1	Clone	018	Ham	dipstick	lt. yellow	sl. cloudy	negative	negative	negative	1.030	large +++	7.5	negative	positive	negative
4-Dec-03	1	Clone	019	Ham	dipstick	dk. yellow	cloudy	negative	negative	negative	1.025	small +	5.0	negative	positive	negative
4-Dec-03	1	Clone	020	Ham	dipstick	brn/yellow	cloudy	negative	negative	negative	1.030	moderate	5.0	negative	negative	negative
4-Dec-03	1	Clone	021	Ham	dipstick	yellow	clear	negative	negative	negative	1.015	negative	5.0	30+	negative	negative
4-Dec-03	1	Clone	022	Ham	dipstick	brn/yellow	clear	100	negative	negative	1.020	large+++	5.0	trace	positive	negative
4-Dec-03	1	Control	025456	Ham	dipstick	lt. yellow	clear	negative	negative	negative	1.000	trace	7.5	negative	negative	negative
4-Dec-03	1	Control	025463	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	mod ++	8.0	trace	positive	negative
4-Dec-03	1	Control	025413	Ham	dipstick	lt. yellow	clear	negative	negative	negative	1.005	negative	7.5	negative	positive	negative
4-Dec-03	1	Control	025461	Ham	dipstick	yellow	sl. cloudy	negative	negative	negative	1.010	negative	7.5	negative	negative	negative
4-Dec-03	1	Control	025436	Ham	dipstick	yellow	sl. cloudy	negative	negative	negative	1.005	moderate	8.0	negative	negative	negative
4-Dec-03	1	Control	025462	Ham	dipstick	yellow	clear	negative	negative	negative	1.005	negative	8.0	negative	negative	negative
4-Dec-03	1	Control	025415	Ham	dipstick	lt. yellow	cloudy	negative	negative	negative	1.000	moderate	8.0	negative	positive	negative
4-Dec-03	1	Control	025457	Ham	dipstick	lt. yellow	sl. cloudy	negative	negative	negative	1.000	large +++	7.5	negative	positive	negative
4-Dec-03	1	Control	025416	Ham	dipstick	lt. yellow	clear	negative	negative	negative	1.010	negative	8.0	negative	positive	negative
4-Dec-03	1	Control	025437	Ham	dipstick	dk. yellow	clear	negative	negative	negative	1.015	moderate	7.0	trace	negative	negative
23-Jan-04	1	Clone	018	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	negative	7.5	negative	positive	negative
23-Jan-04	1	Clone	019	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	negative	5.0	negative	negative	negative
23-Jan-04	1	Clone	020	Ham	dipstick	yellow	clear	negative	negative	negative	1.020	negative	5.0	negative	negative	negative
23-Jan-04	1	Clone	021	Ham	dipstick	yellow	clear	negative	negative	negative	1.015	negative	6.0	negative	negative	negative
23-Jan-04	1	Clone	022	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	negative	6.5	negative	positive	negative
23-Jan-04	1	Control	025456	Ham	dipstick	yellow	clear	negative	negative	negative	1.015	negative	6.0	negative	negative	negative
23-Jan-04	1	Control	025463	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	negative	8.5	negative	positive	negative
23-Jan-04	1	Control	025413	Ham	dipstick	dk. yellow	cloudy	negative	negative	negative	1.030	large+++	5.0	30+	negative	negative
23-Jan-04	1	Control	025461	Ham	dipstick	yellow	clear	negative	negative	negative	1.020	negative	6.0	negative	negative	negative
23-Jan-04	1	Control	025436	Ham	dipstick	yellow	clear	negative	negative	negative	1.015	negative	7.0	negative	negative	negative
23-Jan-04	1	Control	025462	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	negative	6.0	negative	negative	negative
23-Jan-04	1	Control	025415	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	negative	6.0	negative	negative	negative
23-Jan-04	1	Control	025457	Ham	dipstick	yellow	cloudy	negative	negative	negative	1.005	moderate	8.0	negative	positive	negative
23-Jan-04	1	Control	025416	Ham	dipstick	dk. yellow	cloudy	negative	negative	negative	1.025	large+++	6.5	trace	negative	negative
23-Jan-04	1	Control	025437	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	negative	8.5	negative	negative	negative
8-Apr-04	1	Clone	018	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	moderate	7.0	negative	negative	negative
8-Apr-04	1	Clone	019	Ham	dipstick	yellow	clear	negative	negative	negative	1.005	trace	7.5	trace	negative	negative
8-Apr-04	1	Clone	020	Ham	dipstick	yellow	cloudy	100	negative	negative	1.015	large+++	6.5	30+	negative	negative
8-Apr-04	1	Control	025456	Ham	dipstick	yellow	clear	negative	negative	negative	1.015	negative	6.5	300+++	negative	negative
8-Apr-04	1	Control	025463	Ham	dipstick	yellow	clear	negative	negative	negative	1.015	negative	6.5	trace	negative	negative
8-Apr-04	1	Control	025413	Ham	dipstick	yellow	clear	negative	negative	negative	1.005	negative	7.5	trace	negative	negative
8-Apr-04	1	Control	025461	Ham	dipstick	lt. yellow	clear	negative	negative	negative	1.005	negative	6.5	negative	negative	negative
8-Apr-04	1	Control	025462	Ham	dipstick	yellow	clear	negative	negative	negative	1.005	negative	6.0	negative	negative	negative
8-Apr-04	1	Control	025415	Ham	dipstick	yellow	clear	negative	negative	negative	1.020	negative	6.0	trace	negative	negative
8-Apr-04	1	Control	025457	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	moderate	7.0	30+	negative	negative
8-Apr-04	1	Control	025416	Ham	dipstick	yellow	clear	negative	negative	negative	1.015	moderate	6.5	trace	negative	negative

Collection Date	Trial	TRT	Barrow ID	Line	Measurement											
					Urine source	Color	Turbidity	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Nitrite	Leukocytes
8-Apr-04	1	Control	025437	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	negative	7.0	trace	negative	negative
8-Apr-04	1	Clone	021	Ham	dipstick	yellow	clear	negative	negative	negative	1.005	negative	8.5	trace	positive	negative
8-Apr-04	1	Clone	022	Ham	dipstick*	dk. yellow	clear	negative	negative	negative	1.005	negative	8.0	trace	negative	negative
16-Jan-04	2	Clone	023	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	negative	6.0	negative	negative	negative
16-Jan-04	2	Clone	024	Duroc	dipstick	yellow	clear	negative	negative	negative	1.010	negative	7.5	negative	negative	negative
16-Jan-04	2	Control	027446	Ham	dipstick	yellow	clear	negative	negative	negative	1.020	negative	6.5	negative	negative	negative
16-Jan-04	2	Control	044584	Ham	dipstick	yellow	clear	negative	negative	negative	1.020	negative	6.5	negative	negative	negative
16-Jan-04	2	Control	044615	Ham	dipstick	dk. yellow	clear	negative	negative	negative	1.030	negative	5.0	negative	negative	negative
16-Jan-04	2	Control	044661	Ham	dipstick	dk. yellow	clear	negative	negative	negative	1.030	negative	5.0	negative	negative	negative
16-Jan-04	2	Control	044662	Ham	dipstick	yellow	cloudy	negative	negative	negative	1.010	negative	8.0	30+	negative	negative
16-Mar-04	2	Clone	023	Ham	dipstick	dk. yellow	cloudy	negative	negative	negative	1.020	negative	6.5	trace	negative	negative
16-Mar-04	2	Clone	024	Duroc	dipstick	yellow	clear	negative	negative	negative	1.010	negative	7.0	trace	negative	negative
16-Mar-04	2	Control	027446	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	negative	7.5	trace	negative	negative
16-Mar-04	2	Control	044584	Ham	dipstick	dk. yellow	clear	negative	negative	negative	1.010	negative	7.0	negative	negative	negative
16-Mar-04	2	Control	044615	Ham	dipstick	yellow	clear	negative	negative	negative	1.010	negative	6.5	negative	negative	negative
16-Mar-04	2	Control	044661	Ham	dipstick	yellow	cloudy	negative	negative	negative	1.005	mod ++	8.5	trace	negative	negative
16-Mar-04	2	Control	044662	Ham	dipstick	yellow	clear	negative	negative	negative	1.015	negative	6.5	trace	negative	negative
27-May-04	2	Clone	023	Ham	dipstick	yellow	clear	negative	negative	negative	1.000	trace	8.5	100++	negative	negative
27-May-04	2	Clone	024	Duroc	dipstick	dk. yellow	clear	negative	negative	negative	1.015	negative	6.0	trace	negative	negative
27-May-04	2	Control	027446	Ham	dipstick	dk. yellow	clear	negative	negative	negative	1.020	negative	5.0	trace	negative	negative
27-May-04	2	Control	044584	Ham	dipstick	dk. yellow	clear	negative	negative	negative	1.005	trace	8.5	30+	negative	negative
27-May-04	2	Control	044661	Ham	dipstick	yellow	clear	negative	negative	negative	1.005	trace	8.0	30+	negative	negative
27-May-04	2	Control	044615	Ham	dipstick	brown	cloudy	1/10	negative	negative	1.005	large+++	8.0	30+	negative	trace
27-May-04	2	Control	044662	Ham	dipstick	yellow	clear	negative	negative	negative	1.005	trace	8.0	30+	negative	negative

Date	Trial	TRT	#/TRT	Turbidity	% Positive Animals from total			Specific Gravity	%Positive			% Positive Animals from total		
					Glucose	Bilirubin	Ketone		Blood	pH	Protein	Nitrite	Leukocytes	
4-Dec-03	1	Clone	5		20.00	0.00	0.00	1.02	80.00	5.50	40.00	60.00	0.00	
4-Dec-03	1	Control	10		0.00	0.00	0.00	1.01	70.00	7.70	20.00	50.00	0.00	
23-Jan-04	1	Clone	5		0.00	0.00	0.00	1.01	0.00	6.00	0.00	40.00	0.00	
23-Jan-04	1	Control	10		0.00	0.00	0.00	1.02	30.00	6.75	20.00	20.00	0.00	
8-Apr-04	1	Clone	5		20.00	0.00	0.00	1.01	60.00	7.50	80.00	20.00	0.00	
8-Apr-04	1	Control	9		0.00	0.00	0.00	1.01	22.22	6.61	55.00	0.00	0.00	
16-Jan-04	2	Clone	2		0.00	0.00	0.00	1.01	0.00	6.75	0.00	0.00	0.00	
16-Jan-04	2	Control	5		0.00	0.00	0.00	1.02	0.00	6.20	0.00	0.00	0.00	
16-Mar-04	2	Clone	2		0.00	0.00	0.00	1.02	0.00	6.75	100.00	0.00	0.00	
16-Mar-04	2	Control	5		0.00	0.00	0.00	1.01	20.00	7.20	50.00	0.00	0.00	
27-May-04	2	Clone	2		0.00	0.00	0.00	1.01	50.00	7.25	100.00	0.00	0.00	
27-May-04	2	Control	5		20.00	0.00	0.00	1.01	80.00	7.50	100.00	0.00	0.00	

Treatment	Trial #	Barrow ID	Line	DOB	SOT Date	EOT WT	Slaughter Date	Age (Days)	Hot Carcass WT	% Dress	Fat Thickness (mm)		
											First rib	tenth rib	last rib
Clone	1	Clone O18	Ham	3-Oct-03	6-Apr-04	265	8-Apr-04	186	187.5	70.75	35	18	17
Clone	1	Clone O19	Ham	3-Oct-03	6-Apr-04	244	8-Apr-04	186	170	69.67	33	17	21
Clone	1	Clone O20	Ham	3-Oct-03	6-Apr-04	280	8-Apr-04	186	198.5	70.89	35	16	17
Clone	1	Clone O22	Ham	12-Oct-03	26-May-04	289	27-May-04	227	200	69.20	38	23	27
Control	1	O25456	Ham	11-Oct-03	6-Apr-04	303	8-Apr-04	178	212.5	70.13	40	29	24
Control	1	O25463	Ham	11-Oct-03	6-Apr-04	283	8-Apr-04	178	198.5	70.14	41	24	25
Control	1	O25413	Ham	8-Oct-03	23-Mar-04	280	25-Mar-04	167	195.5	69.82	37	19	20
Control	1	O25461	Ham	11-Oct-03	23-Mar-04	304	25-Mar-04	164	220	72.37	33	25	25
Control	1	O25436	Ham	10-Oct-03	23-Mar-04	268	25-Mar-04	165	181.5	67.72	41	25	24
Control	1	O25462	Ham	11-Oct-03	23-Mar-04	266	25-Mar-04	164	186	69.92	35	18	17
Control	1	O25415	Ham	8-Oct-03	23-Mar-04	293	25-Mar-04	167	204	69.62	40	18	22
Control	1	O25457	Ham	11-Oct-03	23-Mar-04	259	25-Mar-04	164	181	69.88	39	18	22
Control	1	O25416	Ham	8-Oct-03	23-Mar-04	278	25-Mar-04	167	195	70.14	41	24	24
Control	1	O25437	Ham	10-Oct-03	6-Apr-04	280	8-Apr-04	179	196	70.00	39	25	22
Control	2	O27446	Ham	10-Nov-03	26-May-04	286	27-May-04	198	209.5	73.25	38	19	25
Control	2	O44584	Ham	13-Nov-03	26-May-04	310	27-May-04	195	217.4	70.13	46	34	31
Control	2	O44615	Ham	14-Nov-03	19-Apr-04	278	20-Apr-04	157	195.5	70.32	36	19	18
Control	2	O44661	Ham	15-Nov-03	10-May-04	306	11-May-04	177	218.5	71.41	38	18	24
Control	2	O44662	Ham	15-Nov-03	19-Apr-04	268	20-Apr-04	156	181.1	67.57	36	17.5	26
Averages				Trt	Number	EOT WT	Slaughter Date	Age (Days)	Hot Carcass WT	% Dress	Fat Thickness (mm)		
				Clone	4	264.8+7.58a		194.8+5.89a	85.91+3.13a	70.13+0.68a	35.25+1.49a	18.50+2.33a	20.50+1.83a
				Control	15	284.1+4.38b		171.7+3.40b	90.67+1.61a	70.16+0.35a	38.67+0.77a	22.17+1.20a	23.27+0.95a

Note: clone O24 was not included due to a lung adhesion to the body wall at time of slaughter and was condemned; adhesion contained a persistent infection

Barrow ID	Last Lumbar	Loin Eye Area (cm ²)	USDA Carcass Muscle Score	Carcass length (cm)	NPPC Subjective Quality Scores			Hunter Color			Longissimus pH
					Color	Marbling	Firmness	L*	a*	b*	
Clone O18	15	43.56	2	81.1	3	1	3	54.23	8.68	17.46	5.67
Clone O19	16	37.99	2	81.2	3	2	4	50.45	11.48	18.6	5.6
Clone O20	16	48.09	2	83.6	3	1	3	50.49	9.41	17.23	5.65
Clone O22	22	46.5	2	83.8	3	2	4	53.62	8.4	17.09	5.47
O25456	20	47.74	2	88.4	2	2	2	61.21	6.43	17.04	5.7
O25463	25	39.61	2	84.5	3	2	4	55.29	9.66	18.85	5.64
O25413	17	40.33	2	83.4	3	2	3	57.41	6.71	15.85	5.76
O25461	22	51.89	3	85.7	2	1	1	59.68	5.95	16.66	5.6
O25436	23	43.07	2	83.8	3	2	3	56.31	6.91	15.75	5.66
O25462	16	47.6	2	83.2	2	2	2	59.91	5.56	15.94	5.7
O25415	19	42.75	2	85.5	3	2	3	54.69	7.93	16.79	5.62
O25457	20	46.57	2	86.3	2	2	2	61.78	7.44	17.11	5.67
O25416	22	44.75	3	80.1	3	2	3	55.18	7.7	16.88	5.67
O25437	24	43.76	2	86.3	3	3	4	52.2	8.94	17.3	5.66
027446	21	53.2	3	80.1	4	2	4	44.95	8.01	14.04	6.08
044584	28	43.24	2	86.2	3	2	4	55.85	8.44	17.15	5.56
044615	20	49.38	2	85	3	2	3	54.4	8.63	17.84	5.52
044661	20	49.4	2	88.7	3	2	3	54.19	8.5	18.05	5.58
044662	18	44.14	2	80.5	2	1	2	61.55	7.11	17.74	5.57
Trt	Last Lumbar	Loin Eye Area (cm ²)	USDA Carcass Muscle Score	Carcass length (cm)	NPPC Subjective Quality Scores			Hunter Color			Longissimus pH
Clone	17.25+1.58a	44.04+2.04a	2.0+0.19a	82.43+1.27a	3.00+0.27a	1.50+0.24a	3.50+0.43a	52.20+2.02a	9.49+0.60a	17.60+0.54a	5.60+0.06a
Control	21.00+0.81b	45.83+1.06a	2.2+0.10a	84.51+0.66a	2.73+0.14a	1.93+0.12a	2.87+0.22a	56.30+1.04a	7.59+0.31b	16.87+0.28b	5.67+0.03a

Treatment	Trial #	Barrow ID	Line	DOB	EOT Date	EOT WT (lbs)	Slaughter Date	Hot Carcass WT (lbs)	Lungs (lbs)	% BWT1	Heart (lbs)	%BWT2	Kidney (BOTH; lbs)	%BWT3	Liver (lbs)	%BWT4	Spleen (lbs)	%BWT5
Clone	1	Clone O18	Ham	3-Oct-03	6-Apr-04	265	8-Apr-04	187.5	1.7	0.64	1.02	0.38	0.64	0.24	3.98	1.50	0.32	0.12
Clone	1	Clone O19	Ham	3-Oct-03	6-Apr-04	244	8-Apr-04	170	1.76	0.72	1	0.41	0.66	0.27	4.02	1.65	0.3	0.12
Clone	1	Clone O20	Ham	3-Oct-03	6-Apr-04	280	8-Apr-04	198.5	1.86	0.66	1.16	0.41	0.72	0.26	5.04	1.80	0.3	0.11
Clone	1	Clone O22	Ham		26-May-04	285	27-May-04	200	1.62	0.57	1.14	0.40	0.66	0.23	4.8	1.68	0.34	
Control	1	O25456	Ham	11-Oct-03	6-Apr-04	303	8-Apr-04	212.5	1.72	0.57	1.16	0.38	1.08	0.36	4.54	1.50	0.4	0.13
Control	1	O25463	Ham	11-Oct-03	6-Apr-04	283	8-Apr-04	198.5	1.76	0.62	1.18	0.42	0.86	0.30	4.3	1.52	0.4	0.14
Control	1	O25413	Ham	8-Oct-03	23-Mar-04	280	25-Mar-04	195.5	2.36	0.84	1.06	0.38	0.9	0.32	4.18	1.49	0.42	0.15
Control	1	O25461	Ham	11-Oct-03	23-Mar-04	304	25-Mar-04	220	2.18	0.72	1.1	0.36	1.12	0.37	4.86	1.60	0.44	0.14
Control	1	O25436	Ham	10-Oct-03	23-Mar-04	268	25-Mar-04	181.5	1.7	0.63	0.96	0.36	0.9	0.34	4.52	1.69	0.24	0.09
Control	1	O25462	Ham	11-Oct-03	23-Mar-04	266	25-Mar-04	186	1.3	0.49	1.14	0.43	0.96	0.36	4.28	1.61	0.3	0.11
Control	1	O25415	Ham	8-Oct-03	23-Mar-04	293	25-Mar-04	204	2.44	0.83	1.26	0.43	1.04	0.35	4.96	1.69	0.44	0.15
Control	1	O25457	Ham	11-Oct-03	23-Mar-04	259	25-Mar-04	181	2.4	0.93	1.06	0.41	0.92	0.36	4.16	1.61	0.42	0.16
Control	1	O25416	Ham	8-Oct-03	23-Mar-04	278	25-Mar-04	195	2.06	0.74	0.88	0.32	0.74	0.27	4.26	1.53	0.42	0.15
Control	1	O25437	Ham	10-Oct-03	6-Apr-04	280	8-Apr-04	196	1.76	0.63	1.12	0.40	0.96	0.34	4.38	1.56	0.34	0.12
Clone	2	Clone O24	Duroc	14-Nov-03	26-May-04	246	27-May-04			0.00	1.1	0.45	0.8	0.33	3.82	1.55	0.3	0.12
Control	2	O27446	Ham	10-Nov-03	26-May-04	286	27-May-04	209.5	1.96	0.69	1.12	0.39	0.74	0.26	4.06	1.42	0.36	0.13
Control	2	O44584	Ham	13-Nov-03	26-May-04	310	27-May-04	217.4	1.62	0.52	0.92	0.30	0.94	0.30	4.96	1.60	0.34	0.11
Control	2	O44615	Ham	14-Nov-03	19-Apr-04	278	20-Apr-04	195.5	1.98	0.71	1.28	0.46	0.96	0.35	5.94	2.14	0.36	0.13
Control	2	O44661	Ham	15-Nov-03	10-May-04	306	11-May-04	218.5	2.04	0.67	1.66	0.54	1.14	0.37	4.9	1.60	0.36	0.12
Control	2	O44662	Ham	15-Nov-03	19-Apr-04	268	20-Apr-04	181.5	2.14	0.80	1.02	0.38	0.98	0.37	4.9	1.83	0.4	0.15

Trt	#/Trt	EOT WT (lbs)	Hot Carcass WT (lbs)	Lungs (lbs)	% BWT1	Heart (lbs)	%BWT2	Kidney (BOTH; lbs)	%BWT3	Liver (lbs)	%BWT4	Spleen (lbs)	%BWT5
Clone	4	258.75	185.33	1.77	0.51	1.07	0.41	0.71	0.28	4.22	1.63	0.31	0.12
Control	15	284.13	199.49	1.96	0.69	1.13	0.40	0.95	0.33	4.61	1.63	0.38	0.13

Trt	#/Trt	EOT WT (kgs)	Hot Carcass WT (kgs)	Lungs (kgs)	% BWT1	Heart (kgs)	%BWT2	Kidney (BOTH; kgs)	%BWT3	Liver (kgs)	%BWT4	Spleen (kgs)	%BWT5
Clone	5	119.55+4.26a	84.24+3.62a	0.81+0.08a	0.31+0.03a	0.5+0.04a	0.18+0.01a	0.31+0.03a	0.12+0.01a	1.98+0.13a	0.75+0.04a	0.14+0.01a	0.05+0.00a
Control	15	129.15+1.91a	90.68+1.62a	0.89+0.04a	0.31+0.01a	0.5+0.02a	0.18+0.01a	0.43+0.01b	0.15+0.00b	2.10+0.06a	0.74+0.02a	0.17+0.01a	0.06+0.00a

Treatment	Trial	Barrow ID	Line	Date	Amino Acid Profile (excluding Tryptophan)																
					Aspartic Acid %	Threonine %	Serine %	Glutamic Acid %	Proline %	Glycine %	Alanine %	Cystine %	Valine %	Methionine %	Isoleucine %	Leucine %	Tyrosine %	Phenylalanine %	Histidine %	Lysine, Total %	Arginine %
Clone	1	Clone O18	Ham	4/8/2004	2.75	1.15	0.92	3.57	0.86	0.97	1.29	0.27	1.14	0.59	1.07	1.79	0.79	0.88	1.03	1.97	1.43
Clone	1	Clone O19	Ham	4/8/2004	2.76	1.12	0.91	3.44	0.88	1.05	1.28	0.26	1.11	0.56	1.04	1.74	0.77	0.86	1.00	1.91	1.43
Clone	1	Clone O20	Ham	4/8/2004	2.74	1.14	0.92	3.43	0.85	0.98	1.28	0.27	1.13	0.58	1.06	1.78	0.78	0.88	1.03	1.95	1.43
Clone	1	Clone O22	Ham	5/27/2004	2.32	1.10	0.87	3.37	0.81	0.95	1.25	0.24	1.12	0.54	1.06	1.74	0.77	0.86	0.99	1.89	1.41
Control	1	O25456	Ham	4/8/2004	2.81	1.16	0.93	3.60	0.88	1.00	1.29	0.27	1.15	0.57	1.07	1.78	0.79	0.89	1.03	1.96	1.45
Control	1	O25463	Ham	4/8/2004	2.73	1.13	0.91	3.40	0.86	1.01	1.26	0.22	1.10	0.54	1.03	1.75	0.77	0.87	0.98	1.92	1.43
Control	1	O25413	Ham	3/25/2004	2.21	1.12	0.92	3.35	0.85	0.92	1.26	0.28	1.06	0.60	1.01	1.76	0.79	0.87	0.97	1.94	1.43
Control	1	O25461	Ham	3/25/2004	2.24	1.13	0.92	3.40	0.86	0.92	1.27	0.28	1.09	0.60	1.03	1.79	0.80	0.89	1.04	1.97	1.46
Control	1	O25436	Ham	3/25/2004	2.33	1.14	0.93	3.40	0.88	0.99	1.29	0.28	1.10	0.59	1.03	1.79	0.80	0.89	1.03	1.96	1.47
Control	1	O25462	Ham	3/25/2004	2.33	1.15	0.92	3.39	0.88	0.95	1.28	0.28	1.10	0.59	1.04	1.79	0.80	0.89	1.05	1.98	1.47
Control	1	O25415	Ham	3/25/2004	2.37	1.17	0.95	3.57	0.93	1.02	1.34	0.28	1.14	0.61	1.06	1.83	0.82	0.92	1.10	2.01	1.51
Control	1	O25457	Ham	3/25/2004	2.30	1.15	0.93	3.53	0.87	0.94	1.29	0.27	1.11	0.59	1.05	1.81	0.81	0.90	1.01	2.00	1.50
Control	1	O25416	Ham	3/25/2004	2.36	1.17	0.94	3.46	0.88	0.97	1.31	0.29	1.12	0.60	1.04	1.81	0.81	0.91	1.11	2.01	1.49
Control	1	O25437	Ham	4/8/2004	2.74	1.12	0.93	3.37	0.93	1.10	1.30	0.27	1.11	0.58	1.03	1.74	0.77	0.88	1.02	1.92	1.44
Clone	2	Clone O24	Duroc	5/27/2004	2.17	1.04	0.86	3.26	0.83	0.95	1.19	0.20	1.02	0.45	0.96	1.66	0.73	0.82	0.90	1.81	1.36
Control	2	027446	Ham	5/27/2004	2.54	1.18	0.97	3.58	0.99	1.27	1.41	0.26	1.19	0.59	1.09	1.84	0.81	0.93	1.08	1.98	1.56
Control	2	044584	Ham	5/27/2004	2.31	1.10	0.89	3.36	0.77	0.94	1.23	0.23	1.09	0.52	1.02	1.71	0.76	0.85	0.93	1.85	1.39
Control	2	044615	Ham	4/20/2004	2.45	1.12	0.90	3.59	0.95	1.04	1.32	0.24	1.17	0.61	1.11	1.82	0.80	0.90	1.02	2.01	1.50
Control	2	044661	Ham	5/11/2004	2.27	1.11	0.91	3.40	0.98	1.12	1.34	0.23	1.15	0.55	1.06	1.80	0.78	0.89	1.05	1.98	1.48
Control	2	044662	Ham	4/20/2004	2.40	1.10	0.89	3.54	1.01	1.13	1.33	0.23	1.15	0.60	1.08	1.78	0.77	0.89	1.00	1.97	1.50

Trt	# per TRT	Aspartic Acid %	Threonine %	Serine %	Glutamic Acid %	Proline %	Glycine %	Alanine %	Cystine %	Valine %	Methionine %	Isoleucine %	Leucine %	Tyrosine %	Phenylalanine %	Histidine %	Lysine, Total %	Arginine %
Clone	5	2.55+0.10a	1.11+0.01a	0.10+0.01a	3.41+0.04a	0.85+0.03a	0.98+0.04a	1.26+0.02a	0.25+0.01a	1.10+0.02a	0.54+0.02a	1.04+0.01a	1.74+0.02a	0.77+0.01a	0.86+0.01a	0.99+0.02a	1.91+0.02a	1.41+0.02a
Control	15	2.42+0.06a	1.14+0.06a	0.92+0.01b	3.46+0.03a	0.90+0.01a	1.02+0.02a	1.30+0.01a	0.26+0.01a	1.12+0.01a	0.58+0.01b	1.05+0.01a	1.79+0.01b	0.79+0.00b	0.89+0.01b	1.03+0.01a	1.96+0.01b	1.47+0.01b

Treatment	Trial	Barrow ID	Line	Date	C08:0 Octanoic (Caprylic) %	C10:0 Decanoic (Capric) %	C11:0 Undecanoic (Hendecanoic) %	C12:0 Dodecanoic (Lauric) %	C14:0 Tetradeconoic (Myristic) %	C14:1 Tetradeconoic (Myristoleic) %	C15:0 Pentadeconoic %	C15:1 Pentadeconoic %	C16:0 Hexadecanoic (Palmitic) %	C16:1 Hexadecenoic (Palmitoleic) %	C17:0 Heptadecanoic (Margaric) %
Clone	1	Clone O18	Ham	4/8/2004	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.65	0.09	0.00
Clone	1	Clone O19	Ham	4/8/2004	0.00	0.00	0.00	0.00	0.06	0.00	0.00	0.00	0.84	0.12	0.00
Clone	1	Clone O20	Ham	4/8/2004	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.64	0.09	0.00
Clone	1	Clone O22	Ham	5/27/2004	0.00	0.00	0.00	0.00	0.14	0.00	0.00	0.00	2.15	0.19	0.02
Control	1	O25456	Ham	4/8/2004	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	1.03	0.08	0.00
Control	1	O25463	Ham	4/8/2004	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.90	0.11	0.00
Control	1	O25413	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.92	0.08	0.00
Control	1	O25461	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.03	0.00	0.00	0.00	0.58	0.05	0.00
Control	1	O25436	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.92	0.12	0.00
Control	1	O25462	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.95	0.08	0.01
Control	1	O25415	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.03	0.00	0.00	0.00	0.55	0.04	0.00
Control	1	O25457	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.06	0.00	0.00	0.00	1.09	0.10	0.00
Control	1	O25416	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.03	0.00	0.00	0.00	0.65	0.06	0.00
Control	1	O25437	Ham	4/8/2004	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.88	0.11	0.00
Clone	2	024	Duroc	5/27/2004	0.00	0.00	0.00	0.00	0.15	0.00	0.00	0.00	2.26	0.20	0.02
Control	2	027446	Ham	5/27/2004	0.00	0.00	0.00	0.00	0.07	0.00	0.00	0.00	1.23	0.11	0.01
Control	2	044584	Ham	5/27/2004	0.00	0.01	0.00	0.00	0.17	0.00	0.00	0.00	2.57	0.19	0.03
Control	2	044615	Ham	4/20/2004	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.66	0.08	0.00
Control	2	044661	Ham	5/11/2004	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.64	0.07	0.00
Control	2	044662	Ham	4/20/2004	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.62	0.07	0.00

TRT	# per TRT	C08:0 Octanoic (Caprylic) %	C10:0 Decanoic (Capric) %	C11:0 Undecanoic (Hendecanoic) %	C12:0 Dodecanoic (Lauric) %	C14:0 Tetradeconoic (Myristic) %	C14:1 Tetradeconoic (Myristoleic) %	C15:0 Pentadeconoic %	C15:1 Pentadeconoic %	C16:0 Hexadecanoic (Palmitic) %	C16:1 Hexadecenoic (Palmitoleic) %	C17:0 Heptadecanoic (Margaric) %
Clone	5	0	0.00+0.001a	0	0	0.086+0.018a	0	0	0	1.308+0.261a	0.138+0.018a	0.008+0.004a
Control	15	0	0.001+0.001a	0	0	0.053+0.010a	0	0	0	0.946+0.151a	0.090+0.011b	0.003+0.002a

Treatment	Trial	Barrow ID	C17:1 Heptadecenoic Margaroleic %	C18:0 Octadecanoic (Stearic) %	C18:1 Octadecenoic (Oleic) %	C18:2 Octadecadienoic (Linoleic) %	C18:3 Octadecatrienoic (Linolenic) %	C18:4 Octadecatetraenoic %	C20:0 Eicosanoic (Arachidic) %	C20:1 Eicosenoic (Gadoleic) %	C20:2 Eicosadienoic %	C20:3 Eicosatrienoic %
Clone	1	Clone O18	0.00	0.34	1.09	0.30	0.02	0.00	0.00	0.03	0.01	0.00
Clone	1	Clone O19	0.00	0.42	1.51	0.32	0.01	0.00	0.00	0.04	0.02	0.00
Clone	1	Clone O20	0.00	0.32	1.13	0.33	0.02	0.00	0.00	0.03	0.01	0.00
Clone	1	Clone O22	0.02	1.05	2.81	0.14	0.00	0.00	0.03	0.09	0.00	0.00
Control	1	O25456	0.00	0.70	1.78	0.25	0.01	0.00	0.01	0.04	0.01	0.00
Control	1	O25463	0.00	0.45	1.56	0.21	0.00	0.00	0.01	0.04	0.01	0.00
Control	1	O25413	0.00	0.60	1.63	0.28	0.02	0.00	0.00	0.05	0.02	0.00
Control	1	O25461	0.00	0.39	0.95	0.16	0.00	0.00	0.00	0.02	0.00	0.00
Control	1	O25436	0.00	0.46	1.76	0.25	0.01	0.00	0.00	0.05	0.02	0.00
Control	1	O25462	0.00	0.58	1.56	0.22	0.00	0.00	0.00	0.04	0.01	0.00
Control	1	O25415	0.00	0.37	0.79	0.10	0.00	0.00	0.00	0.02	0.00	0.00
Control	1	O25457	0.00	0.65	1.97	0.25	0.01	0.00	0.00	0.05	0.01	0.00
Control	1	O25416	0.00	0.43	1.16	0.21	0.01	0.00	0.00	0.03	0.01	0.00
Control	1	O25437	0.00	0.44	1.65	0.23	0.01	0.00	0.00	0.05	0.01	0.00
Clone	2	O24	0.01	1.16	2.67	0.20	0.00	0.00	0.02	0.08	0.01	0.00
Control	2	O27446	0.00	0.74	1.75	0.13	0.00	0.00	0.01	0.04	0.00	0.00
Control	2	O44584	0.02	1.39	2.71	0.13	0.00	0.00	0.02	0.08	0.00	0.00
Control	2	O44615	0.00	0.36	1.08	0.17	0.00	0.00	0.00	0.02	0.00	0.00
Control	2	O44661	0.00	0.34	1.11	0.17	0.00	0.00	0.00	0.03	0.00	0.00
Control	2	O44662	0.00	0.34	0.95	0.13	0.00	0.00	0.00	0.03	0.00	0.00

C17:1 Heptadecenoic Margaroleic %	C18:0 Octadecanoic (Stearic) %	C18:1 Octadecenoic (Oleic) %	C18:2 Octadecadienoic (Linoleic) %	C18:3 Octadecatrienoic (Linolenic) %	C18:4 Octadecatetraenoic %	C20:0 Eicosanoic (Arachidic) %	C20:1 Eicosenoic (Gadoleic) %	C20:2 Eicosadienoic %	C20:3 Eicosatrienoic %
0.006+0.003a	0.658+0.137a	1.842+0.264a	0.258+0.028a	0.010+0.003a	0	0.010+0.004a	0.054+0.009a	0.010+0.003a	0
0.001+0.001a	0.549+0.079a	1.494+0.153a	0.193+0.016a	0.005+0.002a	0	0.003+0.002a	0.039+0.005a	0.007+0.002a	0

Treatment	Trial	Barrow ID	C20:4 Eicosatetraenoic (Arachidonic) %	C20:5 Eicosapentaenoic %	C21:5 Henicosapentaenoic c %	C22:0 Docosanoic (Behenic) %	C22:1 Docosenoic (Erucic) %	C22:2 Docosadienoic %	C22:3 Docosatienoic %	C22:4 Docosatetraenoic %	C22:5 Docosapentaenoic %	C22:6 Docosahexaenoic %	C24:0 Tetracosanoic (Lignoceric) %	C24:1 Tetracosenoic (Nervonic) %
Clone	1	Clone O18	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Clone	1	Clone O19	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Clone	1	Clone O20	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Clone	1	Clone O22	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.07	0.00	0.00
Control	1	O25456	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25463	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25413	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25461	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25436	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25462	0.00	0.00	0.00	0.00	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25415	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25457	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.00
Control	1	O25416	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25437	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Clone	2	024	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.00
Control	2	027446	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.00	0.00
Control	2	044584	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.05	0.00	0.00
Control	2	044615	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	2	044661	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	2	044662	0.00	0.00	0.00	0.00	0.01	0.00	0.00	0.00	0.00	0.01	0.00	0.00

C24:1 Tetracosenoic (Nervonic) %	C24:0 Tetracosanoic (Lignoceric) %	C22:6 Docosahexaenoic %	C22:5 Docosapentaenoic %	C22:4 Docosatetraenoic %	C22:3 Docosatienoic %	C22:2 Docosadienoic %	C22:1 Docosenoic (Erucic) %	C22:0 Docosanoic (Behenic) %	C21:5 Henicosapentaenoic c %	C20:5 Eicosapentaenoic %	C20:4 Eicosatetraenoic (Arachidonic) %
0	0	0.016+0.008a	0	0	0	0	0.000+0.001a	0	0	0	0
0	0	0.006+0.005a	0	0	0	0	0.001+0.001a	0	0	0	0

**Proximate Analysis
Fatty Acids
Experiment 1, Trials 1 + 2**

Treatment	Trial	Barrow ID	Line	Date	Calcium	Iron	Phosphor	Zinc	Cholesterol	Niacin	Vitamin B6	Vitamin B12
					%	%	%	%	mg/100 g	mg/100 g	mcg/100 g	mg/100 g
Clone	2	024	Duroc	5/27/2004	0.0033	0.0005	0.2	0.0014	67.4	9.69	0.466	0.56
Clone	1	Clone O18	Ham	4/8/2004	0.0036	0.00061	0.21	0.0016	54.2	11.3	0.319	0
Clone	1	Clone O19	Ham	4/8/2004	0.0038	0.00066	0.2	0.0017	54.8	11.4	0.363	0
Clone	1	Clone O20	Ham	4/8/2004	0.0039	0.00063	0.2	0.0014	50.3	11.7	0.37	0
Clone	2	Clone 022	Ham	5/27/2004	0.0033	0.00037	0.19	0.0013	50.8	10.4	0.531	0.468
Control	1	O25456	Ham	4/8/2004	0.0047	0.0006	0.2	0.0014	55.5	9.87	0.262	0
Control	1	O25463	Ham	4/8/2004	0.0040	0.00054	0.21	0.0014	50.4	11.5	0.348	0
Control	1	O25413	Ham	3/25/2004	0.0084	0.00033	0.2	0.0011	57.3	11.9	0.43	0
Control	1	O25461	Ham	3/25/2004	0.0032	0.00043	0.22	0.0011	54.8	10.5	0.38	0
Control	1	O25436	Ham	3/25/2004	0.0032	0.00041	0.21	0.0011	53.5	12.6	0.39	0
Control	1	O25462	Ham	3/25/2004	0.0036	0.00039	0.2	0.001	52.6	12.7	0.46	0
Control	1	O25415	Ham	3/25/2004	0.0036	0.001	0.22	0.0013	48.4	14.3	0.45	0
Control	1	O25457	Ham	3/25/2004	0.0040	0.0006	0.21	0.001	57.7	10.2	0.47	0
Control	1	O25416	Ham	3/25/2004	0.0032	0.00055	0.21	0.0012	51.6	12.9	0.46	0
Control	1	O25437	Ham	4/8/2004	0.0041	0.00054	0.2	0.0014	52.3	10.6	0.432	0
Control	2	027446	Ham	5/27/2004	0.0140	0.00	0.20	0.00	52.80	9.05	0.61	0.00
Control	2	044584	Ham	5/27/2004	0.0034	0.00037	0.2	0.0011	52.2	8.7	0.684	0
Control	2	044615	Ham	4/20/2004	0.0043	<0.0002	0.21	0.0014	49.1	11	0.469	0
Control	2	044661	Ham	5/11/2004	0.0033	0.00051	0.19	0.0012	50.8	9.56	0.716	0
Control	2	044662	Ham	4/20/2004	0.0041	0.0062	0.19	0.0019	53.2	12	0.595	0
					Calcium	Iron	Phosphor	Zinc	Cholesterol	Niacin	Vitamin B6	Vitamin B12
#/trt					%	%	%	%	mg/100 g	mg/100 g	mcg/100 g	mg/100 g
		control	15	mean	0.0047	0.0009	0.2047	0.0013	52.8133	11.1587	0.4767	0.0000
				stdev	0.0029	0.0015	0.0092	0.0002	2.6872	1.5840	0.1241	0.0000
				stdev	0.0007	0.0004	0.0024	0.0001	0.6938	0.4090	0.0321	0.0000
Cloned		Ham	4	mean	0.0037	0.0006	0.2000	0.0015	52.5250	11.2000	0.3958	0.1170
				stdev	0.0003	0.0001	0.0082	0.0002	2.3027	0.5598	0.0929	0.2340
				stdev	0.0001	0.0001	0.0041	0.0001	1.1514	0.2799	0.0465	0.1170
		Duroc	1		0.0033	0.0005	0.2000	0.0014	67.4000	9.6900	0.4660	0.5600

					Amino Acid Profile (excluding Tryptophan)																
					Aspartic Acid	Threonine	Serine	Glutamic Acid	Proline	Glycine	Alanine	Cysteine	Valine	Methionine	Isoleucine	Leucine	Tyrosine	Phenylalanine	Histidine	Lysine, Total	Arginine
Treatment	Trial	Barrow ID	Line	Date	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%
Clone	1	Clone O18	Ham	4/8/2004	2.75	1.15	0.92	3.57	0.86	0.97	1.29	0.27	1.14	0.59	1.07	1.79	0.79	0.88	1.03	1.97	1.43
Clone	1	Clone O19	Ham	4/8/2004	2.76	1.12	0.91	3.44	0.88	1.05	1.28	0.26	1.11	0.56	1.04	1.74	0.77	0.86	1.00	1.91	1.43
Clone	1	Clone O20	Ham	4/8/2004	2.74	1.14	0.92	3.43	0.85	0.98	1.28	0.27	1.13	0.58	1.06	1.78	0.78	0.88	1.03	1.95	1.43
Clone	1	Clone O22	Ham	5/27/2004	2.32	1.10	0.87	3.37	0.81	0.95	1.25	0.24	1.12	0.54	1.06	1.74	0.77	0.86	0.99	1.89	1.41
Control	1	O25456	Ham	4/8/2004	2.81	1.16	0.93	3.60	0.88	1.00	1.29	0.27	1.15	0.57	1.07	1.78	0.79	0.89	1.03	1.96	1.45
Control	1	O25463	Ham	4/8/2004	2.73	1.13	0.91	3.40	0.86	1.01	1.26	0.22	1.10	0.54	1.03	1.75	0.77	0.87	0.98	1.92	1.43
Control	1	O25413	Ham	3/25/2004	2.21	1.12	0.92	3.35	0.85	0.92	1.26	0.28	1.06	0.60	1.01	1.76	0.79	0.87	0.97	1.94	1.43
Control	1	O25461	Ham	3/25/2004	2.24	1.13	0.92	3.40	0.86	0.92	1.27	0.28	1.09	0.60	1.03	1.79	0.80	0.89	1.04	1.97	1.46
Control	1	O25436	Ham	3/25/2004	2.33	1.14	0.93	3.40	0.88	0.99	1.29	0.28	1.10	0.59	1.03	1.79	0.80	0.89	1.03	1.96	1.47
Control	1	O25462	Ham	3/25/2004	2.33	1.15	0.92	3.39	0.88	0.95	1.28	0.28	1.10	0.59	1.04	1.79	0.80	0.89	1.05	1.98	1.47
Control	1	O25415	Ham	3/25/2004	2.37	1.17	0.95	3.57	0.93	1.02	1.34	0.28	1.14	0.61	1.06	1.83	0.82	0.92	1.10	2.01	1.51
Control	1	O25457	Ham	3/25/2004	2.30	1.15	0.93	3.53	0.87	0.94	1.29	0.27	1.11	0.59	1.05	1.81	0.81	0.90	1.01	2.00	1.50
Control	1	O25416	Ham	3/25/2004	2.36	1.17	0.94	3.46	0.88	0.97	1.31	0.29	1.12	0.60	1.04	1.81	0.81	0.91	1.11	2.01	1.49
Control	1	O25437	Ham	4/8/2004	2.74	1.12	0.93	3.37	0.93	1.10	1.30	0.27	1.11	0.58	1.03	1.74	0.77	0.88	1.02	1.92	1.44
Clone	2	024	Duroc	5/27/2004	2.17	1.04	0.86	3.26	0.83	0.95	1.19	0.20	1.02	0.45	0.96	1.66	0.73	0.82	0.90	1.81	1.36
Control	2	027446	Ham	5/27/2004	2.54	1.18	0.97	3.58	0.99	1.27	1.41	0.26	1.19	0.59	1.09	1.84	0.81	0.93	1.08	1.98	1.56
Control	2	044584	Ham	5/27/2004	2.31	1.10	0.89	3.36	0.77	0.94	1.23	0.23	1.09	0.52	1.02	1.71	0.76	0.85	0.93	1.85	1.39
Control	2	044615	Ham	4/20/2004	2.45	1.12	0.90	3.59	0.95	1.04	1.32	0.24	1.17	0.61	1.11	1.82	0.80	0.90	1.02	2.01	1.50
Control	2	044661	Ham	5/11/2004	2.27	1.11	0.91	3.40	0.98	1.12	1.34	0.23	1.15	0.55	1.06	1.80	0.78	0.89	1.05	1.98	1.48
Control	2	044662	Ham	4/20/2004	2.40	1.10	0.89	3.54	1.01	1.13	1.33	0.23	1.15	0.60	1.08	1.78	0.77	0.89	1.00	1.97	1.50

Treatment	# per TRT																				
Clone	5	2.55	1.11	0.90	3.41	0.85	0.98	1.26	0.25	1.10	0.54	1.04	1.74	0.77	0.86	0.99	1.91	1.41			
Control	15	2.43	1.14	0.92	3.46	0.90	1.02	1.30	0.26	1.12	0.58	1.05	1.79	0.79	0.89	1.03	1.96	1.47			

Control	Ham	15	mean	2.43	1.14	0.92	3.46	0.90	1.02	1.30	0.26	1.12	0.58	1.05	1.79	0.79	0.89	1.03	1.96	1.47
			stdev	0.19	0.03	0.02	0.09	0.06	0.10	0.04	0.02	0.04	0.03	0.03	0.04	0.02	0.02	0.05	0.04	0.04
			sterr	0.05	0.01	0.01	0.02	0.02	0.03	0.01	0.01	0.01	0.01	0.01	0.01	0.00	0.01	0.01	0.01	0.01
Clone	Ham	4	mean	2.64	1.13	0.91	3.45	0.85	0.99	1.28	0.26	1.13	0.57	1.06	1.76	0.78	0.87	1.01	1.93	1.43
			stdev	0.22	0.02	0.02	0.08	0.03	0.04	0.02	0.01	0.01	0.02	0.01	0.03	0.01	0.01	0.02	0.04	0.01
			sterr	0.11	0.01	0.01	0.04	0.01	0.02	0.01	0.01	0.01	0.01	0.01	0.01	0.00	0.01	0.01	0.02	0.01
Clone	Duroc	1		2.17	1.04	0.86	3.26	0.83	0.95	1.19	0.2	1.02	0.45	0.96	1.66	0.73	0.82	0.9	1.81	1.36

Nutrient Analysis

Amino Acids

Experiment 1, Trials 1 + 2

Treatment	Trial	Barrow ID	Line	Date	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%	
Clone	1	Clone O18	Ham	4/8/2004	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.65	0.09	0.00	0.00	0.34	1.09	0.30	0.02	0.00
Clone	1	Clone O19	Ham	4/8/2004	0.00	0.00	0.00	0.00	0.06	0.00	0.00	0.00	0.84	0.12	0.00	0.00	0.42	1.51	0.32	0.01	0.00
Clone	1	Clone O20	Ham	4/8/2004	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.64	0.09	0.00	0.00	0.32	1.13	0.33	0.02	0.00
Clone	1	Clone O22	Ham	5/27/2004	0.00	0.00	0.00	0.00	0.14	0.00	0.00	0.00	2.15	0.19	0.02	0.02	1.05	2.81	0.14	0.00	0.00
Control	1	O25456	Ham	4/8/2004	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	1.03	0.08	0.00	0.00	0.70	1.78	0.25	0.01	0.00
Control	1	O25463	Ham	4/8/2004	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.90	0.11	0.00	0.00	0.45	1.56	0.21	0.00	0.00
Control	1	O25413	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.92	0.08	0.00	0.00	0.60	1.63	0.28	0.02	0.00
Control	1	O25461	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.03	0.00	0.00	0.00	0.58	0.05	0.00	0.00	0.39	0.95	0.16	0.00	0.00
Control	1	O25436	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.92	0.12	0.00	0.00	0.46	1.76	0.25	0.01	0.00
Control	1	O25462	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.95	0.08	0.01	0.00	0.58	1.56	0.22	0.00	0.00
Control	1	O25415	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.03	0.00	0.00	0.00	0.55	0.04	0.00	0.00	0.37	0.79	0.10	0.00	0.00
Control	1	O25457	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.06	0.00	0.00	0.00	1.09	0.10	0.00	0.00	0.65	1.97	0.25	0.01	0.00
Control	1	O25416	Ham	3/25/2004	0.00	0.00	0.00	0.00	0.03	0.00	0.00	0.00	0.65	0.06	0.00	0.00	0.43	1.16	0.21	0.01	0.00
Control	1	O25437	Ham	4/8/2004	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.88	0.11	0.00	0.00	0.44	1.65	0.23	0.01	0.00
Clone	2	024	Duroc	5/27/2004	0.00	0.00	0.00	0.00	0.15	0.00	0.00	0.00	2.26	0.20	0.02	0.01	1.16	2.67	0.20	0.00	0.00
Control	2	027446	Ham	5/27/2004	0.00	0.00	0.00	0.00	0.07	0.00	0.00	0.00	1.23	0.11	0.01	0.00	0.74	1.75	0.13	0.00	0.00
Control	2	044584	Ham	5/27/2004	0.00	0.01	0.00	0.00	0.17	0.00	0.00	0.00	2.57	0.19	0.03	0.02	1.39	2.71	0.13	0.00	0.00
Control	2	044615	Ham	4/20/2004	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.66	0.08	0.00	0.00	0.36	1.08	0.17	0.00	0.00
Control	2	044661	Ham	5/11/2004	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.64	0.07	0.00	0.00	0.34	1.11	0.17	0.00	0.00
Control	2	044662	Ham	4/20/2004	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.62	0.07	0.00	0.00	0.34	0.95	0.13	0.00	0.00

Treatment	# per TRT	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%
Clone	5	0.00	0.00	0.00	0.00	0.09	0.00	0.00	0.00	0.00	1.31	0.14	0.01	0.01	0.66	1.84	0.26	0.01	0.00	
Control	15	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.00	0.95	0.09	0.00	0.00	0.55	1.49	0.19	0.00	0.00	

Control	Ham	mean	0.00	0.00	0.00	0.00	0.06	0.00	0.00	0.00	1.03	0.10	0.00	0.00	0.58	1.55	0.19	0.00	0.00
		stdev	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.60	0.05	0.01	0.01	0.31	0.58	0.05	0.01	0.00
		sterr	0.00	0.00	0.00	0.00	0.01	0.00	0.00	0.00	0.15	0.01	0.00	0.00	0.08	0.15	0.01	0.00	0.00
Clone	Ham	mean	0.00	0.00	0.00	0.00	0.07	0.00	0.00	0.00	1.17	0.12	0.01	0.01	0.62	1.81	0.26	0.01	0.00
		stdev	0.00	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.68	0.05	0.01	0.01	0.33	0.72	0.09	0.01	0.00
		sterr	0.00	0.00	0.00	0.00	0.02	0.00	0.00	0.00	0.34	0.02	0.01	0.01	0.16	0.36	0.04	0.00	0.00
Clone	Duroc		0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.65	0.09	0.00	0.00	0.34	1.09	0.30	0.02	0.00

					C20:0 Eicosanoic (Arachidic)	C20:1 Eicosenoic (Gadoleic)	C20:2 Eicosadienoic	C20:3 Eicosatrienoic	C20:4 Eicosatetraenoic (Arachidonic)	C20:5 Eicosapentaenoic	Henicosapentaenoic C21:5	C22:0 Docosanoic (Behenic)	C22:1 Docosenoic (Erucic)	C22:2 Docosadienoic	C22:3 Docosatrenoic	C22:4 Docosatetraenoic	C22:5 Docosapentaenoic	C22:6 Docosahexaenoic	C24:0 Tetracosanoic (Lignoceric)	C24:1 Tetracosenoic (Nervonic)
Treatment	Trial	Barrow ID	Line	Date	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%
Clone	1	Clone O18	Ham	4/8/2004	0.00	0.03	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Clone	1	Clone O19	Ham	4/8/2004	0.00	0.04	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Clone	1	Clone O20	Ham	4/8/2004	0.00	0.03	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Clone	1	Clone O22	Ham	5/27/2004	0.03	0.09	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.07	0.00	0.00
Control	1	O25456	Ham	4/8/2004	0.01	0.04	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25463	Ham	4/8/2004	0.01	0.04	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25413	Ham	3/25/2004	0.00	0.05	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25461	Ham	3/25/2004	0.00	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25436	Ham	3/25/2004	0.00	0.05	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25462	Ham	3/25/2004	0.00	0.04	0.01	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25415	Ham	3/25/2004	0.00	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25457	Ham	3/25/2004	0.00	0.05	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.00
Control	1	O25416	Ham	3/25/2004	0.00	0.03	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	1	O25437	Ham	4/8/2004	0.00	0.05	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Clone	2	024	Duroc	5/27/2004	0.02	0.08	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.00
Control	2	027446	Ham	5/27/2004	0.01	0.04	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.00	0.00
Control	2	044584	Ham	5/27/2004	0.02	0.08	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.05	0.00	0.00
Control	2	044615	Ham	4/20/2004	0.00	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	2	044661	Ham	5/11/2004	0.00	0.03	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control	2	044662	Ham	4/20/2004	0.00	0.03	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.00

Treatment	# per TRT	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%	%
Clone	5	0.01	0.05	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.00	0.00	0.00
Control	15	0.00	0.04	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.00	0.00

Control	Ham	mean	0.00	0.04	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.00	0.00
		stdev	0.01	0.02	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.00
		sterr	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Clone	Ham	mean	0.01	0.05	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.00	0.00	0.00
		stdev	0.01	0.03	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00
		sterr	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.00	0.00	0.00
Clone	Duroc		0.00	0.03	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00

Treatment	Trial	Barrow ID	Line	Date	Calcium %	Iron %	Phosphorus %	Zinc %	Cholesterol mg/100 g	Niacin mg/100 g	Vitamin B6 mcg/100 g	Vitamin B12 mg/100 g
Clone	1	Clone O18	Ham	4/8/2004	0.0036	0.00061	0.21	0.0016	54.2	11.3	0.319	0
Clone	1	Clone O19	Ham	4/8/2004	0.0038	0.00066	0.20	0.0017	54.8	11.4	0.363	0
Clone	1	Clone O20	Ham	4/8/2004	0.0039	0.00063	0.20	0.0014	50.3	11.7	0.370	0
Clone	2	Clone O22	Ham	5/27/2004	0.0033	0.00037	0.19	0.0013	50.8	10.4	0.531	0.468
Control	1	O25456	Ham	4/8/2004	0.0047	0.00060	0.20	0.0014	55.5	9.87	0.262	0
Control	1	O25463	Ham	4/8/2004	0.0040	0.00054	0.21	0.0014	50.4	11.5	0.348	0
Control	1	O25413	Ham	3/25/2004	0.0084	0.00033	0.20	0.0011	57.3	11.9	0.43	0
Control	1	O25461	Ham	3/25/2004	0.0032	0.00043	0.22	0.0011	54.8	10.5	0.38	0
Control	1	O25436	Ham	3/25/2004	0.0032	0.00041	0.21	0.0011	53.5	12.6	0.39	0
Control	1	O25462	Ham	3/25/2004	0.0036	0.00039	0.20	0.0010	52.6	12.7	0.46	0
Control	1	O25415	Ham	3/25/2004	0.0036	0.0010	0.22	0.0013	48.4	14.3	0.45	0
Control	1	O25457	Ham	3/25/2004	0.0040	0.00060	0.21	0.0010	57.7	10.2	0.47	0
Control	1	O25416	Ham	3/25/2004	0.0032	0.00055	0.21	0.0012	51.6	12.9	0.46	0
Control	1	O25437	Ham	4/8/2004	0.0041	0.00054	0.20	0.0014	52.3	10.6	0.432	0
Clone	2	Clone O24	Duroc	5/27/2004	0.0033	0.00050	0.20	0.0014	67.4	9.69	0.466	0.560
Control	2	O27446	Ham	5/27/2004	0.014	0.00050	0.20	0.0013	52.8	9.05	0.605	0.00
Control	2	O44584	Ham	5/27/2004	0.0034	0.00037	0.20	0.0011	52.2	8.70	0.684	0
Control	2	O44615	Ham	4/20/2004	0.0043	<0.0002	0.21	0.0014	49.1	11.0	0.469	0
Control	2	O44661	Ham	5/11/2004	0.0033	0.00051	0.19	0.0012	50.8	9.56	0.716	0
Control	2	O44662	Ham	4/20/2004	0.0041	0.0062	0.19	0.0019	53.2	12.0	0.595	0

TRT	#/TRT	Calcium %	Iron %	Phosphorus %	Zinc %	Cholesterol mg/100 g	Niacin mg/100 g	Vitamin B6 mcg/100 g	Vitamin B12 mg/100 g
Clone	5	0.00358	0.000554	0.2	0.00148	55.5	10.898	0.4098	0.2056
Control	15	0.00474	0.000878	0.20466667	0.00126	52.8133333	11.1586667	0.47673333	0

Control	Ham	15	mean	0.0047	0.0009	0.2047	0.0013	52.8133	11.1587	0.4767	0.0000
			stdev	0.0029	0.0015	0.0092	0.0002	2.6872	1.5840	0.1241	0.0000
			stderr	0.0007	0.0004	0.0024	0.0001	0.6938	0.4090	0.0321	0.0000
Clone	Ham	4	mean	0.0037	0.0006	0.2000	0.0015	52.5250	11.2000	0.3958	0.1170
			stdev	0.0003	0.0001	0.0082	0.0002	2.3027	0.5598	0.0929	0.2340
			stderr	0.0001	0.0001	0.0041	0.0001	1.1514	0.2799	0.0465	0.1170
Clone	Duroc	1		0.0033	0.0005	0.2000	0.0014	67.4000	9.6900	0.4660	0.5600

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200430501	U.S. Meat Animal Research	2	Control	Ham	498	200342304	H	7/7/2004
200430502	U.S. Meat Animal Research	2	Control	Ham	498	200342304	H	7/7/2004
200430503	U.S. Meat Animal Research	2	Control	Ham	498	200342304	H	7/7/2004
200430504	U.S. Meat Animal Research	2	Control	Ham	498	200342304	H	7/7/2004
200430505	U.S. Meat Animal Research	2	Control	Ham	498	200342304	H	7/7/2004
200430506	U.S. Meat Animal Research	2	Control	Ham	498	200342304	H	7/7/2004
200430507	U.S. Meat Animal Research	1	Control	Ham	498	200342304	H	7/7/2004
200430508	U.S. Meat Animal Research	1	Control	Ham	498	200342304	H	7/7/2004
200430509	U.S. Meat Animal Research	1	Control	Ham	498	200342304	H	7/7/2004
200430510	U.S. Meat Animal Research	1	Control	Ham	498	200342304	H	7/7/2004
200430601	U.S. Meat Animal Research	2	Clone	Ham	5	200336705	G	7/7/2004
200430602	U.S. Meat Animal Research	2	Clone	Ham	5	200336705	G	7/7/2004
200430603	U.S. Meat Animal Research	2	Clone	Ham	5	200336705	G	7/7/2004
200430604	U.S. Meat Animal Research	1	Clone	Ham	5	200336705	G	7/7/2004
200430605	U.S. Meat Animal Research	1	Clone	Ham	5	200336705	G	7/7/2004
200430606	U.S. Meat Animal Research	1	Clone	Ham	5	200336705	G	7/7/2004
200430607	U.S. Meat Animal Research	1	Clone	Ham	5	200336705	G	7/7/2004
200430608	U.S. Meat Animal Research	1	Clone	Ham	5	200336705	G	7/7/2004
200430609	U.S. Meat Animal Research	1	Clone	Ham	5	200336705	G	7/7/2004
200430610	U.S. Meat Animal Research	1	Clone	Ham	5	200336705	G	7/7/2004
200430611	U.S. Meat Animal Research	1	Clone	Ham	5	200336705	G	7/7/2004
200430701	U.S. Meat Animal Research	2	Clone	Ham	2	200338205	C	7/7/2004
200430702	U.S. Meat Animal Research	2	Clone	Ham	2	200338205	C	7/7/2004
200430703	U.S. Meat Animal Research	2	Clone	Ham	2	200338205	C	7/7/2004
200430704	U.S. Meat Animal Research	2	Clone	Ham	2	200338205	C	7/7/2004
200430705	U.S. Meat Animal Research	2	Clone	Ham	2	200338205	C	7/7/2004
200430706	U.S. Meat Animal Research	1	Clone	Ham	2	200338205	C	7/7/2004
200430707	U.S. Meat Animal Research	1	Clone	Ham	2	200338205	C	7/7/2004
200430708	U.S. Meat Animal Research	1	Clone	Ham	2	200338205	C	7/7/2004
200430709	U.S. Meat Animal Research	1	Clone	Ham	2	200338205	C	7/7/2004
200430710	U.S. Meat Animal Research	1	Clone	Ham	2	200338205	C	7/7/2004
200430711	U.S. Meat Animal Research	1	Clone	Ham	2	200338205	C	7/7/2004
200430801	U.S. Meat Animal Research	2	Clone	Ham	5	200345501	F	7/7/2004
200430802	U.S. Meat Animal Research	2	Clone	Ham	5	200345501	F	7/7/2004
200430803	U.S. Meat Animal Research	1	Clone	Ham	5	200345501	F	7/7/2004
200430804	U.S. Meat Animal Research	1	Clone	Ham	5	200345501	F	7/7/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200430805	U.S. Meat Animal Research	1	Clone	Ham		5 200345501	F	7/7/2004
200430902	U.S. Meat Animal Research	1	Clone	Ham		3 200340904	F	7/8/2004
200430903	U.S. Meat Animal Research	1	Clone	Ham		3 200340904	F	7/8/2004
200430905	U.S. Meat Animal Research	1	Clone	Ham		3 200340904	F	7/8/2004
200430906	U.S. Meat Animal Research	1	Clone	Ham		3 200340904	F	7/8/2004
200431001	U.S. Meat Animal Research	2	Clone	Ham		2 200344904	D	7/8/2004
200431002	U.S. Meat Animal Research	2	Clone	Ham		2 200344904	D	7/8/2004
200431003	U.S. Meat Animal Research	2	Clone	Ham		2 200344904	D	7/8/2004
200431004	U.S. Meat Animal Research	2	Clone	Ham		2 200344904	D	7/8/2004
200431005	U.S. Meat Animal Research	2	Clone	Ham		2 200344904	D	7/8/2004
200431006	U.S. Meat Animal Research	1	Clone	Ham		2 200344904	D	7/8/2004
200431007	U.S. Meat Animal Research	1	Clone	Ham		2 200344904	D	7/8/2004
200431008	U.S. Meat Animal Research	1	Clone	Ham		2 200344904	D	7/8/2004
200431009	U.S. Meat Animal Research	1	Clone	Ham		2 200344904	D	7/8/2004
200431010	U.S. Meat Animal Research	1	Clone	Ham		2 200344904	D	7/8/2004
200431011	U.S. Meat Animal Research	1	Clone	Ham		2 200344904	D	7/8/2004
200431012	U.S. Meat Animal Research	1	Clone	Ham		2 200344904	D	7/8/2004
200431401	U.S. Meat Animal Research	2	Clone	Ham		5 200344902	D	7/10/2004
200431402	U.S. Meat Animal Research	2	Clone	Ham		5 200344902	D	7/10/2004
200431403	U.S. Meat Animal Research	2	Clone	Ham		5 200344902	D	7/10/2004
200431404	U.S. Meat Animal Research	2	Clone	Ham		5 200344902	D	7/10/2004
200431405	U.S. Meat Animal Research	2	Clone	Ham		5 200344902	D	7/10/2004
200431406	U.S. Meat Animal Research	2	Clone	Ham		5 200344902	D	7/10/2004
200431407	U.S. Meat Animal Research	2	Clone	Ham		5 200344902	D	7/10/2004
200431408	U.S. Meat Animal Research	2	Clone	Ham		5 200344902	D	7/10/2004
200431409	U.S. Meat Animal Research	1	Clone	Ham		5 200344902	D	7/10/2004
200431410	U.S. Meat Animal Research	1	Clone	Ham		5 200344902	D	7/10/2004
200431411	U.S. Meat Animal Research	1	Clone	Ham		5 200344902	D	7/10/2004
200431501	U.S. Meat Animal Research	2	Clone	Ham		2 200340903	F	7/10/2004
200431502	U.S. Meat Animal Research	2	Clone	Ham		2 200340903	F	7/10/2004
200431503	U.S. Meat Animal Research	2	Clone	Ham		2 200340903	F	7/10/2004
200431504	U.S. Meat Animal Research	1	Clone	Ham		2 200340903	F	7/10/2004
200431505	U.S. Meat Animal Research	1	Clone	Ham		2 200340903	F	7/10/2004
200431506	U.S. Meat Animal Research	1	Clone	Ham		2 200340903	F	7/10/2004
200431601	U.S. Meat Animal Research	2	Clone	Ham		2 200343201	A	7/10/2004
200431602	U.S. Meat Animal Research	2	Clone	Ham		2 200343201	A	7/10/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200431603	U.S. Meat Animal Research	2	Clone	Ham		2 200343201	A	7/10/2004
200431604	U.S. Meat Animal Research	2	Clone	Ham		2 200343201	A	7/10/2004
200431605	U.S. Meat Animal Research	2	Clone	Ham		2 200343201	A	7/10/2004
200431606	U.S. Meat Animal Research	1	Clone	Ham		2 200343201	A	7/10/2004
200431607	U.S. Meat Animal Research	1	Clone	Ham		2 200343201	A	7/10/2004
200431608	U.S. Meat Animal Research	1	Clone	Ham		2 200343201	A	7/10/2004
200431610	U.S. Meat Animal Research	1	Clone	Ham		2 200343201	A	7/10/2004
200431701	U.S. Meat Animal Research	2	Clone	Ham		5 200345103	A	7/11/2004
200431702	U.S. Meat Animal Research	1	Clone	Ham		5 200345103	A	7/11/2004
200431703	U.S. Meat Animal Research	1	Clone	Ham		5 200345103	A	7/11/2004
200431704	U.S. Meat Animal Research	1	Clone	Ham		5 200345103	A	7/11/2004
200431706	U.S. Meat Animal Research	1	Clone	Ham		5 200345103	A	7/11/2004
200431708	U.S. Meat Animal Research	1	Clone	Ham		5 200345103	A	7/11/2004
200431801	U.S. Meat Animal Research	2	Control	Duroc	18128	200342905	C	7/12/2004
200431802	U.S. Meat Animal Research	2	Control	Duroc	18128	200342905	C	7/12/2004
200431803	U.S. Meat Animal Research	2	Control	Duroc	18128	200342905	C	7/12/2004
200431804	U.S. Meat Animal Research	1	Control	Duroc	18128	200342905	C	7/12/2004
200431805	U.S. Meat Animal Research	1	Control	Duroc	18128	200342905	C	7/12/2004
200432001	U.S. Meat Animal Research	2	Clone	Ham		2 200337401	E	7/12/2004
200432002	U.S. Meat Animal Research	2	Clone	Ham		2 200337401	E	7/12/2004
200432003	U.S. Meat Animal Research	2	Clone	Ham		2 200337401	E	7/12/2004
200432004	U.S. Meat Animal Research	1	Clone	Ham		2 200337401	E	7/12/2004
200432005	U.S. Meat Animal Research	1	Clone	Ham		2 200337401	E	7/12/2004
200432006	U.S. Meat Animal Research	1	Clone	Ham		2 200337401	E	7/12/2004
200432007	U.S. Meat Animal Research	1	Clone	Ham		2 200337401	E	7/12/2004
200432008	U.S. Meat Animal Research	1	Clone	Ham		2 200337401	E	7/12/2004
200432101	U.S. Meat Animal Research	2	Clone	Ham		3 200342004	A	7/12/2004
200432102	U.S. Meat Animal Research	2	Clone	Ham		3 200342004	A	7/12/2004
200432103	U.S. Meat Animal Research	2	Clone	Ham		3 200342004	A	7/12/2004
200432104	U.S. Meat Animal Research	1	Clone	Ham		3 200342004	A	7/12/2004
200432105	U.S. Meat Animal Research	1	Clone	Ham		3 200342004	A	7/12/2004
200432106	U.S. Meat Animal Research	1	Clone	Ham		3 200342004	A	7/12/2004
200432108	U.S. Meat Animal Research	1	Clone	Ham		3 200342004	A	7/12/2004
200432109	U.S. Meat Animal Research	1	Clone	Ham		3 200342004	A	7/12/2004
200432301	U.S. Meat Animal Research	2	Clone	Ham		3 200336707	G	7/12/2004
200432302	U.S. Meat Animal Research	2	Clone	Ham		3 200336707	G	7/12/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200432303	U.S. Meat Animal Research	2	Clone	Ham		3 200336707	G	7/12/2004
200432304	U.S. Meat Animal Research	2	Clone	Ham		3 200336707	G	7/12/2004
200432305	U.S. Meat Animal Research	2	Clone	Ham		3 200336707	G	7/12/2004
200432306	U.S. Meat Animal Research	2	Clone	Ham		3 200336707	G	7/12/2004
200432307	U.S. Meat Animal Research	1	Clone	Ham		3 200336707	G	7/12/2004
200432308	U.S. Meat Animal Research	1	Clone	Ham		3 200336707	G	7/12/2004
200432309	U.S. Meat Animal Research	1	Clone	Ham		3 200336707	G	7/12/2004
200432310	U.S. Meat Animal Research	1	Clone	Ham		3 200336707	G	7/12/2004
200432311	U.S. Meat Animal Research	1	Clone	Ham		3 200336707	G	7/12/2004
200432312	U.S. Meat Animal Research	1	Clone	Ham		3 200336707	G	7/12/2004
200432401	U.S. Meat Animal Research	2	Clone	Ham		2 200344002	N	7/12/2004
200432402	U.S. Meat Animal Research	2	Clone	Ham		2 200344002	N	7/12/2004
200432403	U.S. Meat Animal Research	2	Clone	Ham		2 200344002	N	7/12/2004
200432404	U.S. Meat Animal Research	2	Clone	Ham		2 200344002	N	7/12/2004
200432405	U.S. Meat Animal Research	1	Clone	Ham		2 200344002	N	7/12/2004
200432406	U.S. Meat Animal Research	1	Clone	Ham		2 200344002	N	7/12/2004
200432407	U.S. Meat Animal Research	1	Clone	Ham		2 200344002	N	7/12/2004
200432501	U.S. Meat Animal Research	2	Clone	Ham		3 200345003	J	7/13/2004
200432502	U.S. Meat Animal Research	2	Clone	Ham		3 200345003	J	7/13/2004
200432503	U.S. Meat Animal Research	2	Clone	Ham		3 200345003	J	7/13/2004
200432504	U.S. Meat Animal Research	2	Clone	Ham		3 200345003	J	7/13/2004
200432506	U.S. Meat Animal Research	2	Clone	Ham		3 200345003	J	7/13/2004
200432507	U.S. Meat Animal Research	1	Clone	Ham		3 200345003	J	7/13/2004
200432508	U.S. Meat Animal Research	1	Clone	Ham		3 200345003	J	7/13/2004
200432601	U.S. Meat Animal Research	2	Control	Duroc	18128	200337403	E	7/13/2004
200432602	U.S. Meat Animal Research	2	Control	Duroc	18128	200337403	E	7/13/2004
200432603	U.S. Meat Animal Research	2	Control	Duroc	18128	200337403	E	7/13/2004
200432604	U.S. Meat Animal Research	2	Control	Duroc	18128	200337403	E	7/13/2004
200432605	U.S. Meat Animal Research	2	Control	Duroc	18128	200337403	E	7/13/2004
200432607	U.S. Meat Animal Research	1	Control	Duroc	18128	200337403	E	7/13/2004
200432608	U.S. Meat Animal Research	1	Control	Duroc	18128	200337403	E	7/13/2004
200432609	U.S. Meat Animal Research	1	Control	Duroc	18128	200337403	E	7/13/2004
200432610	U.S. Meat Animal Research	1	Control	Duroc	18128	200337403	E	7/13/2004
200432701	U.S. Meat Animal Research	2	Clone	Ham		3 200339303	D	7/13/2004
200432702	U.S. Meat Animal Research	2	Clone	Ham		3 200339303	D	7/13/2004
200432703	U.S. Meat Animal Research	1	Clone	Ham		3 200339303	D	7/13/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200432704	U.S. Meat Animal Research	1	Clone	Ham		3 200339303	D	7/13/2004
200432705	U.S. Meat Animal Research	1	Clone	Ham		3 200339303	D	7/13/2004
200432706	U.S. Meat Animal Research	1	Clone	Ham		3 200339303	D	7/13/2004
200432901	U.S. Meat Animal Research	2	Control	Duroc	25515	200346104	D	7/13/2004
200432902	U.S. Meat Animal Research	2	Control	Duroc	25515	200346104	D	7/13/2004
200432903	U.S. Meat Animal Research	2	Control	Duroc	25515	200346104	D	7/13/2004
200432904	U.S. Meat Animal Research	2	Control	Duroc	25515	200346104	D	7/13/2004
200432905	U.S. Meat Animal Research	2	Control	Duroc	25515	200346104	D	7/13/2004
200432906	U.S. Meat Animal Research	2	Control	Duroc	25515	200346104	D	7/13/2004
200432907	U.S. Meat Animal Research	1	Control	Duroc	25515	200346104	D	7/13/2004
200432908	U.S. Meat Animal Research	1	Control	Duroc	25515	200346104	D	7/13/2004
200432909	U.S. Meat Animal Research	1	Control	Duroc	25515	200346104	D	7/13/2004
200432910	U.S. Meat Animal Research	1	Control	Duroc	25515	200346104	D	7/13/2004
200432911	U.S. Meat Animal Research	1	Control	Duroc	25515	200346104	D	7/13/2004
200432912	U.S. Meat Animal Research	1	Control	Duroc	25515	200346104	D	7/13/2004
200432913	U.S. Meat Animal Research	1	Control	Duroc	25515	200346104	D	7/13/2004
200433001	U.S. Meat Animal Research	2	Clone	Ham		3 200341501	H	7/13/2004
200433002	U.S. Meat Animal Research	2	Clone	Ham		3 200341501	H	7/13/2004
200433003	U.S. Meat Animal Research	2	Clone	Ham		3 200341501	H	7/13/2004
200433004	U.S. Meat Animal Research	1	Clone	Ham		3 200341501	H	7/13/2004
200433005	U.S. Meat Animal Research	1	Clone	Ham		3 200341501	H	7/13/2004
200433006	U.S. Meat Animal Research	1	Clone	Ham		3 200341501	H	7/13/2004
200433007	U.S. Meat Animal Research	1	Clone	Ham		3 200341501	H	7/13/2004
200433008	U.S. Meat Animal Research	1	Clone	Ham		3 200341501	H	7/13/2004
200433010	U.S. Meat Animal Research	1	Clone	Ham		3 200341501	H	7/13/2004
200433011	U.S. Meat Animal Research	1	Clone	Ham		3 200341501	H	7/13/2004
200433201	U.S. Meat Animal Research	2	Clone	Ham		5 200342902	C	7/13/2004
200433202	U.S. Meat Animal Research	2	Clone	Ham		5 200342902	C	7/13/2004
200433203	U.S. Meat Animal Research	2	Clone	Ham		5 200342902	C	7/13/2004
200433204	U.S. Meat Animal Research	2	Clone	Ham		5 200342902	C	7/13/2004
200433205	U.S. Meat Animal Research	1	Clone	Ham		5 200342902	C	7/13/2004
200433206	U.S. Meat Animal Research	1	Clone	Ham		5 200342902	C	7/13/2004
200433207	U.S. Meat Animal Research	1	Clone	Ham		5 200342902	C	7/13/2004
200433208	U.S. Meat Animal Research	1	Clone	Ham		5 200342902	C	7/13/2004
200433301	U.S. Meat Animal Research	2	Control	Ham	498	200340902	F	7/13/2004
200433302	U.S. Meat Animal Research	2	Control	Ham	498	200340902	F	7/13/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200433303	U.S. Meat Animal Research	2	Control	Ham	498	200340902	F	7/13/2004
200433304	U.S. Meat Animal Research	2	Control	Ham	498	200340902	F	7/13/2004
200433305	U.S. Meat Animal Research	2	Control	Ham	498	200340902	F	7/13/2004
200433306	U.S. Meat Animal Research	1	Control	Ham	498	200340902	F	7/13/2004
200433307	U.S. Meat Animal Research	1	Control	Ham	498	200340902	F	7/13/2004
200433401	U.S. Meat Animal Research	2	Control	Ham	498	200342601	E	7/13/2004
200433402	U.S. Meat Animal Research	2	Control	Ham	498	200342601	E	7/13/2004
200433403	U.S. Meat Animal Research	2	Control	Ham	498	200342601	E	7/13/2004
200433404	U.S. Meat Animal Research	2	Control	Ham	498	200342601	E	7/13/2004
200433405	U.S. Meat Animal Research	2	Control	Ham	498	200342601	E	7/13/2004
200433406	U.S. Meat Animal Research	2	Control	Ham	498	200342601	E	7/13/2004
200433407	U.S. Meat Animal Research	2	Control	Ham	498	200342601	E	7/13/2004
200433408	U.S. Meat Animal Research	1	Control	Ham	498	200342601	E	7/13/2004
200433409	U.S. Meat Animal Research	1	Control	Ham	498	200342601	E	7/13/2004
200433501	U.S. Meat Animal Research	2	Control	Ham	498	200345803	D	7/13/2004
200433502	U.S. Meat Animal Research	2	Control	Ham	498	200345803	D	7/13/2004
200433503	U.S. Meat Animal Research	2	Control	Ham	498	200345803	D	7/13/2004
200433504	U.S. Meat Animal Research	2	Control	Ham	498	200345803	D	7/13/2004
200433505	U.S. Meat Animal Research	2	Control	Ham	498	200345803	D	7/13/2004
200433506	U.S. Meat Animal Research	2	Control	Ham	498	200345803	D	7/13/2004
200433507	U.S. Meat Animal Research	2	Control	Ham	498	200345803	D	7/13/2004
200433509	U.S. Meat Animal Research	1	Control	Ham	498	200345803	D	7/13/2004
200433510	U.S. Meat Animal Research	1	Control	Ham	498	200345803	D	7/13/2004
200433511	U.S. Meat Animal Research	1	Control	Ham	498	200345803	D	7/13/2004
200433512	U.S. Meat Animal Research	1	Control	Ham	498	200345803	D	7/13/2004
200433601	U.S. Meat Animal Research	2	Clone	Ham	3	200341201	M	7/13/2004
200433602	U.S. Meat Animal Research	2	Clone	Ham	3	200341201	M	7/13/2004
200433603	U.S. Meat Animal Research	2	Clone	Ham	3	200341201	M	7/13/2004
200433604	U.S. Meat Animal Research	2	Clone	Ham	3	200341201	M	7/13/2004
200433605	U.S. Meat Animal Research	1	Clone	Ham	3	200341201	M	7/13/2004
200433701	U.S. Meat Animal Research	2	Clone	Duroc	7	200346101	D	7/14/2004
200433702	U.S. Meat Animal Research	2	Clone	Duroc	7	200346101	D	7/14/2004
200433703	U.S. Meat Animal Research	2	Clone	Duroc	7	200346101	D	7/14/2004
200433704	U.S. Meat Animal Research	2	Clone	Duroc	7	200346101	D	7/14/2004
200433705	U.S. Meat Animal Research	2	Clone	Duroc	7	200346101	D	7/14/2004
200433706	U.S. Meat Animal Research	2	Clone	Duroc	7	200346101	D	7/14/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200433707	U.S. Meat Animal Research	2	Clone	Duroc		7 200346101 D		7/14/2004
200433708	U.S. Meat Animal Research	2	Clone	Duroc		7 200346101 D		7/14/2004
200433709	U.S. Meat Animal Research	2	Clone	Duroc		7 200346101 D		7/14/2004
200433710	U.S. Meat Animal Research	1	Clone	Duroc		7 200346101 D		7/14/2004
200433711	U.S. Meat Animal Research	1	Clone	Duroc		7 200346101 D		7/14/2004
200433712	U.S. Meat Animal Research	1	Clone	Duroc		7 200346101 D		7/14/2004
200433801	U.S. Meat Animal Research	2	Clone	Duroc		7 200336702 G		7/14/2004
200433802	U.S. Meat Animal Research	2	Clone	Duroc		7 200336702 G		7/14/2004
200433803	U.S. Meat Animal Research	2	Clone	Duroc		7 200336702 G		7/14/2004
200433804	U.S. Meat Animal Research	2	Clone	Duroc		7 200336702 G		7/14/2004
200433806	U.S. Meat Animal Research	1	Clone	Duroc		7 200336702 G		7/14/2004
200433807	U.S. Meat Animal Research	1	Clone	Duroc		7 200336702 G		7/14/2004
200433808	U.S. Meat Animal Research	1	Clone	Duroc		7 200336702 G		7/14/2004
200433809	U.S. Meat Animal Research	1	Clone	Duroc		7 200336702 G		7/14/2004
200434001	U.S. Meat Animal Research	2	Control	Duroc	18128	200345603 M		7/15/2004
200434002	U.S. Meat Animal Research	2	Control	Duroc	18128	200345603 M		7/15/2004
200434003	U.S. Meat Animal Research	2	Control	Duroc	18128	200345603 M		7/15/2004
200434004	U.S. Meat Animal Research	2	Control	Duroc	18128	200345603 M		7/15/2004
200434005	U.S. Meat Animal Research	2	Control	Duroc	18128	200345603 M		7/15/2004
200434006	U.S. Meat Animal Research	1	Control	Duroc	18128	200345603 M		7/15/2004
200434007	U.S. Meat Animal Research	1	Control	Duroc	18128	200345603 M		7/15/2004
200434008	U.S. Meat Animal Research	1	Control	Duroc	18128	200345603 M		7/15/2004
200434009	U.S. Meat Animal Research	1	Control	Duroc	18128	200345603 M		7/15/2004
200434101	U.S. Meat Animal Research	2	Clone	Ham		3 200337003 E		7/15/2004
200434102	U.S. Meat Animal Research	2	Clone	Ham		3 200337003 E		7/15/2004
200434103	U.S. Meat Animal Research	2	Clone	Ham		3 200337003 E		7/15/2004
200434104	U.S. Meat Animal Research	2	Clone	Ham		3 200337003 E		7/15/2004
200434105	U.S. Meat Animal Research	2	Clone	Ham		3 200337003 E		7/15/2004
200434106	U.S. Meat Animal Research	1	Clone	Ham		3 200337003 E		7/15/2004
200434107	U.S. Meat Animal Research	1	Clone	Ham		3 200337003 E		7/15/2004
200434108	U.S. Meat Animal Research	1	Clone	Ham		3 200337003 E		7/15/2004
200434109	U.S. Meat Animal Research	1	Clone	Ham		3 200337003 E		7/15/2004
200434110	U.S. Meat Animal Research	1	Clone	Ham		3 200337003 E		7/15/2004
200434111	U.S. Meat Animal Research	1	Clone	Ham		3 200337003 E		7/15/2004
200434112	U.S. Meat Animal Research	1	Clone	Ham		3 200337003 E		7/15/2004
200434113	U.S. Meat Animal Research	1	Clone	Ham		3 200337003 E		7/15/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200434114	U.S. Meat Animal Research	1	Clone	Ham		3 200337003	E	7/15/2004
200434115	U.S. Meat Animal Research	1	Clone	Ham		3 200337003	E	7/15/2004
200434201	U.S. Meat Animal Research	2	Control	Duroc	18128	200346102	D	7/15/2004
200434202	U.S. Meat Animal Research	1	Control	Duroc	18128	200346102	D	7/15/2004
200434203	U.S. Meat Animal Research	1	Control	Duroc	18128	200346102	D	7/15/2004
200434204	U.S. Meat Animal Research	1	Control	Duroc	18128	200346102	D	7/15/2004
200434205	U.S. Meat Animal Research	1	Control	Duroc	18128	200346102	D	7/15/2004
200434206	U.S. Meat Animal Research	1	Control	Duroc	18128	200346102	D	7/15/2004
200434207	U.S. Meat Animal Research	1	Control	Duroc	18128	200346102	D	7/15/2004
200434208	U.S. Meat Animal Research	1	Control	Duroc	18128	200346102	D	7/15/2004
200434209	U.S. Meat Animal Research	1	Control	Duroc	18128	200346102	D	7/15/2004
200434210	U.S. Meat Animal Research	1	Control	Duroc	18128	200346102	D	7/15/2004
200434301	U.S. Meat Animal Research	2	Control	Duroc	25515	200340204	M	7/15/2004
200434302	U.S. Meat Animal Research	2	Control	Duroc	25515	200340204	M	7/15/2004
200434303	U.S. Meat Animal Research	2	Control	Duroc	25515	200340204	M	7/15/2004
200434304	U.S. Meat Animal Research	1	Control	Duroc	25515	200340204	M	7/15/2004
200434305	U.S. Meat Animal Research	1	Control	Duroc	25515	200340204	M	7/15/2004
200434306	U.S. Meat Animal Research	1	Control	Duroc	25515	200340204	M	7/15/2004
200434307	U.S. Meat Animal Research	1	Control	Duroc	25515	200340204	M	7/15/2004
200434308	U.S. Meat Animal Research	1	Control	Duroc	25515	200340204	M	7/15/2004
200434309	U.S. Meat Animal Research	1	Control	Duroc	25515	200340204	M	7/15/2004
200434310	U.S. Meat Animal Research	1	Control	Duroc	25515	200340204	M	7/15/2004
200434401	U.S. Meat Animal Research	2	Clone	Ham		5 200345302	H	7/15/2004
200434402	U.S. Meat Animal Research	2	Clone	Ham		5 200345302	H	7/15/2004
200434403	U.S. Meat Animal Research	1	Clone	Ham		5 200345302	H	7/15/2004
200434501	U.S. Meat Animal Research	2	Clone	Ham		3 200344402	B	7/16/2004
200434502	U.S. Meat Animal Research	2	Clone	Ham		3 200344402	B	7/16/2004
200434503	U.S. Meat Animal Research	2	Clone	Ham		3 200344402	B	7/16/2004
200434504	U.S. Meat Animal Research	2	Clone	Ham		3 200344402	B	7/16/2004
200434506	U.S. Meat Animal Research	1	Clone	Ham		3 200344402	B	7/16/2004
200434507	U.S. Meat Animal Research	1	Clone	Ham		3 200344402	B	7/16/2004
200434508	U.S. Meat Animal Research	1	Clone	Ham		3 200344402	B	7/16/2004
200434509	U.S. Meat Animal Research	1	Clone	Ham		3 200344402	B	7/16/2004
200434510	U.S. Meat Animal Research	1	Clone	Ham		3 200344402	B	7/16/2004
200434511	U.S. Meat Animal Research	1	Clone	Ham		3 200344402	B	7/16/2004
200434512	U.S. Meat Animal Research	1	Clone	Ham		3 200344402	B	7/16/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200434601	U.S. Meat Animal Research	2	Clone	Ham		2 200344102	H	7/16/2004
200434602	U.S. Meat Animal Research	2	Clone	Ham		2 200344102	H	7/16/2004
200434603	U.S. Meat Animal Research	2	Clone	Ham		2 200344102	H	7/16/2004
200434604	U.S. Meat Animal Research	2	Clone	Ham		2 200344102	H	7/16/2004
200434605	U.S. Meat Animal Research	2	Clone	Ham		2 200344102	H	7/16/2004
200434606	U.S. Meat Animal Research	2	Clone	Ham		2 200344102	H	7/16/2004
200434607	U.S. Meat Animal Research	1	Clone	Ham		2 200344102	H	7/16/2004
200434608	U.S. Meat Animal Research	1	Clone	Ham		2 200344102	H	7/16/2004
200434801	U.S. Meat Animal Research	2	Control	Duroc	25515	200339403	H	7/17/2004
200434802	U.S. Meat Animal Research	2	Control	Duroc	25515	200339403	H	7/17/2004
200434803	U.S. Meat Animal Research	2	Control	Duroc	25515	200339403	H	7/17/2004
200434804	U.S. Meat Animal Research	2	Control	Duroc	25515	200339403	H	7/17/2004
200434805	U.S. Meat Animal Research	2	Control	Duroc	25515	200339403	H	7/17/2004
200434806	U.S. Meat Animal Research	1	Control	Duroc	25515	200339403	H	7/17/2004
200434807	U.S. Meat Animal Research	1	Control	Duroc	25515	200339403	H	7/17/2004
200434808	U.S. Meat Animal Research	1	Control	Duroc	25515	200339403	H	7/17/2004
200434809	U.S. Meat Animal Research	1	Control	Duroc	25515	200339403	H	7/17/2004
200434901	U.S. Meat Animal Research	2	Control	Duroc	18128	200344403	B	7/17/2004
200434902	U.S. Meat Animal Research	2	Control	Duroc	18128	200344403	B	7/17/2004
200434903	U.S. Meat Animal Research	2	Control	Duroc	18128	200344403	B	7/17/2004
200434904	U.S. Meat Animal Research	2	Control	Duroc	18128	200344403	B	7/17/2004
200434905	U.S. Meat Animal Research	1	Control	Duroc	18128	200344403	B	7/17/2004
200435301	U.S. Meat Animal Research	2	Control	Ham	498	200338103	A	7/17/2004
200435302	U.S. Meat Animal Research	2	Control	Ham	498	200338103	A	7/17/2004
200435303	U.S. Meat Animal Research	2	Control	Ham	498	200338103	A	7/17/2004
200435304	U.S. Meat Animal Research	2	Control	Ham	498	200338103	A	7/17/2004
200435305	U.S. Meat Animal Research	1	Control	Ham	498	200338103	A	7/17/2004
200435306	U.S. Meat Animal Research	1	Control	Ham	498	200338103	A	7/17/2004
200435307	U.S. Meat Animal Research	1	Control	Ham	498	200338103	A	7/17/2004
200435308	U.S. Meat Animal Research	1	Control	Ham	498	200338103	A	7/17/2004
200435309	U.S. Meat Animal Research	1	Control	Ham	498	200338103	A	7/17/2004
200435310	U.S. Meat Animal Research	1	Control	Ham	498	200338103	A	7/17/2004
200435311	U.S. Meat Animal Research	1	Control	Ham	498	200338103	A	7/17/2004
200435401	U.S. Meat Animal Research	2	Control	Duroc	25515	200343605	F	7/18/2004
200435402	U.S. Meat Animal Research	2	Control	Duroc	25515	200343605	F	7/18/2004
200435403	U.S. Meat Animal Research	2	Control	Duroc	25515	200343605	F	7/18/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200435404	U.S. Meat Animal Research	2	Control	Duroc	25515	200343605	F	7/18/2004
200435405	U.S. Meat Animal Research	2	Control	Duroc	25515	200343605	F	7/18/2004
200435407	U.S. Meat Animal Research	1	Control	Duroc	25515	200343605	F	7/18/2004
200435408	U.S. Meat Animal Research	1	Control	Duroc	25515	200343605	F	7/18/2004
200435409	U.S. Meat Animal Research	1	Control	Duroc	25515	200343605	F	7/18/2004
200435410	U.S. Meat Animal Research	1	Control	Duroc	25515	200343605	F	7/18/2004
200435411	U.S. Meat Animal Research	1	Control	Duroc	25515	200343605	F	7/18/2004
200435412	U.S. Meat Animal Research	1	Control	Duroc	25515	200343605	F	7/18/2004
200435413	U.S. Meat Animal Research	1	Control	Duroc	25515	200343605	F	7/18/2004
200435801	U.S. Meat Animal Research	2	Clone	Duroc		7 200338102	A	7/19/2004
200435802	U.S. Meat Animal Research	1	Clone	Duroc		7 200338102	A	7/19/2004
200435803	U.S. Meat Animal Research	1	Clone	Duroc		7 200338102	A	7/19/2004
200436001	U.S. Meat Animal Research	2	Clone	Ham		5 200344201	K	7/19/2004
200436002	U.S. Meat Animal Research	2	Clone	Ham		5 200344201	K	7/19/2004
200436003	U.S. Meat Animal Research	2	Clone	Ham		5 200344201	K	7/19/2004
200436004	U.S. Meat Animal Research	2	Clone	Ham		5 200344201	K	7/19/2004
200436005	U.S. Meat Animal Research	2	Clone	Ham		5 200344201	K	7/19/2004
200436006	U.S. Meat Animal Research	2	Clone	Ham		5 200344201	K	7/19/2004
200436007	U.S. Meat Animal Research	2	Clone	Ham		5 200344201	K	7/19/2004
200436008	U.S. Meat Animal Research	2	Clone	Ham		5 200344201	K	7/19/2004
200436009	U.S. Meat Animal Research	1	Clone	Ham		5 200344201	K	7/19/2004
200436010	U.S. Meat Animal Research	1	Clone	Ham		5 200344201	K	7/19/2004
200436011	U.S. Meat Animal Research	1	Clone	Ham		5 200344201	K	7/19/2004
200436012	U.S. Meat Animal Research	1	Clone	Ham		5 200344201	K	7/19/2004
200436013	U.S. Meat Animal Research	1	Clone	Ham		5 200344201	K	7/19/2004
200436301	U.S. Meat Animal Research	2	Control	Duroc	25515	200340001	A	7/19/2004
200436302	U.S. Meat Animal Research	2	Control	Duroc	25515	200340001	A	7/19/2004
200436303	U.S. Meat Animal Research	2	Control	Duroc	25515	200340001	A	7/19/2004
200436304	U.S. Meat Animal Research	2	Control	Duroc	25515	200340001	A	7/19/2004
200436305	U.S. Meat Animal Research	2	Control	Duroc	25515	200340001	A	7/19/2004
200436306	U.S. Meat Animal Research	2	Control	Duroc	25515	200340001	A	7/19/2004
200436307	U.S. Meat Animal Research	2	Control	Duroc	25515	200340001	A	7/19/2004
200436309	U.S. Meat Animal Research	1	Control	Duroc	25515	200340001	A	7/19/2004
200436310	U.S. Meat Animal Research	1	Control	Duroc	25515	200340001	A	7/19/2004
200436311	U.S. Meat Animal Research	1	Control	Duroc	25515	200340001	A	7/19/2004
200436312	U.S. Meat Animal Research	1	Control	Duroc	25515	200340001	A	7/19/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200436401	U.S. Meat Animal Research	2	Clone	Ham		5 200339104	N	7/19/2004
200436402	U.S. Meat Animal Research	1	Clone	Ham		5 200339104	N	7/19/2004
200436403	U.S. Meat Animal Research	1	Clone	Ham		5 200339104	N	7/19/2004
200436404	U.S. Meat Animal Research	1	Clone	Ham		5 200339104	N	7/19/2004
200436405	U.S. Meat Animal Research	1	Clone	Ham		5 200339104	N	7/19/2004
200436601	U.S. Meat Animal Research	2	Clone	Duroc		7 200340206	M	7/20/2004
200436602	U.S. Meat Animal Research	2	Clone	Duroc		7 200340206	M	7/20/2004
200436603	U.S. Meat Animal Research	2	Clone	Duroc		7 200340206	M	7/20/2004
200436604	U.S. Meat Animal Research	2	Clone	Duroc		7 200340206	M	7/20/2004
200436605	U.S. Meat Animal Research	1	Clone	Duroc		7 200340206	M	7/20/2004
200436606	U.S. Meat Animal Research	1	Clone	Duroc		7 200340206	M	7/20/2004
200436607	U.S. Meat Animal Research	1	Clone	Duroc		7 200340206	M	7/20/2004
200436608	U.S. Meat Animal Research	1	Clone	Duroc		7 200340206	M	7/20/2004
200436609	U.S. Meat Animal Research	1	Clone	Duroc		7 200340206	M	7/20/2004
200436610	U.S. Meat Animal Research	1	Clone	Duroc		7 200340206	M	7/20/2004
200436611	U.S. Meat Animal Research	1	Clone	Duroc		7 200340206	M	7/20/2004
200436612	U.S. Meat Animal Research	1	Clone	Duroc		7 200340206	M	7/20/2004
200436901	U.S. Meat Animal Research	2	Clone	Ham		3 200337501	K	7/20/2004
200436902	U.S. Meat Animal Research	2	Clone	Ham		3 200337501	K	7/20/2004
200436903	U.S. Meat Animal Research	2	Clone	Ham		3 200337501	K	7/20/2004
200436904	U.S. Meat Animal Research	2	Clone	Ham		3 200337501	K	7/20/2004
200436905	U.S. Meat Animal Research	2	Clone	Ham		3 200337501	K	7/20/2004
200436906	U.S. Meat Animal Research	2	Clone	Ham		3 200337501	K	7/20/2004
200436907	U.S. Meat Animal Research	2	Clone	Ham		3 200337501	K	7/20/2004
200436908	U.S. Meat Animal Research	2	Clone	Ham		3 200337501	K	7/20/2004
200436909	U.S. Meat Animal Research	1	Clone	Ham		3 200337501	K	7/20/2004
200436910	U.S. Meat Animal Research	1	Clone	Ham		3 200337501	K	7/20/2004
200436911	U.S. Meat Animal Research	1	Clone	Ham		3 200337501	K	7/20/2004
200436912	U.S. Meat Animal Research	1	Clone	Ham		3 200337501	K	7/20/2004
200436913	U.S. Meat Animal Research	1	Clone	Ham		3 200337501	K	7/20/2004
200437301	U.S. Meat Animal Research	2	Control	Duroc	25515	200340702	J	7/22/2004
200437302	U.S. Meat Animal Research	2	Control	Duroc	25515	200340702	J	7/22/2004
200437303	U.S. Meat Animal Research	2	Control	Duroc	25515	200340702	J	7/22/2004
200437304	U.S. Meat Animal Research	1	Control	Duroc	25515	200340702	J	7/22/2004
200437305	U.S. Meat Animal Research	1	Control	Duroc	25515	200340702	J	7/22/2004
200437306	U.S. Meat Animal Research	1	Control	Duroc	25515	200340702	J	7/22/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200437307	U.S. Meat Animal Research	1	Control	Duroc	25515	200340702	J	7/22/2004
200437308	U.S. Meat Animal Research	1	Control	Duroc	25515	200340702	J	7/22/2004
200437309	U.S. Meat Animal Research	1	Control	Duroc	25515	200340702	J	7/22/2004
200437401	U.S. Meat Animal Research	2	Clone	Duroc	7	200338203	C	7/22/2004
200437402	U.S. Meat Animal Research	2	Clone	Duroc	7	200338203	C	7/22/2004
200437403	U.S. Meat Animal Research	1	Clone	Duroc	7	200338203	C	7/22/2004
200437404	U.S. Meat Animal Research	1	Clone	Duroc	7	200338203	C	7/22/2004
200437405	U.S. Meat Animal Research	1	Clone	Duroc	7	200338203	C	7/22/2004
200437406	U.S. Meat Animal Research	1	Clone	Duroc	7	200338203	C	7/22/2004
200437407	U.S. Meat Animal Research	1	Clone	Duroc	7	200338203	C	7/22/2004
200437408	U.S. Meat Animal Research	1	Clone	Duroc	7	200338203	C	7/22/2004
200437409	U.S. Meat Animal Research	1	Clone	Duroc	7	200338203	C	7/22/2004
200437410	U.S. Meat Animal Research	1	Clone	Duroc	7	200338203	C	7/22/2004
200437411	U.S. Meat Animal Research	1	Clone	Duroc	7	200338203	C	7/22/2004
200437501	U.S. Meat Animal Research	2	Clone	Duroc	7	200342201	K	7/22/2004
200437502	U.S. Meat Animal Research	2	Clone	Duroc	7	200342201	K	7/22/2004
200437504	U.S. Meat Animal Research	1	Clone	Duroc	7	200342201	K	7/22/2004
200437505	U.S. Meat Animal Research	1	Clone	Duroc	7	200342201	K	7/22/2004
200437506	U.S. Meat Animal Research	1	Clone	Duroc	7	200342201	K	7/22/2004
200437507	U.S. Meat Animal Research	1	Clone	Duroc	7	200342201	K	7/22/2004
200437509	U.S. Meat Animal Research	1	Clone	Duroc	7	200342201	K	7/22/2004
200437510	U.S. Meat Animal Research	1	Clone	Duroc	7	200342201	K	7/22/2004
200437801	U.S. Meat Animal Research	2	Clone	Duroc	7	200339102	N	7/22/2004
200437802	U.S. Meat Animal Research	1	Clone	Duroc	7	200339102	N	7/22/2004
200437803	U.S. Meat Animal Research	1	Clone	Duroc	7	200339102	N	7/22/2004
200437804	U.S. Meat Animal Research	1	Clone	Duroc	7	200339102	N	7/22/2004
200438101	U.S. Meat Animal Research	2	Clone	Duroc	7	200339704	J	7/23/2004
200438102	U.S. Meat Animal Research	2	Clone	Duroc	7	200339704	J	7/23/2004
200438103	U.S. Meat Animal Research	2	Clone	Duroc	7	200339704	J	7/23/2004
200438104	U.S. Meat Animal Research	2	Clone	Duroc	7	200339704	J	7/23/2004
200438105	U.S. Meat Animal Research	2	Clone	Duroc	7	200339704	J	7/23/2004
200438106	U.S. Meat Animal Research	1	Clone	Duroc	7	200339704	J	7/23/2004
200438107	U.S. Meat Animal Research	1	Clone	Duroc	7	200339704	J	7/23/2004
200438401	U.S. Meat Animal Research	2	Control	Duroc	18128	200339603	N	7/23/2004
200438402	U.S. Meat Animal Research	2	Control	Duroc	18128	200339603	N	7/23/2004
200438403	U.S. Meat Animal Research	2	Control	Duroc	18128	200339603	N	7/23/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200438404	U.S. Meat Animal Research	1	Control	Duroc	18128	200339603	N	7/23/2004
200438405	U.S. Meat Animal Research	1	Control	Duroc	18128	200339603	N	7/23/2004
200438406	U.S. Meat Animal Research	1	Control	Duroc	18128	200339603	N	7/23/2004
200438407	U.S. Meat Animal Research	1	Control	Duroc	18128	200339603	N	7/23/2004
200438408	U.S. Meat Animal Research	1	Control	Duroc	18128	200339603	N	7/23/2004
200438409	U.S. Meat Animal Research	1	Control	Duroc	18128	200339603	N	7/23/2004
200438410	U.S. Meat Animal Research	1	Control	Duroc	18128	200339603	N	7/23/2004
200438411	U.S. Meat Animal Research	1	Control	Duroc	18128	200339603	N	7/23/2004
200438412	U.S. Meat Animal Research	1	Control	Duroc	18128	200339603	N	7/23/2004
200438413	U.S. Meat Animal Research	1	Control	Duroc	18128	200339603	N	7/23/2004
200438414	U.S. Meat Animal Research	1	Control	Duroc	18128	200339603	N	7/23/2004
200438501	U.S. Meat Animal Research	2	Control	Duroc	18128	200336502	A	7/23/2004
200438502	U.S. Meat Animal Research	2	Control	Duroc	18128	200336502	A	7/23/2004
200438503	U.S. Meat Animal Research	2	Control	Duroc	18128	200336502	A	7/23/2004
200438504	U.S. Meat Animal Research	2	Control	Duroc	18128	200336502	A	7/23/2004
200438505	U.S. Meat Animal Research	2	Control	Duroc	18128	200336502	A	7/23/2004
200438506	U.S. Meat Animal Research	2	Control	Duroc	18128	200336502	A	7/23/2004
200438507	U.S. Meat Animal Research	2	Control	Duroc	18128	200336502	A	7/23/2004
200438508	U.S. Meat Animal Research	2	Control	Duroc	18128	200336502	A	7/23/2004
200438509	U.S. Meat Animal Research	2	Control	Duroc	18128	200336502	A	7/23/2004
200438510	U.S. Meat Animal Research	2	Control	Duroc	18128	200336502	A	7/23/2004
200438511	U.S. Meat Animal Research	1	Control	Duroc	18128	200336502	A	7/23/2004
200438512	U.S. Meat Animal Research	1	Control	Duroc	18128	200336502	A	7/23/2004
200438513	U.S. Meat Animal Research	1	Control	Duroc	18128	200336502	A	7/23/2004
200438514	U.S. Meat Animal Research	1	Control	Duroc	18128	200336502	A	7/23/2004
200438601	U.S. Meat Animal Research	2	Clone	Duroc	7	200340906	F	7/23/2004
200438602	U.S. Meat Animal Research	2	Clone	Duroc	7	200340906	F	7/23/2004
200438603	U.S. Meat Animal Research	2	Clone	Duroc	7	200340906	F	7/23/2004
200438604	U.S. Meat Animal Research	2	Clone	Duroc	7	200340906	F	7/23/2004
200438605	U.S. Meat Animal Research	2	Clone	Duroc	7	200340906	F	7/23/2004
200438606	U.S. Meat Animal Research	2	Clone	Duroc	7	200340906	F	7/23/2004
200438607	U.S. Meat Animal Research	1	Clone	Duroc	7	200340906	F	7/23/2004
200438608	U.S. Meat Animal Research	1	Clone	Duroc	7	200340906	F	7/23/2004
200438609	U.S. Meat Animal Research	1	Clone	Duroc	7	200340906	F	7/23/2004
200438610	U.S. Meat Animal Research	1	Clone	Duroc	7	200340906	F	7/23/2004
200438611	U.S. Meat Animal Research	1	Clone	Duroc	7	200340906	F	7/23/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200438701	U.S. Meat Animal Research	2	Control	Duroc	25515	200339105	N	7/23/2004
200438702	U.S. Meat Animal Research	2	Control	Duroc	25515	200339105	N	7/23/2004
200438703	U.S. Meat Animal Research	2	Control	Duroc	25515	200339105	N	7/23/2004
200438704	U.S. Meat Animal Research	2	Control	Duroc	25515	200339105	N	7/23/2004
200438705	U.S. Meat Animal Research	2	Control	Duroc	25515	200339105	N	7/23/2004
200438706	U.S. Meat Animal Research	1	Control	Duroc	25515	200339105	N	7/23/2004
200438707	U.S. Meat Animal Research	1	Control	Duroc	25515	200339105	N	7/23/2004
200438709	U.S. Meat Animal Research	1	Control	Duroc	25515	200339105	N	7/23/2004
200439201	U.S. Meat Animal Research	2	Control	Duroc	18128	200336204	G	7/24/2004
200439202	U.S. Meat Animal Research	2	Control	Duroc	18128	200336204	G	7/24/2004
200439203	U.S. Meat Animal Research	2	Control	Duroc	18128	200336204	G	7/24/2004
200439204	U.S. Meat Animal Research	2	Control	Duroc	18128	200336204	G	7/24/2004
200439205	U.S. Meat Animal Research	2	Control	Duroc	18128	200336204	G	7/24/2004
200439206	U.S. Meat Animal Research	1	Control	Duroc	18128	200336204	G	7/24/2004
200439207	U.S. Meat Animal Research	1	Control	Duroc	18128	200336204	G	7/24/2004
200439208	U.S. Meat Animal Research	1	Control	Duroc	18128	200336204	G	7/24/2004
200439209	U.S. Meat Animal Research	1	Control	Duroc	18128	200336204	G	7/24/2004
200439210	U.S. Meat Animal Research	1	Control	Duroc	18128	200336204	G	7/24/2004
200439211	U.S. Meat Animal Research	1	Control	Duroc	18128	200336204	G	7/24/2004
200439212	U.S. Meat Animal Research	1	Control	Duroc	18128	200336204	G	7/24/2004
200439215	U.S. Meat Animal Research	1	Control	Duroc	18128	200336204	G	7/24/2004
200439401	U.S. Meat Animal Research	2	Control	Duroc	18128	200343802	C	7/24/2004
200439403	U.S. Meat Animal Research	1	Control	Duroc	18128	200343802	C	7/24/2004
200439404	U.S. Meat Animal Research	1	Control	Duroc	18128	200343802	C	7/24/2004
200439405	U.S. Meat Animal Research	1	Control	Duroc	18128	200343802	C	7/24/2004
200439406	U.S. Meat Animal Research	1	Control	Duroc	18128	200343802	C	7/24/2004
200439407	U.S. Meat Animal Research	1	Control	Duroc	18128	200343802	C	7/24/2004
200439408	U.S. Meat Animal Research	1	Control	Duroc	18128	200343802	C	7/24/2004
200439409	U.S. Meat Animal Research	1	Control	Duroc	18128	200343802	C	7/24/2004
200439501	U.S. Meat Animal Research	2	Clone	Duroc	7	200342502	E	7/24/2004
200439502	U.S. Meat Animal Research	2	Clone	Duroc	7	200342502	E	7/24/2004
200439503	U.S. Meat Animal Research	2	Clone	Duroc	7	200342502	E	7/24/2004
200439504	U.S. Meat Animal Research	2	Clone	Duroc	7	200342502	E	7/24/2004
200439505	U.S. Meat Animal Research	1	Clone	Duroc	7	200342502	E	7/24/2004
200439901	U.S. Meat Animal Research	1	Control	Duroc	25515	200338501	G	7/24/2004
200439902	U.S. Meat Animal Research	1	Control	Duroc	25515	200338501	G	7/24/2004

Animal ID	Owner	Sex	Treatment	Sire Genetics	Sire	Dam	Dam Sire Group	Birth Date
200439903	U.S. Meat Animal Research	1	Control	Duroc	25515	200338501	G	7/24/2004
200439904	U.S. Meat Animal Research	1	Control	Duroc	25515	200338501	G	7/24/2004
200440601	U.S. Meat Animal Research	2	Control	Duroc	18128	200340901	F	7/25/2004
200440602	U.S. Meat Animal Research	2	Control	Duroc	18128	200340901	F	7/25/2004
200440603	U.S. Meat Animal Research	1	Control	Duroc	18128	200340901	F	7/25/2004
200440604	U.S. Meat Animal Research	1	Control	Duroc	18128	200340901	F	7/25/2004
200440605	U.S. Meat Animal Research	1	Control	Duroc	18128	200340901	F	7/25/2004
200440606	U.S. Meat Animal Research	1	Control	Duroc	18128	200340901	F	7/25/2004
200440607	U.S. Meat Animal Research	1	Control	Duroc	18128	200340901	F	7/25/2004
200440608	U.S. Meat Animal Research	1	Control	Duroc	18128	200340901	F	7/25/2004
200440609	U.S. Meat Animal Research	1	Control	Duroc	18128	200340901	F	7/25/2004
200440901	U.S. Meat Animal Research	2	Control	Duroc	18128	200341702	K	7/26/2004
200440902	U.S. Meat Animal Research	2	Control	Duroc	18128	200341702	K	7/26/2004
200440903	U.S. Meat Animal Research	2	Control	Duroc	18128	200341702	K	7/26/2004
200440904	U.S. Meat Animal Research	2	Control	Duroc	18128	200341702	K	7/26/2004
200440905	U.S. Meat Animal Research	2	Control	Duroc	18128	200341702	K	7/26/2004
200440906	U.S. Meat Animal Research	2	Control	Duroc	18128	200341702	K	7/26/2004
200440907	U.S. Meat Animal Research	1	Control	Duroc	18128	200341702	K	7/26/2004
200440908	U.S. Meat Animal Research	1	Control	Duroc	18128	200341702	K	7/26/2004
200440909	U.S. Meat Animal Research	1	Control	Duroc	18128	200341702	K	7/26/2004
200440910	U.S. Meat Animal Research	1	Control	Duroc	18128	200341702	K	7/26/2004
200440911	U.S. Meat Animal Research	1	Control	Duroc	18128	200341702	K	7/26/2004
200440912	U.S. Meat Animal Research	1	Control	Duroc	18128	200341702	K	7/26/2004
200440913	U.S. Meat Animal Research	1	Control	Duroc	18128	200341702	K	7/26/2004
200441201	U.S. Meat Animal Research	2	Control	Duroc	25515	200337303	B	7/27/2004
200441202	U.S. Meat Animal Research	2	Control	Duroc	25515	200337303	B	7/27/2004
200441203	U.S. Meat Animal Research	1	Control	Duroc	25515	200337303	B	7/27/2004
200441204	U.S. Meat Animal Research	1	Control	Duroc	25515	200337303	B	7/27/2004
200441205	U.S. Meat Animal Research	1	Control	Duroc	25515	200337303	B	7/27/2004
200441206	U.S. Meat Animal Research	1	Control	Duroc	25515	200337303	B	7/27/2004
200441207	U.S. Meat Animal Research	1	Control	Duroc	25515	200337303	B	7/27/2004
200441208	U.S. Meat Animal Research	1	Control	Duroc	25515	200337303	B	7/27/2004
200441209	U.S. Meat Animal Research	1	Control	Duroc	25515	200337303	B	7/27/2004
200441210	U.S. Meat Animal Research	1	Control	Duroc	25515	200337303	B	7/27/2004
200441211	U.S. Meat Animal Research	1	Control	Duroc	25515	200337303	B	7/27/2004

Experiment 2

Sire ID	Sire ID	Sire ID	Sire ID	Sire ID	Sire ID	Sire ID
Clone 002 (H498)	Clone 003 (H498)	Clone 005 (H498)	Clone 007 -5001	Control H498 Hamline	Control 18128 5001 Son	Control 25515 5001 Son
Offspring ID	Offspring ID	Offspring ID	Offspring ID	Offspring ID	Offspring ID	Offspring ID
200430701	200430901	200430601	200433701	200430501	200431801	200432901
200430702	200430902	200430602	200433702	200430502	200431802	200432902
200430703	200430903	200430603	200433703	200430503	200431803	200432903
200430704	200430904	200430604	200433704	200430504	200431804	200432904
200430705	200430905	200430605	200433705	200430505	200431805	200432905
200430706	200430906	200430606	200433706	200430506	200432601	200432906
200430707	200432101	200430607	200433707	200430507	200432602	200432907
200430708	200432102	200430608	200433708	200430508	200432603	200432908
200430709	200432103	200430609	200433709	200430509	200432604	200432909
200430710	200432104	200430610	200433710	200430510	200432605	200432910
200430711	200432105	200430611	200433711	200433301	200432606	200432911
200431001	200432106	200430801	200433712	200433302	200432607	200432912
200431002	200432107	200430802	200433801	200433303	200432608	200432913
200431003	200432108	200430803	200433802	200433304	200432609	200434301
200431004	200432109	200430804	200433803	200433305	200432610	200434302
200431005	200432301	200430805	200433804	200433306	200434001	200434303
200431006	200432302	200431401	200433805	200433307	200434002	200434304
200431007	200432303	200431402	200433806	200433401	200434003	200434305
200431008	200432304	200431403	200433807	200433402	200434004	200434306
200431009	200432305	200431404	200433808	200433403	200434005	200434307
200431010	200432306	200431405	200433809	200433404	200434006	200434308
200431011	200432307	200431406	200435801	200433405	200434007	200434309
200431012	200432308	200431407	200435802	200433406	200434008	200434310
200431013	200432309	200431408	200435803	200433407	200434009	200434801
200431501	200432310	200431409	200436601	200433408	200434201	200434802
200431502	200432311	200431410	200436602	200433409	200434202	200434803
200431503	200432312	200431411	200436603	200433501	200434203	200434804
200431504	200432501	200431701	200436604	200433502	200434204	200434805
200431505	200432502	200431702	200436605	200433503	200434205	200434806
200431506	200432503	200431703	200436606	200433504	200434206	200434807
200431601	200432504	200431704	200436607	200433505	200434207	200434808
200431602	200432505	200431705	200436608	200433506	200434208	200434809
200431603	200432506	200431706	200436609	200433507	200434209	200435401
200431604	200432507	200431707	200436610	200433508	200434210	200435402
200431605	200432508	200431708	200436611	200433509	200434901	200435403
200431606	200432701	200433201	200436612	200433510	200434902	200435404
200431607	200432702	200433202	200437401	200433511	200434903	200435405
200431608	200432703	200433203	200437402	200433512	200434904	200435406
200431609	200432704	200433204	200437403	200435301	200434905	200435407
200431610	200432705	200433205	200437404	200435302	200434906	200435408
200432001	200432706	200433206	200437405	200435303	200438401	200435409
200432002	200433001	200433207	200437406	200435304	200438402	200435410
200432003	200433002	200433208	200437407	200435305	200438403	200435411
200432004	200433003	200434401	200437408	200435306	200438404	200435412
200432005	200433004	200434402	200437409	200435307	200438405	200435413
200432006	200433005	200434403	200437410	200435308	200438406	200436301
200432007	200433006	200436001	200437411	200435309	200438407	200436302
200432008	200433007	200436002	200437501	200435310	200438408	200436303
200432401	200433008	200436003	200437502	200435311	200438409	200436304
200432402	200433009	200436004	200437503		200438410	200436305
200432403	200433010	200436005	200437504		200438411	200436306
200432404	200433011	200436006	200437505		200438412	200436307
200432405	200433601	200436007	200437506		200438413	200436308
200432406	200433602	200436008	200437507		200438414	200436309
200432407	200433603	200436009	200437508		200438501	200436310

Sire ID	Sire ID	Sire ID	Sire ID	Sire ID	Sire ID	Sire ID
Clone 002 (H498)	Clone 003 (H498)	Clone 005 (H498)	Clone 007 -5001	Control H498 Hamline	Control 18128 5001 Son	Control 25515 5001 Son
Offspring ID	Offspring ID	Offspring ID	Offspring ID	Offspring ID	Offspring ID	Offspring ID
200432408	200433604	200436010	200437509		200438502	200436311
200434601	200433605	200436011	200437510		200438503	200436312
200434602	200434101	200436012	200437801		200438504	200437301
200434603	200434102	200436013	200437802		200438505	200437302
200434604	200434103	200436014	200437803		200438506	200437303
200434605	200434104	200436401	200437804		200438507	200437304
200434606	200434105	200436402	200438101		200438508	200437305
200434607	200434106	200436403	200438102		200438509	200437306
200434608	200434107	200436404	200438103		200438510	200437307
	200434108	200436405	200438104		200438511	200437308
	200434109		200438105		200438512	200437309
	200434110		200438106		200438513	200438701
	200434111		200438107		200438514	200438702
	200434112		200438601		200439201	200438703
	200434113		200438602		200439202	200438704
	200434114		200438603		200439203	200438705
	200434115		200438604		200439204	200438706
	200434501		200438605		200439205	200438707
	200434502		200438606		200439206	200438708
	200434503		200438607		200439207	200438709
	200434504		200438608		200439208	200439901
	200434505		200438609		200439209	200439902
	200434506		200438610		200439210	200439903
	200434507		200438611		200439211	200439904
	200434508		200439501		200439212	200441201
	200434509		200439502		200439213	200441202
	200434510		200439503		200439401	200441203
	200434511		200439504		200439402	200441204
	200434512		200439505		200439403	200441205
	200436901				200439404	200441206
	200436902				200439405	200441207
	200436903				200439406	200441208
	200436904				200439407	200441209
	200436905				200439408	200441210
	200436906				200439409	200441211
	200436907				200440601	200441212
	200436908				200440602	
	200436909				200440603	
	200436910				200440604	
	200436911				200440605	
	200436912				200440606	
	200436913				200440607	
					200440608	
					200440609	
					200440901	
					200440902	
					200440903	
					200440904	
					200440905	
					200440906	
					200440907	
					200440908	
					200440909	
					200440910	
					200440911	
					200440912	
					200440913	

Date	Treatment	Genetics	Age	Boar ID	Gel WT (g)	Temp °F	Volume (ml)	Concentration (x10 ⁶)	Motility %	Total Concentration	Viable Sperm Concentration	# Doses (3x10 ⁹)	Morph	Glutination
2/13/2004	Clone	TX1	12	2	25.13		190.9		80					3
2/20/2004	Clone	TX1	12	2	12.5	97.7	134.7	416	80	56.04	44.83	14.94		2
3/1/2004	Clone	TX1	12	2	31.3	96.2	184	368	80	67.71	54.17	18.06	1	3
3/10/2004	Clone	TX1	12	2	28.6		183	315	90	57.65	51.88	17.29		2
3/15/2004	Clone	TX1	12	2	23.4	98.7	199.7	268	95	53.52	50.84	16.95	10	2
3/23/2004	Clone	TX1	12	2	39	96.8	267.6	280	95	74.93	71.18	23.73	10	2
4/2/2004	Clone	TX1	14	2	27.2	94.6	217.6	349	95	75.94	72.15	24.05	10	2
4/12/2004	Clone	TX1	14	2	35.6	98.9	289.4	239	95	69.17	65.71	21.90	10	3
4/19/2004	Clone	TX1	14	2	38.92	98.7	258.62	230	95	59.48	56.51	18.84	1	2
5/5/2004	Clone	TX1	15	2	18.04	98.2	164.8	480	95	79.10	75.15	25.05	1	2
5/11/2004	Clone	TX1	15	2		97.5	92.4	468	95	10.00	2.00	0.67	10.00	2
5/18/2004	Clone	TX1	15	2		96.4	88.72	493	95				1.00	2
5/24/2004	Clone	TX1	15	2		96.9	72.8	504	95				1.00	2
6/10/2004	Clone	TX1	16	2	19.98	97.5	206.42	333	95				10.00	2
3/2/2004	Clone	TX1	12	3	4.2	91	27.1	557	80	15.09	12.08	4.03	20	2
3/5/2004	Clone	TX1	12	3	37	95.3	86.4	400	90	34.56	31.10	10.37		2
3/10/2004	Clone	TX1	12	3	31.9	91	89.6	319	90	28.58	25.72	8.57		2
3/15/2004	Clone	TX1	12	3	6.71	94.8	81	227	90	18.39	16.55	5.52	10	2
3/17/2004	Clone	TX1	12	3	12.6	95.5	95.3	363	95	34.59	32.86	10.95	1	2
3/22/2004	Clone	TX1	12	3	21.4	93.3	86.7	297	95	25.75	24.46	8.15	10	2
4/2/2004	Clone	TX1	14	3	7.15	93.9	98.3	375	95	36.86	35.02	11.67	10	2
4/12/2004	Clone	TX1	14	3	9.5	96.9	137.6	397	95	54.63	51.90	17.30	10	2
4/19/2004	Clone	TX1	14	3	18.43	96.6	134.68	370	95	49.83	47.34	15.78	1	2
5/5/2004	Clone	TX1	15	3	7.47	94.7	81.4	468	95	38.10	36.19	12.06	10	2
5/11/2004	Clone	TX1	15	3		98.2	85.3	392	95	33.44	31.77	10.59	10	2
5/18/2004	Clone	TX1	15	3		94.6	70.5	462	95	32.57	30.94	10.31	5	2
5/24/2004	Clone	TX1	15	3		96.4	92.8	499	95	46.31	43.99	14.66	1	2
2/13/2004	Clone	TX1	10	5	30.4	85.4	56.2		60	0.00	0.00	0.00		2
2/14/2004	Clone	TX1	10	5	23.5		99.5		80	0.00	0.00	0.00		2
2/16/2004	Clone	TX1	10	5	14	92.4	116.5		75	0.00	0.00	0.00		2
2/20/2004	Clone	TX1	10	5	12.5	95.5	114	166	80	18.92	15.14	5.05		2
3/10/2004	Clone	TX1	10	5	20.8	88.7	121.5	368	95	44.71	42.48	14.16		2
3/15/2004	Clone	TX1	10	5	30.71	91.2	139.9	218	95	30.50	28.97	9.66	10	2
3/17/2004	Clone	TX1	10	5	11.3	91.7	120.3	176	90	21.17	19.06	6.35	10	2
3/23/2004	Clone	TX1	10	5	19.1	94.4	169.2	194	95	32.82	31.18	10.39	10	2
4/2/2004	Clone	TX1	10	5	16.8	95.7	208.8	236	90	49.28	44.35	14.78	5	2
4/12/2004	Clone	TX1	10	5	29.1	97.1	177.2	379	95	67.16	63.80	21.27	10	2
4/19/2004	Clone	TX1	11	5	22.49	98.4	169.33	253	95	42.84	40.70	13.57	10	3
5/5/2004	Clone	TX1	12	5		94.3	50.63	541	95	27.39	26.02	8.67	3	2
5/11/2004	Clone	TX1	12	5			71	400	95	28.40	26.98	8.99	5	2
5/18/2004	Clone	TX1	12	5		93.7	76.77	411	95	31.55	29.97	9.99	5	2
5/24/2004	Clone	TX1	12	5		95.9	85.5	534	95	45.66	43.37	14.46	1	2
2/14/2004	Clone	TX2	6.5	7			18.9		75	0.00	0.00	0.00		2
2/16/2004	Clone	TX2	6.5	7	4	85.6	56.7		80	0.00	0.00	0.00		2
2/20/2004	Clone	TX2	6.5	7	10	89.2	76.9	316	80	24.30	19.44	6.48		3
3/10/2004	Clone	TX2	6.5	7	9.6		44.2	516	85	22.81	19.39	6.46		2
3/15/2004	Clone	TX2	6.5	7	17.48	91.5	52.7	378	95	19.92	18.92	6.31	15	2
3/23/2004	Clone	TX2	6.5	7	14.9	92.3	71.3	242	95	17.25	16.39	5.46	10	2
3/29/2004	Clone	TX2	6.5	7	22.1	88.4	87.4	430	95	37.58	35.70	11.90	15	2
4/6/2004	Clone	TX2	7	7	14.9	90.1	60.1	301	95	18.09	17.19	5.73	10	2
4/13/2004	Clone	TX2	7.5	7	9.52	79.9	39.75	343	80	13.63	10.91	3.64	15	2
4/20/2004	Clone	TX2	7.5	7	10.82	93.3	61.41	403	85	24.75	21.04	7.01	15	3

Treatment	Genetics	Boar ID	# Collections	Gel WT (g)	Temp °F	Volume (ml)	Concentration (x10 ⁶)	Motility %	Total Concentration	Viable Sperm Concentration	# Doses (3x10 ⁹)	Morph	Glutination	
Clone	TX1		2	14	27.24	97.34	182.19	364.85	91.43	60.35	54.44	18.15	5.91	2.21
Clone	TX1		3	13	15.64	94.78	89.74	394.31	92.69	34.52	32.30	10.77	8.00	2.00
Clone	TX1		5	15	20.97	93.42	118.42	323.00	88.67	29.36	27.47	9.16	6.90	2.07
Clone	TX2		7	10	12.59	88.79	56.94	366.13	86.50	17.83	15.90	5.30	13.33	2.20

Denotes collections that were shipped and used to inseminate gilts for the production of offspring

										Gross Morphology					SpermVision Viable Cells per Dose (Billion)**	Total straws
Treatment	Boar ID	Date	Arrival Temp.	Pre/Post Freeze	Dose Volume	Conc. (million/ml)	Total Cells per Dose	Motility (%)	% dead cells	Abnormal Heads (%)	Abnormal Tails (%)	Proximal Drops	Distal Drops	%Normal (alive)*		
Control	H498	5/19/04	16.7	Pre	288.0	0.340	97.92	70.00%	30.00%	2.00%	19.50%	0.00%	6.50%	78.50%	53.81	
Control	H498	5/25/2004	16.4	Pre	263.0	0.300	78.90	66.00%	34.00%	0.00%	22.50%	0.00%	22.50%	77.50%	40.36	
Control	H498	6/16/2004	14.4	Pre	147.0	0.340	49.98	80.00%	20.00%	0.00%	9.50%	4.00%	21.00%	86.50%	34.59	
Control	H498	6/2/2004	14.5	Pre	264.0	0.340	89.76	78.00%	22.00%	1.50%	13.50%	0.00%	15.00%	85.00%	59.51	
Control	H498	6/9/2004	15.2	Pre	233.0	0.330	76.89	84.00%	16.00%	4.00%	7.00%	2.00%	18.00%	87.00%	56.19	
Control	H498	6/23/2004	16.8	Pre	224.2	0.280	62.78	79.00%	21.00%	2.50%	10.00%	0.00%	14.00%	87.50%	43.39	
Control	H498	7/8/2004		Pre	264.2	0.210	55.48	87.00%	13.00%	0.00%	8.50%	0.00%	17.00%	91.50%	44.17	
Clone	02	5/19/2004	15.1	Pre	245.0	0.290	71.05	52.00%	48.00%	0.00%	2.50%	0.00%	2.50%	97.50%	36.02	
Clone	03	5/19/2004	15.1	Pre	245.0	0.230	56.35	29.00%	71.00%	0.00%	2.50%	2.50%	10.00%	95.00%	15.52	
Clone	05	5/19/2004	15.1	Pre	246.0	0.190	46.74	33.00%	67.00%	0.00%	6.00%	12.00%	0.00%	82.00%	12.65	
Clone	03	5/25/2004	15	Pre	235.0	0.280	65.80	92.00%	8.00%	0.00%	12.00%	0.00%	0.00%	88.00%	53.27	
Clone	05	5/25/2004	15	Pre	245.0	0.180	44.10	93.00%	7.00%	4.00%	0.00%	0.00%	0.00%	96.00%	39.37	
Clone	02	5/25/2004	15	Pre	246.0	0.220	54.12	85.00%	15.00%	0.00%	3.00%	0.00%	9.00%	97.00%	44.62	
Control	H498	5/19/2004	16.7	Post	5.0	0.720	3.60	49.00%	51.00%	1.00%	25.00%	1.00%	6.00%	73.00%	1.29	13
Control	H498	5/25/2004	16.4	Post	5.0	0.980	4.90	64.00%	36.00%	1.00%	8.00%	1.00%	12.50%	90.00%	2.82	11
Control	H498	6/16/2004	14.4	Post	5.0	1.070	5.35	63.00%	37.00%	2.00%	8.00%	2.00%	6.50%	88.00%	2.97	7
Control	H498	6/2/2004	14.5	Post	5.0	1.000	5.00	74.00%	26.00%	1.00%	8.50%	1.50%	5.00%	89.00%	3.29	12
Control	H498	6/23/2004	16.8	Post	5.0	1.050	5.25	60.00%	40.00%	1.00%	6.50%	1.00%	6.00%	91.50%	2.88	9
Control	H498	6/9/2004	15.2	Post	5.0	1.100	5.50	65.00%	35.00%	2.00%	7.00%	0.00%	9.00%	91.00%	3.25	10
Control	H498	7/8/2004		Post	5.0	1.240	6.20	65.00%	35.00%	0.00%	2.00%	0.00%	3.50%	98.00%	3.95	8
Clone	02	5/19/2004	15.1	Post	5.0	1.050	5.25	75.00%	25.00%	0.00%	4.50%	0.00%	1.50%	95.50%	3.76	8
Clone	03	5/19/2004	15.1	Post	5.0	1.090	5.45	69.00%	31.00%	4.50%	1.50%	0.00%	1.50%	94.00%	3.53	6
Clone	05	5/19/2004	15.1	Post	5.0	1.170	5.85	71.00%	29.00%	1.00%	0.00%	1.00%	2.50%	98.00%	4.07	4
Clone	03	5/25/2004	15	Post	5.0	0.640	3.20	79.00%	21.00%	0.50%	3.00%	1.50%	2.50%	95.00%	2.40	7
Clone	05	5/25/2004	15	Post	5.0	0.640	3.20	84.00%	16.00%	1.00%	1.50%	1.00%	1.00%	96.50%	2.59	6
Clone	02	5/25/2004	15	Post	5.0	0.600	3.00	88.00%	12.00%	0.00%	1.50%	0.00%	1.50%	98.50%	2.60	7
Treatment	Genetics	Boar ID	Pre/Post Freeze	# of shipments	Dose Volume	Conc. (million/ml)	Total Cells per Dose	Motility (%)	% dead cells	Abnormal Heads (%)	Abnormal Tails (%)	Proximal Drops	Distal Drops	%Normal (alive)*	SpermVision Viable Cells per Dose (Billion)**	Total straws
Clone	TX1	2	Pre	2	245.50	0.26	62.59	68.50%	31.50%	0.00%	3.00%	0.00%	6.00%	97.50%	40.32	
Clone	TX1	3	Pre	2	240.00	0.26	61.08	60.50%	39.50%	0.00%	7.50%	1.50%	5.00%	91.50%	34.40	
Clone	TX1	5	Pre	2	245.50	0.19	45.42	63.00%	37.00%	2.00%	3.00%	6.00%	0.00%	89.00%	26.01	
Control	TX1	H498	Pre	7	240.49	0.31	73.10	77.71%	22.29%	1.57%	13.29%	0.86%	16.43%	85.14%	47.43	
Clone	TX1	2	Post	2	5.00	0.83	4.13	81.50%	18.50%	0.00%	3.50%	0.00%	2.00%	97.50%	3.18	15
Clone	TX1	3	Post	2	5.00	0.87	4.33	74.00%	26.00%	3.00%	2.50%	1.00%	2.50%	94.50%	2.97	13
Clone	TX1	5	Post	2	5.00	0.91	4.53	77.50%	22.50%	1.00%	1.00%	1.00%	2.00%	97.50%	3.33	10
Control	TX1	H498	Post	7	5.00	1.02	5.11	62.86%	37.14%	1.14%	9.43%	1.00%	7.14%	88.71%	2.92	70

Boar ID	Treatment	Line	Age (Months)	Location	Sample Date	Tech	Vol (ml)	Conc/mlx10 ⁹	Total Conc.x 10 ⁹	Total Motility %	Progressive Motility %	% Normal Heads	% Normal Tails
2	Clone	TX1	12	ViaGen	11-Mar-04	RC	85	0.061	5.19	87.93	76.72	20	86
2	Clone	TX1	12	ViaGen	16-Mar-04	RC	85	0.046	3.91	68.88	60	96	94
2	Clone	TX1	12	ViaGen	3-Mar-04	RC	85	0.074	6.29	90.22	77.44		
3	Clone	TX1	12	ViaGen	11-Mar-04	RC	85	0.108	9.18	78.28	65.65	39	84
3	Clone	TX1	12	ViaGen	16-Mar-04	RC	85	0.133	11.31	81.1	74.01	97	95
3	Clone	TX1	12	ViaGen	18-Mar-04	RC	85	0.043	3.66	75.32	71.42	97	93
3	Clone	TX1	12	ViaGen	23-Mar-04	RC	85	0.05	4.25	85.1	76.59	94	92
5	Clone	TX1	10	ViaGen	11-Mar-04	RC	85	0.096	8.16	82.35	75.49	17	92
5	Clone	TX1	10	ViaGen	18-Mar-04	RC	85	0.027	2.30	90	76	100	83
7	Clone	TX2	7	ViaGen	16-Mar-04	RC	85	0.043	3.66	79.26	70.73	69	96
7	Clone	TX2	7	ViaGen	25-Mar-04	RC	85	0.035	2.98	73.01	50.79	97	68
7	Clone	TX2	7	ViaGen	26-Mar-04	RC	85	0.089	7.57	88.63	60.22	96	84
7	Clone	TX2	7	ViaGen	30-Mar-04	RC	85	0.03	2.55	75.86	68.96	100	70
H498	Control	TX1	60	Farm A	11-Mar-04	RC	85	0.074	6.29	77.08	70.83	26	76
H498	Control	TX1	60	Farm A	16-Mar-04	RC	85	0.039	3.32	68.49	63.01	79	100
H498	Control	TX1	60	Farm A	18-Mar-04	RC	85	0.046	3.91	82.02	77.52	94	86
18128	Control	TX2	7	Farm A	16-Mar-04	RC	85	0.03	2.55	68.33	65	83	96
18128	Control	TX2	7	Farm A	25-Mar-04	RC	85	0.046	3.91	13.33	6.74	93	65
18128	Control	TX2	7	Farm A	30-Mar-04	RC	85	0.03	2.55	56.45	53.22	94	78
18128	Control	TX2	7	Farm A	2-Apr-04	RC	85	0.013	1.11	75	72.72	100	86
25515	Control	TX2	7	Farm A	18-Mar-04	RC	85	0.03	2.55	83.33	68.33	96	80
25515	Control	TX2	7	Farm A	25-Mar-04	RC	85	0.03	2.55	67.74	50	97	90
25515	Control	TX2	7	Farm A	26-Mar-04	RC	85	0.067	5.70	72.72	40.9	100	69
25515	Control	TX2	7	Farm A	30-Mar-04	RC	85	0.023	1.96	84.78	69.56	96	96

Treatment	Genetics	Boar ID	# Observations	Vol (ml)	Conc/mlx10 ⁹	Total Conc.x 10 ⁹	Total Motility %	Progressive Motility %	% Normal Heads	% Normal Tails
Clone	TX1	2	3	85	0.060	5.13	82.34	71.39	58.00	90.00
Clone	TX1	3	4	85	0.084	7.10	79.95	71.92	81.75	91.00
Clone	TX1	5	2	85	0.062	5.23	86.18	75.75	58.50	87.50
Clone	TX2	7	4	85	0.049	4.19	79.19	62.68	90.50	79.50
Control	TX1	H498	3	85	0.053	4.51	75.86	70.45	66.33	87.33
Control	TX2	18128	4	85	0.030	2.53	53.28	49.42	92.50	81.25
Control	TX2	25515	4	85	0.038	3.19	77.14	57.20	97.25	83.75

Boar ID	Genetics	Treatment	Gilt ID	Dam Sire Code	First Date Bred	Second Date Bred	Bred	30 Day Preg. Check		110 Day Preg. Check		Farrowing Date	Response
								Date	Response	Date	Response		
2	Ham	Clone	0337302	B	3/20/2000	3/21/2000	1	4/19/2000	0	7/8/2000	0		0
2	Ham	Clone	0337401	E	3/18/2000	3/19/2000	1	4/17/2000	1	7/6/2000	1	7/12/2004	1
2	Ham	Clone	0338205	C	3/15/2000	3/16/2000	1	4/14/2000	1	7/3/2000	1	7/7/2004	1
2	Ham	Clone	0340903	F	3/15/2000	3/16/2000	1	4/14/2000	1	7/3/2000	1	7/10/2004	1
2	Ham	Clone	0343201	A	3/15/2000	3/16/2000	1	4/14/2000	1	7/3/2000	1	7/10/2004	1
2	Ham	Clone	0343301	G	3/17/2000	3/18/2000	1	4/16/2000	1	7/5/2000	0		0
2	Ham	Clone	0343904	J	3/20/2000	3/21/2000	1	4/19/2000	0	7/8/2000	0		0
2	Ham	Clone	0344002	N	3/17/2000	3/18/2000	1	4/16/2000	1	7/5/2000	1	7/12/2004	1
2	Ham	Clone	0344102	H	3/21/2000	3/22/2000	1	4/20/2000	1	7/9/2000	1	7/16/2004	1
2	Ham	Clone	0344701	K	3/24/2000	3/25/2000	1	4/23/2000	0	7/12/2000	0		0
2	Ham	Clone	0344904	D	3/15/2000	3/16/2000	1	4/14/2000	1	7/3/2000	1	7/8/2004	1
2	Ham	Clone	0345601	M	3/16/2000	3/17/2000	1	4/15/2000	1	7/4/2000	0		0
3	Ham	Clone	0336707	G	3/19/2000	3/20/2000	1	4/18/2000	1	7/7/2000	1	7/12/2004	1
3	Ham	Clone	0337003	E	3/22/2000	3/23/2000	1	4/21/2000	1	7/10/2000	1	7/15/2004	1
3	Ham	Clone	0337501	K	3/26/2000	2/27/2000	1	4/25/2000	1	7/14/2000	1	7/20/2004	1
3	Ham	Clone	0339303	D	3/19/2000	3/20/2000	1	4/18/2000	1	7/7/2000	1	7/13/2004	1
3	Ham	Clone	0340904	F	3/15/2000	3/16/2000	1	4/14/2000	1	7/3/2000	1	7/8/2004	1
3	Ham	Clone	0341201	M	3/18/2000	3/19/2000	1	4/17/2000	1	7/6/2000	1	7/13/2004	1
3	Ham	Clone	0341501	H	3/21/2000	3/22/2000	1	4/20/2000	1	7/9/2000	1	7/13/2004	1
3	Ham	Clone	0342004	A	3/18/2000	3/19/2000	1	4/17/2000	1	7/6/2000	1	7/12/2004	1
3	Ham	Clone	0342401	N	3/16/2000	3/17/2000	1	4/15/2000	0	7/4/2000	0		0
3	Ham	Clone	0343801	C	3/15/2000	3/16/2000	1	4/14/2000	0	7/3/2000	0		0
3	Ham	Clone	0344402	B	3/21/2000	3/22/2000	1	4/20/2000	1	7/9/2000	1	7/16/2004	1
3	Ham	Clone	0345003	J	3/22/2000	3/23/2000	1	4/21/2000	1	7/10/2000	1	7/13/2004	1
5	Ham	Clone	0336305	B	3/21/2000	3/22/2000	1	4/20/2000	0	7/9/2000	0		0
5	Ham	Clone	0336705	G	3/16/2000	3/17/2000	1	4/15/2000	1	7/4/2000	1	7/7/2004	1
5	Ham	Clone	0339104	N	3/24/2000	3/25/2000	1	4/23/2000	1	7/12/2000	1	7/19/2004	1
5	Ham	Clone	0340601	M	3/16/2000	3/17/2000	1	4/15/2000	1	7/4/2000	0		0
5	Ham	Clone	0342501	E	3/15/2000	3/16/2000	1	4/14/2000	0	7/3/2000	0		0
5	Ham	Clone	0342902	C	3/18/2000	3/19/2000	1	4/17/2000	1	7/6/2000	1	7/13/2004	1
5	Ham	Clone	0343105	J	3/22/2000	3/23/2000	1	4/21/2000	0	7/10/2000	0		0
5	Ham	Clone	0344201	K	3/25/2000	3/26/2000	1	4/24/2000	1	7/13/2000	1	7/19/2004	1
5	Ham	Clone	0344902	D	3/15/2000	3/16/2000	1	4/14/2000	1	7/3/2000	1	7/10/2004	1
5	Ham	Clone	0345103	A	3/17/2000	3/18/2000	1	4/16/2000	1	7/5/2000	1	7/11/2004	1
5	Ham	Clone	0345302	H	3/20/2000	3/21/2000	1	4/19/2000	1	7/8/2000	1	7/15/2004	1
5	Ham	Clone	0345501	F	3/15/2000	3/16/2000	1	4/14/2000	1	7/3/2000	1	7/7/2004	1

Boar ID	Genetics	Treatment	Gilt ID	Dam Sire Code	First Date Bred	Second Date Bred	Bred	30 Day Preg. Check		110 Day Preg. Check		Farrowing Date	Response
								Date	Response	Date	Response		
7	Duroc	Clone	0337102	K	3/26/2000	3/27/2000	1	4/25/2000	0	7/14/2000	0		0
7	Duroc	Clone	0337602	A	3/26/2000	3/27/2000	1	4/25/2000	1	7/14/2000	1	7/15/2004	1
7	Duroc	Clone	0338102	A	3/26/2000	3/27/2000	1	4/25/2000	1	7/14/2000	1	7/19/2004	1
7	Duroc	Clone	0338203	C	3/30/2000	3/31/2000	1	4/29/2000	1	7/18/2000	1	7/22/2004	1
7	Duroc	Clone	0339102	N	3/27/2000	3/28/2000	1	4/26/2000	1	7/15/2000	1	7/22/2004	1
7	Duroc	Clone	0339704	J	3/30/2000		1	4/29/2000	1	7/18/2000	1	7/23/2004	1
7	Duroc	Clone	0340206	M	3/26/2000	3/27/2000	1	4/25/2000	1	7/14/2000	1	7/20/2004	1
7	Duroc	Clone	0340906	F	3/30/2000	3/31/2000	1	4/29/2000	1	7/18/2000	1	7/23/2004	1
7	Duroc	Clone	0342201	K	3/28/2000	3/29/2000	1	4/27/2000	1	7/16/2000	1	7/22/2004	1
7	Duroc	Clone	0342502	E	4/1/2000	4/2/2000	1	5/1/2000	1	7/20/2000	1	7/24/2004	1
7	Duroc	Clone	0346101	D	3/20/2000	3/21/2000	1	4/19/2000	1	7/8/2000	1	7/14/2004	1
H498	Ham	Control	0336706	G	3/18/2000	3/19/2000	1	4/17/2000	0	7/6/2000	0		0
H498	Ham	Control	0338103	A	3/24/2000	3/25/2000	1	4/23/2000	1	7/12/2000	1	7/17/2004	1
H498	Ham	Control	0340902	F	3/18/2000	3/19/2000	1	4/17/2000	1	7/6/2000	1	7/13/2004	1
H498	Ham	Control	0342304	H	3/15/2000	3/16/2000	1	4/14/2000	1	7/3/2000	1	7/7/2004	1
H498	Ham	Control	0342601	E	3/20/2000	3/21/2000	1	4/19/2000	1	7/8/2000	1	7/13/2004	1
H498	Ham	Control	0342802	B	3/16/2000	3/18/04	1	4/15/2000	0	7/4/2000	0		0
H498	Ham	Control	0343106	J	3/21/2000	3/21/2000	1	4/20/2000	1	7/9/2000	1		0
H498	Ham	Control	0344601	M	3/16/2000	3/17/2000	1	4/15/2000	0	7/4/2000	0		0
H498	Ham	Control	0344706	K	3/17/2000	3/18/2000	1	4/16/2000	1	7/5/2000	1		0
H498	Ham	Control	0345405	C	3/20/2000	3/21/2000	1	4/19/2000	0	7/8/2000	0		0
H498	Ham	Control	0345803	D	3/18/2000	3/19/2000	1	4/17/2000	1	7/6/2000	1	7/13/2004	1
H498	Ham	Control	0346006	N	3/15/2000	3/16/2000	1	4/14/2000	0	7/3/2000	0		0
18128	Duroc	Control	0336204	G	4/2/2000		1	5/2/2000	1	7/21/2000	1	7/24/2004	1
18128	Duroc	Control	0336502	A	3/31/2000	4/1/2000	1	4/30/2000	1	7/19/2000	1	7/23/2004	1
18128	Duroc	Control	0337403	E	3/22/2000	3/23/2000	1	4/21/2000	1	7/10/2000	1	7/13/2004	1
18128	Duroc	Control	0339501	H	3/30/2000	3/31/2000	1	4/29/2000	1	7/18/2000	1		0
18128	Duroc	Control	0339603	N	4/2/2000	4/3/2000	1	5/2/2000	1	7/21/2000	1	7/23/2004	1
18128	Duroc	Control	0340901	F	3/31/2000		1	4/30/2000	1	7/19/2000	1	7/25/2004	1
18128	Duroc	Control	0341702	K	4/2/2000	4/3/2000	1	5/2/2000	1	7/21/2000	1	7/26/2004	1
18128	Duroc	Control	0342905	C	3/22/2000	3/23/2000	1	4/21/2000	1	7/10/2000	1	7/12/2004	1
18128	Duroc	Control	0343107	J	4/2/2000	4/3/2000	1	5/2/2000	1	7/21/2000	1		0
18128	Duroc	Control	0343802	C	4/2/2000	4/3/2000	1	5/2/2000	1	7/21/2000	1	7/24/2004	1
18128	Duroc	Control	0344403	B	3/23/2000	3/23/2000	1	4/22/2000	1	7/11/2000	1	7/17/2004	1
18128	Duroc	Control	0345603	M	3/22/2000	3/23/2000	1	4/21/2000	1	7/10/2000	1	7/15/2004	1
18128	Duroc	Control	0346003	N	3/31/2000	4/1/2000	1	4/30/2000	0	7/19/2000	0		0

Boar ID	Genetics	Treatment	Gilt ID	Dam Sire Code	First Date Bred	Second Date Bred	Bred	30 Day Preg. Check		110 Day Preg. Check		Farrowing Date	Response
								Date	Response	Date	Response		
25515	Duroc	Control	0337303	B	3/31/2000	4/1/2000	1	4/30/2000	1	7/19/2000	1	7/27/2004	1
25515	Duroc	Control	0337901	N	3/30/2000	3/31/2000	1	4/29/2000	0	7/18/2000	0		0
25515	Duroc	Control	0338501	G	3/30/2000	3/31/2000	1	4/29/2000	1	7/18/2000	1	7/24/2004	1
25515	Duroc	Control	0339105	N	3/31/2000	4/1/2000	1	4/30/2000	1	7/19/2000	1	7/23/2004	1
25515	Duroc	Control	0339403	H	3/26/2000	3/27/2000	1	4/25/2000	1	7/14/2000	1	7/17/2004	1
25515	Duroc	Control	0340001	A	3/26/2000	3/27/2000	1	4/25/2000	1	7/14/2000	1	7/19/2004	1
25515	Duroc	Control	0340204	M	3/22/2000	3/23/2000	1	4/21/2000	1	7/10/2000	1	7/15/2004	1
25515	Duroc	Control	0340301	E	3/22/2000	3/23/2000	1	4/21/2000	0	7/10/2000	0		0
25515	Duroc	Control	0340702	J	3/31/2000	4/1/2000	1	4/30/2000	1	7/19/2000	1	7/22/2004	1
25515	Duroc	Control	0343605	F	3/26/2000	3/27/2000	1	4/25/2000	1	7/14/2000	1	7/18/2004	1
25515	Duroc	Control	0344501	K	3/26/2000	3/27/2000	1	4/25/2000	0	7/14/2000	0		0
25515	Duroc	Control	0345404	C	3/23/2000	3/24/2000	1	4/22/2000	0	7/11/2000	0		0
25515	Duroc	Control	0346104	D	3/22/2000	3/23/2000	1	4/21/2000	1	7/10/2000	1	7/13/2004	1

Boar ID	# Gilts Bred	# Gilts Preg.	% Preg	# Gilts Preg	% Preg	# Gilts Farrowed	% Gilts Farrowed
		Day 30	Day 30	Day 110	Day 110		
2	12	9	75.00%	7	58.33%	7	58.33%
3	12	10	83.33%	10	83.33%	10	83.33%
5	12	9	75.00%	8	66.67%	8	66.67%
7	11	10	90.91%	10	90.91%	10	90.91%
H498	12	7	58.33%	7	58.33%	5	41.67%
18128	13	12	92.31%	12	92.31%	10	76.92%
25515	13	9	69.23%	9	69.23%	9	69.23%
Total	85	66	77.65%	63	74.12%	59	69.41%

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA	
200439201	2	7/24/2004	3.7	200398128	Duroc	Control	200336204	G	14	1			2	1	8/12/04	19	14.2	0.55	9/14/2004	1	52	49.4	1.07	0.88	
200439202	2	7/24/2004	3.3	200398128	Duroc	Control	200336204	G	14	1			2	1	8/12/04	19	16	0.67	9/14/2004	1	52	48.4	0.98	0.87	
200439203	2	7/24/2004	3.1	200398128	Duroc	Control	200336204	G	15	1			2	1	8/12/04	19	12.6	0.50	9/14/2004	1	52	42.0	0.89	0.75	
200439204	2	7/24/2004	2.8	200398128	Duroc	Control	200336204	G	16	1			2	1	8/13/04	20	11.4	0.43	9/15/2004	1	53	42.3	0.94	0.75	
200439205	2	7/24/2004	2.2	200398128	Duroc	Control	200336204	G	12	1			2	1	8/13/04	20	11.4	0.46	9/15/2004	1	53	41.0	0.90	0.73	
200439206	1	7/24/2004	4.1	200398128	Duroc	Control	200336204	G	12	1			3	1	8/13/04	20	15	0.55	9/15/2004	1	53	55.1	1.22	0.96	
200439207	1	7/24/2004	3.2	200398128	Duroc	Control	200336204	G	13	1			3	1	8/13/04	20	11.7	0.43	9/15/2004	1	53	42.4	0.93	0.74	
200439208	1	7/24/2004	3.3	200398128	Duroc	Control	200336204	G	14	1			3	1	8/13/04	20	13.4	0.51	9/15/2004	1	53	45.0	0.96	0.79	
200439209	1	7/24/2004	3.2	200398128	Duroc	Control	200336204	G	14	1			3	1	8/13/04	20	11.5	0.42	9/15/2004	1	53	40.9	0.89	0.71	
200439210	1	7/24/2004	3.2	200398128	Duroc	Control	200336204	G	14	1			3	1	8/13/04	20	12.4	0.46	9/15/2004	1	53	46.7	1.04	0.82	
200439211	1	7/24/2004	3.0	200398128	Duroc	Control	200336204	G	14	1			3	1	8/13/04	20	12.8	0.49	9/15/2004	1	53	49.2	1.10	0.87	
200439212	1	7/24/2004	2.8	200398128	Duroc	Control	200336204	G	14	1			3	1	8/13/04	20	10.9	0.41	9/15/2004	1	53	44.2	1.01	0.78	
200439215	1	7/24/2004	2.8	200398128	Duroc	Control	200336204	G	14	1			3	1	8/13/04	20	9.4	0.36	9/15/2004	1	53	40.2	0.93	0.76	
200438501	2	7/23/2004	3.7	200398128	Duroc	Control	200336502	A	11	1			2	1	8/6/04	14	4.7	0.07	9/9/2004	1	48	30.6	0.76	0.56	
200438502	2	7/23/2004	3.6	200398128	Duroc	Control	200336502	A	12	1			2	1	8/6/04	14	6.2	0.19	9/9/2004	1	48	34.0	0.82	0.63	
200438503	2	7/23/2004	3.7	200398128	Duroc	Control	200336502	A	10	1			2	1	8/6/04	14	8.2	0.32	9/9/2004	1	48	38.2	0.88	0.72	
200438504	2	7/23/2004	3.7	200398128	Duroc	Control	200336502	A	14	1			2	1	8/12/04	20	14.4	0.54	9/14/2004	1	53	45.4	0.94	0.79	
200438505	2	7/23/2004	3.3	200398128	Duroc	Control	200336502	A	14	1			2	1	8/12/04	20	13.2	0.50	9/14/2004	1	53	45.0	0.96	0.79	
200438506	2	7/23/2004	2.8	200398128	Duroc	Control	200336502	A	12	1			2	1	8/12/04	20	12.6	0.49	9/14/2004	1	53	44.0	0.95	0.78	
200438507	2	7/23/2004	3.3	200398128	Duroc	Control	200336502	A	12	1			2	1	8/12/04	20	16.1	0.64	9/14/2004	1	53	53.4	1.13	0.95	
200438508	2	7/23/2004	2.4	200398128	Duroc	Control	200336502	A	13	1			2	1	8/12/04	20	8	0.28	9/14/2004	1	53	34.6	0.81	0.61	
200438509	2	7/23/2004	2.4	200398128	Duroc	Control	200336502	A	11	1			2	1	8/12/04	20	11.2	0.44	9/14/2004	1	53	41.6	0.92	0.74	
200438510	2	7/23/2004	2.0	200398128	Duroc	Control	200336502	A	12	1			2	1	8/12/04	20	11	0.45	9/14/2004	1	53	39.8	0.87	0.71	
200438511	1	7/23/2004	3.8	200398128	Duroc	Control	200336502	A	14	1			3	1	8/12/04	20	15.6	0.59	9/14/2004	1	53	53.6	1.15	0.94	
200438512	1	7/23/2004	3.4	200398128	Duroc	Control	200336502	A	12	1			3	1	8/12/04	20	12.4	0.45	9/14/2004	1	53	45.6	1.01	0.80	
200438513	1	7/23/2004	3.3	200398128	Duroc	Control	200336502	A	12	1			3	1	8/12/04	20	13.5	0.51	9/14/2004	1	53	46.0	0.98	0.81	
200438514	1	7/23/2004	3.3	200398128	Duroc	Control	200336502	A	11	1			3	1	8/12/04	20	14.1	0.54	9/14/2004	1	53	45.6	0.95	0.80	
200433801	2	7/14/2004	4.3	200390007	Duroc	Clone	200336702	G	14	1			2	1	7/30/04	16	10.1	0.36	9/2/2004	1	50	43.0	0.97	0.77	
200433802	2	7/14/2004	3.8	200390007	Duroc	Clone	200336702	G	13	1			2	1	7/30/04	16	9.1	0.33	9/2/2004	1	50	33.4	0.71	0.59	
200433803	2	7/14/2004	3.8	200390007	Duroc	Clone	200336702	G	13	1			2	1	7/30/04	16	9.2	0.34	9/2/2004	1	50	37.6	0.84	0.68	
200433804	2	7/14/2004	3.8	200390007	Duroc	Clone	200336702	G	13	1			2	1	7/30/04	16	9.1	0.33	9/2/2004	1	50	37.4	0.83	0.67	
200433805	2	7/14/2004	2.9	200390007	Duroc	Clone	200336702	G	13	1			0								0				
200433806	1	7/14/2004	4.1	200390007	Duroc	Clone	200336702	G	13	1			3	1	7/30/04	16	10.3	0.39	9/2/2004	1	50	35.4	0.74	0.63	
200433807	1	7/14/2004	4.4	200390007	Duroc	Clone	200336702	G	13	1			3	1	7/30/04	16	11.2	0.43	9/2/2004	1	50	47.6	1.07	0.86	
200433808	1	7/14/2004	4.0	200390007	Duroc	Clone	200336702	G	12	1			3	1	7/30/04	16	10.5	0.41	9/2/2004	1	50	39.8	0.86	0.72	
200433809	1	7/14/2004	3.8	200390007	Duroc	Clone	200336702	G	12	1			3	1	7/30/04	16	10.3	0.41	9/2/2004	1	50	28.2	0.53	0.49	
200433810	2	7/14/2004	3.6	200390007	Duroc	Clone	200336702	G	12	1	7/14/2004	OVRL	0								0				
200433811	1	7/14/2004	3.4	200390007	Duroc	Clone	200336702	G	12	1	7/15/2004	OVRL	0								0				
200430601	2	7/7/2004	2.9	200390005	H498	Clone	200336705	G	13	1			2	1	7/27/04	20	10.9	0.40	9/1/2004	1	56	33.8	0.64	0.55	
200430602	2	7/7/2004	2.3	200390005	H498	Clone	200336705	G	12	1			2	1	7/27/04	20	11.6	0.47	9/1/2004	1	56	39.8	0.78	0.67	
200430603	2	7/7/2004	2.4	200390005	H498	Clone	200336705	G	14	1			2	1	7/27/04	20	9.8	0.37	9/1/2004	1	56	35.6	0.72	0.59	
200430604	1	7/7/2004	3.5	200390005	H498	Clone	200336705	G	14	1			3	1	7/27/04	20	12.1	0.43	9/1/2004	1	56	48.4	1.01	0.80	
200430605	1	7/7/2004	3.4	200390005	H498	Clone	200336705	G	15	1			3	1	7/27/04	20	10.8	0.37	9/1/2004	1	56	41.4	0.85	0.68	
200430606	1	7/7/2004	3.2	200390005	H498	Clone	200336705	G	15	1			3	1	7/27/04	20	11	0.39	9/1/2004	1	56	32.6	0.60	0.53	
200430607	1	7/7/2004	3.1	200390005	H498	Clone	200336705	G	14	1			3	1	7/27/04	20	10.9	0.39	9/1/2004	1	56	37.8	0.75	0.62	
200430608	1	7/7/2004	3.1	200390005	H498	Clone	200336705	G	13	1			3	1	7/27/04	20	9.9	0.34	9/1/2004	1	56	40.8	0.86	0.67	
200430609	1	7/7/2004	3.1	200390005	H498	Clone	200336705	G	12	6			3	1	7/27/04	20	9.1	0.30	9/1/2004	1	56	37.2	0.78	0.61	
200430610	1	7/7/2004	3.0	200390005	H498	Clone	200336705	G	14	1			3	1	7/27/04	20	12.7	0.49	9/1/2004	1	56	37.8	0.70	0.62	
200430611	1	7/7/2004	2.3	200390005	H498	Clone	200336705	G	12	1			3	1	7/27/04	20	10.6	0.42	9/1/2004	1	56	34.8	0.67	0.58	
200430612	1	7/7/2004	3.4	200390005	H498	Clone	200336705	G	14	1	7/7/2004	STLB	0								0				
200432301	2	7/12/2004	3.7	200390003	H498	Clone	200336707	G	14	1			2	1	7/27/04	15	9.2	0.37	9/2/2004	1	52	32.8	0.64	0.56	

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA		
200432302	2	7/12/2004	3.4	200390003	H498	Clone	200336707	G	14	1			2	1	7/27/04	15	8.3	0.33	9/2/2004	1	52	28.6	0.55	0.48		
200432303	2	7/12/2004	3.2	200390003	H498	Clone	200336707	G	15	1			2	1	7/27/04	15	11.3	0.54	9/2/2004	1	52	35.8	0.66	0.63		
200432304	2	7/12/2004	2.9	200390003	H498	Clone	200336707	G	14	1			2	1	7/27/04	15	10.4	0.50	9/2/2004	1	52	37.2	0.72	0.66		
200432305	2	7/12/2004	2.4	200390003	H498	Clone	200336707	G	13	1			2	1	7/27/04	15	8.4	0.40	9/2/2004	1	52	28.2	0.54	0.50		
200432306	2	7/12/2004	2.3	200390003	H498	Clone	200336707	G	14	1			2	1	7/27/04	15	8	0.38	9/2/2004	1	52	26.8	0.51	0.47		
200432307	1	7/12/2004	4.0	200390003	H498	Clone	200336707	G	14	1			3	1	7/27/04	15	10	0.40	9/2/2004	1	52	30.6	0.56	0.51		
200432308	1	7/12/2004	3.9	200390003	H498	Clone	200336707	G	14	1			3	1	7/27/04	15	11.6	0.51	9/2/2004	1	52	38.6	0.73	0.67		
200432309	1	7/12/2004	3.8	200390003	H498	Clone	200336707	G	14	1			3	1	7/27/04	15	11.1	0.49	9/2/2004	1	52	22.0	0.29	0.35		
200432310	1	7/12/2004	3.3	200390003	H498	Clone	200336707	G	14	1			3	1	7/27/04	15	10.1	0.45	9/2/2004	1	52	34.4	0.66	0.60		
200432311	1	7/12/2004	2.5	200390003	H498	Clone	200336707	G	15	1			3	1	7/27/04	15	9.6	0.47	9/2/2004	1	52	29.8	0.55	0.53		
200432312	1	7/12/2004	2.3	200390003	H498	Clone	200336707	G	13	1			3	1	7/27/04	15	7.9	0.37	9/2/2004	1	52	25.0	0.46	0.44		
200434101	2	7/15/2004	3.5	200390003	H498	Clone	200337003	E	14	1			2	1	7/30/04	15	6.8	0.22	9/2/2004	1	49	22.0	0.45	0.38		
200434102	2	7/15/2004	3.0	200390003	H498	Clone	200337003	E	15	1			2	1	7/30/04	15	6	0.20	9/2/2004	1	49	25.4	0.57	0.46		
200434103	2	7/15/2004	2.8	200390003	H498	Clone	200337003	E	14	1			2	1	7/30/04	15	7.4	0.31	9/2/2004	1	49	20.8	0.39	0.37		
200434104	2	7/15/2004	2.7	200390003	H498	Clone	200337003	E	15	1			2	1	7/30/04	15	8	0.35	9/2/2004	1	49	29.4	0.63	0.54		
200434105	2	7/15/2004	2.5	200390003	H498	Clone	200337003	E	14	1			2	1	7/30/04	15	8.2	0.38	9/2/2004	1	49	27.0	0.55	0.50		
200434106	1	7/15/2004	3.4	200390003	H498	Clone	200337003	E	16	1			3	1	7/30/04	15	9.2	0.39	9/2/2004	1	49	30.6	0.63	0.56		
200434107	1	7/15/2004	2.8	200390003	H498	Clone	200337003	E	14	1			3	1	7/30/04	15	8.7	0.39	9/2/2004	1	49	27.9	0.56	0.51		
200434108	1	7/15/2004	2.6	200390003	H498	Clone	200337003	E	14	1			3	1	7/30/04	15	8.2	0.37	9/2/2004	1	49	21.4	0.39	0.38		
200434109	1	7/15/2004	2.9	200390003	H498	Clone	200337003	E	14	1			3	1	7/30/04	15	9.8	0.46	9/2/2004	1	49	30.9	0.62	0.57		
200434110	1	7/15/2004	2.5	200390003	H498	Clone	200337003	E	13	1			3	1	7/30/04	15	7.9	0.36	9/2/2004	1	49	27.2	0.57	0.50		
200434111	1	7/15/2004	2.5	200390003	H498	Clone	200337003	E	15	1			3	1	7/30/04	15	8.3	0.39	9/2/2004	1	49	25.4	0.50	0.47		
200434112	1	7/15/2004	2.1	200390003	H498	Clone	200337003	E	13	1			3	1	7/30/04	15	7.4	0.35	9/2/2004	1	49	22.2	0.44	0.41		
200434113	1	7/15/2004	1.9	200390003	H498	Clone	200337003	E	12	1			3	1	7/30/04	15	5.4	0.23	9/1/2004	1	48	16.2	0.33	0.30		
200434114	1	7/15/2004	1.6	200390003	H498	Clone	200337003	E	14	1			3	1	7/30/04	15	7.3	0.38	9/2/2004	1	49	20.2	0.38	0.38		
200434115	1	7/15/2004	1.4	200390003	H498	Clone	200337003	E	14	1			3	1	7/30/04	15	5.6	0.28		0						
200441201	2	7/27/2004	3.3	200395515	Duroc	Control	200337303	E	15	1			2	1	8/13/04	17	13	0.57	9/15/2004	1	50	37.4	0.74	0.68		
200441202	2	7/27/2004	2.8	200395515	Duroc	Control	200337303	B	14	1			2	1	8/13/04	17	11	0.48	9/15/2004	1	50	37.8	0.81	0.70		
200441203	1	7/27/2004	3.8	200395515	Duroc	Control	200337303	B	15	1			3	1	8/13/04	17	10.6	0.40	9/15/2004	1	50	33.9	0.71	0.60		
200441204	1	7/27/2004	3.6	200395515	Duroc	Control	200337303	B	16	1			3	1	8/13/04	17	10.3	0.39	9/15/2004	1	50	34.6	0.74	0.62		
200441205	1	7/27/2004	3.7	200395515	Duroc	Control	200337303	B	15	1			3	1	8/13/04	17	10.3	0.39	9/15/2004	1	50	34.1	0.72	0.61		
200441206	1	7/27/2004	3.3	200395515	Duroc	Control	200337303	B	14	1			3	1	8/13/04	17	11.5	0.48	9/15/2004	1	50	40.0	0.86	0.73		
200441207	1	7/27/2004	3.2	200395515	Duroc	Control	200337303	B	15	1			3	1	8/13/04	17	9.7	0.38	9/15/2004	1	50	28.5	0.57	0.51		
200441208	1	7/27/2004	3.2	200395515	Duroc	Control	200337303	B	14	1			3	1	8/13/04	17	10	0.40	9/15/2004	1	50	38.8	0.87	0.71		
200441209	1	7/27/2004	2.6	200395515	Duroc	Control	200337303	B	15	1			3	1	8/13/04	17	11	0.49	9/15/2004	1	50	28.7	0.54	0.52		
200441210	1	7/27/2004	2.6	200395515	Duroc	Control	200337303	B	15	1			3	1	8/13/04	17	9.6	0.41	9/15/2004	1	50	32.4	0.69	0.60		
200441211	1	7/27/2004	2.4	200395515	Duroc	Control	200337303	B	14	1			3	1	8/13/04	17	7.4	0.29	9/15/2004	1	50	33.9	0.80	0.63		
200441212	2	7/27/2004	3.1	200395515	Duroc	Control	200337303	B	16	1			0								0					
200441213	1	7/27/2004	2.1	200395515	Duroc	Control	200337303	B	14	1	7/27/2004	STLB	0								0					
200432001	2	7/12/2004	3.4	200390002	H498	Clone	200337401	E	11	1			2	1	7/30/04	18	12.2	0.49	9/2/2004	1	52	42.2	0.88	0.75		
200432002	2	7/12/2004	3.3	200390002	H498	Clone	200337401	E	14	1			2	1	7/30/04	18	12.1	0.49	9/2/2004	1	52	42.6	0.90	0.76		
200432003	2	7/12/2004	3.1	200390002	H498	Clone	200337401	E	13	1			2	1	7/30/04	18	11.9	0.49	9/2/2004	1	52	40.6	0.84	0.72		
200432004	1	7/12/2004	3.6	200390002	H498	Clone	200337401	E	16	1			3	1	7/30/04	18	11.8	0.46	9/2/2004	1	52	45.8	1.00	0.81		
200432005	1	7/12/2004	3.2	200390002	H498	Clone	200337401	E	15	1			3	1	7/30/04	18	11.7	0.47	9/2/2004	1	52	39.8	0.83	0.70		
200432006	1	7/12/2004	3.1	200390002	H498	Clone	200337401	E	14	1			3	1	7/30/04	18	11.9	0.49	9/2/2004	1	52	40.4	0.84	0.72		
200432007	1	7/12/2004	3.1	200390002	H498	Clone	200337401	E	12	1			3	1	7/30/04	18	11.4	0.46	9/2/2004	1	52	41.4	0.88	0.74		
200432008	1	7/12/2004	2.7	200390002	H498	Clone	200337401	E	14	1			3	1	7/30/04	18	9.7	0.39	9/2/2004	1	52	30.0	0.60	0.53		
200432009	2	7/12/2004	2.5	200390002	H498	Clone	200337401	E	14	1	7/13/2004	OVRL	0								0					
200432010	1	7/12/2004	3.1	200390002	H498	Clone	200337401	E	14	1	7/13/2004	OVRL	0								0					
200432011	2	7/12/2004	1.6	200390002	H498	Clone	200337401	E	15	1	7/12/2004	STLB	0								0					
200432601	2	7/13/2004	3.7	200398128	Duroc	Control	200337403	E	10	1			2	1	7/30/04	17	12.1	0.49	9/2/2004	1	51	38.6	0.78	0.68		
200432602	2	7/13/2004	3.4	200398128	Duroc	Control	200337403	E	15	1			2	1	7/30/04	17	12.3	0.52	9/2/2004	1	51	34.8	0.66	0.62		

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA	
200432603	2	7/13/2004	3.3	200398128	Duroc	Control	200337403	E	13	1			2	1	7/30/04	17	10.5	0.42	9/2/2004	1	51	39.0	0.84	0.70	
200432604	2	7/13/2004	3.2	200398128	Duroc	Control	200337403	E	13	1			2	1	7/30/04	17	12.8	0.56	9/2/2004	1	51	40.0	0.80	0.72	
200432605	2	7/13/2004	3.0	200398128	Duroc	Control	200337403	E	12	1			2	1	7/30/04	17	9.8	0.40	9/2/2004	1	51	31.6	0.64	0.56	
200432606	2	7/13/2004	2.9	200398128	Duroc	Control	200337403	E	12	1				0							0				
200432607	1	7/13/2004	3.6	200398128	Duroc	Control	200337403	E	13	1			3	1	7/30/04	17	11.6	0.47	9/2/2004	1	51	37.2	0.75	0.66	
200432608	1	7/13/2004	3.3	200398128	Duroc	Control	200337403	E	12	1			3	1	7/30/04	17	11	0.45	9/2/2004	1	51	32.2	0.62	0.57	
200432609	1	7/13/2004	3.5	200398128	Duroc	Control	200337403	E	15	1			3	1	7/30/04	17	11.5	0.47	9/2/2004	1	51	37.8	0.77	0.67	
200432610	1	7/13/2004	3.3	200398128	Duroc	Control	200337403	E	13	1			3	1	7/30/04	17	10.7	0.44	9/2/2004	1	51	32.0	0.63	0.56	
200436901	2	7/20/2004	3.3	200390003	H498	Clone	200337501	K	14	1			2	1	8/6/04	17	9.7	0.38	9/7/2004	1	49	31.2	0.67	0.57	
200436902	2	7/20/2004	3.2	200390003	H498	Clone	200337501	K	13	1			2	1	8/6/04	17	10	0.40	9/7/2004	1	49	38.2	0.88	0.71	
200436903	2	7/20/2004	2.8	200390003	H498	Clone	200337501	K	12	1			2	1	8/6/04	17	9.7	0.41	9/7/2004	1	49	31.4	0.68	0.58	
200436904	2	7/20/2004	2.8	200390003	H498	Clone	200337501	K	16	1			2	1	8/6/04	17	11.5	0.51	9/9/2004	1	51	35.2	0.70	0.64	
200436905	2	7/20/2004	2.6	200390003	H498	Clone	200337501	K	13	1			2	1	8/6/04	17	8.4	0.34	9/9/2004	1	51	29.2	0.61	0.52	
200436906	2	7/20/2004	2.4	200390003	H498	Clone	200337501	K	13	6			2	1	8/6/04	17	7.9	0.32	9/9/2004	1	51	24.0	0.47	0.42	
200436907	2	7/20/2004	2.3	200390003	H498	Clone	200337501	K	15	1			2	1	8/6/04	17	7	0.28	9/9/2004	1	51	27.4	0.60	0.49	
200436908	2	7/20/2004	2.3	200390003	H498	Clone	200337501	K	13	1			2	1	8/6/04	17	8.8	0.38	9/9/2004	1	51	28.0	0.56	0.50	
200436909	1	7/20/2004	3.6	200390003	H498	Clone	200337501	K	14	1			3	1	8/6/04	17	12.3	0.51	9/9/2004	1	51	42.8	0.90	0.77	
200436910	1	7/20/2004	2.8	200390003	H498	Clone	200337501	K	14	1			3	1	8/6/04	17	11.6	0.52	9/9/2004	1	51	37.0	0.75	0.67	
200436911	1	7/20/2004	2.2	200390003	H498	Clone	200337501	K	14	1			3	1	8/6/04	17	9.8	0.45	9/9/2004	1	51	35.8	0.76	0.66	
200436912	1	7/20/2004	2.2	200390003	H498	Clone	200337501	K	13	1			3	1	8/6/04	17	9.5	0.43	9/9/2004	1	51	30.2	0.61	0.55	
200436913	1	7/20/2004	1.9	200390003	H498	Clone	200337501	K	14	1			3	1	8/6/04	17	9.8	0.46	9/9/2004	1	51	32.0	0.65	0.59	
200436914	2	7/20/2004	1.8	200390003	Duroc	Clone	200337501	K	13	1	7/20/2004	OVRL		0						0					
200433901	2	7/15/2004	2.8	200390007	Duroc	Clone	200337602	A	13	1	7/18/2004	WEAK		0						0					
200433902	2	7/15/2004	2.7	200390007	Duroc	Clone	200337602	A	12	1	7/18/2004	WEAK		0						0					
200433903	2	7/15/2004	2.6	200390007	Duroc	Clone	200337602	A	13	1	7/15/2004	WEAK		0						0					
200433904	2	7/15/2004	3.0	200390007	Duroc	Clone	200337602	A	13	1	7/15/2004	WEAK		0						0					
200433905	2	7/15/2004	3.1	200390007	Duroc	Clone	200337602	A	12	1	7/15/2004	WEAK		0						0					
200433906	2	7/15/2004	2.7	200390007	Duroc	Clone	200337602	A	12	1	7/15/2004	WEAK		0						0					
200433907	2	7/15/2004	2.8	200390007	Duroc	Clone	200337602	A	12	1	7/15/2004	UNK		0						0					
200433908	2	7/15/2004	2.8	200390007	Duroc	Clone	200337602	A	14	1	7/15/2004	UNK		0						0					
200433909	1	7/15/2004	2.5	200390007	Duroc	Clone	200337602	A	14	1	7/15/2004	UNK		0						0					
200433910	1	7/15/2004	2.7	200390007	Duroc	Clone	200337602	A	14	1	7/15/2004	UNK		0						0					
200433911	1	7/15/2004	3.2	200390007	Duroc	Clone	200337602	A	13	1	7/15/2004	UNK		0						0					
200433912	1	7/15/2004	3.1	200390007	Duroc	Clone	200337602	A	14	1	7/15/2004	UNK		0						0					
200433913	1	7/15/2004	2.6	200390007	Duroc	Clone	200337602	A	13	1	7/15/2004	UNK		0						0					
200435801	2	7/19/2004	3.9	200390007	Duroc	Clone	200338102	A	13	1			2	1	8/5/04	17	12.1	0.48	9/7/2004	1	50	43.6	0.95	0.79	
200435802	1	7/19/2004	4.5	200390007	Duroc	Clone	200338102	A	13	1			3	1	8/5/04	17	12.8	0.49	9/7/2004	1	50	48.8	1.09	0.89	
200435803	1	7/19/2004	4.1	200390007	Duroc	Clone	200338102	A	13	1			3	1	8/5/04	17	12.6	0.50	9/7/2004	1	50	42.4	0.90	0.77	
200435804	2	7/19/2004	3.2	200390007	Duroc	Clone	200338102	A	13	1	7/19/2004	OVRL		0						0					
200435805	2	7/19/2004	2.7	200390007	Duroc	Clone	200338102	A	14	99	7/19/2004	OVRL		0						0					
200435806	2	7/19/2004	3.2	200390007	Duroc	Clone	200338102	A	13	99	7/19/2004	STLB		0						0					
200435807	1	7/19/2004	3.6	200390007	Duroc	Clone	200338102	A	14	1	7/19/2004	STLB		0						0					
200435808	1	7/19/2004	3.2	200390007	Duroc	Clone	200338102	A	14	1	7/19/2004	STLB		0						0					
200435301	2	7/17/2004	4.1	200190498	H498	Control	200338103	A	14	1			2	1	8/5/04	19	14.1	0.53	9/7/2004	1	52	43.4	0.89	0.76	
200435302	2	7/17/2004	3.8	200190498	H498	Control	200338103	A	15	1			2	1	8/5/04	19	12.5	0.46	9/7/2004	1	52	38.2	0.78	0.66	
200435303	2	7/17/2004	3.6	200190498	H498	Control	200338103	A	14	1			2	1	8/5/04	19	15.1	0.61	9/7/2004	1	52	40.0	0.75	0.70	
200435304	2	7/17/2004	1.9	200190498	H498	Control	200338103	A	15	1			2	1	8/5/04	19	10.4	0.45	9/7/2004	1	52	29.2	0.57	0.53	
200435305	1	7/17/2004	4.1	200190498	H498	Control	200338103	A	14	1			3	1	8/5/04	19	13.3	0.48	9/7/2004	1	52	33.8	0.62	0.57	
200435306	1	7/17/2004	3.7	200190498	H498	Control	200338103	A	14	1			3	1	8/5/04	19	12.1	0.44	9/7/2004	1	52	38.2	0.79	0.66	
200435307	1	7/17/2004	3.8	200190498	H498	Control	200338103	A	16	1			3	1	8/5/04	19	13.2	0.49	9/7/2004	1	52	39.6	0.80	0.69	
200435308	1	7/17/2004	3.8	200190498	H498	Control	200338103	A	14	1			3	1	8/5/04	19	13.1	0.49	9/7/2004	1	52	41.0	0.85	0.72	
200435309	1	7/17/2004	3.4	200190498	H498	Control	200338103	A	15	1			3	1	8/5/04	19	14.2	0.57	9/7/2004	1	52	45.8	0.96	0.82	
200435310	1	7/17/2004	2.9	200190498	H498	Control	200338103	A	14	1			3	1	8/5/04	19	11.9	0.47	9/7/2004	1	52	30.0	0.55	0.52	

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA	
200435311	1	7/17/2004	3.0	200190498	H498	Control	200338103	A	14	1			3		8/5/04	19	11.9	0.47	9/7/2004	1	52	37.4	0.77	0.66	
200435312	2	7/17/2004	2.7	200190498	H498	Control	200338103	A	15	1	7/17/2004	OVRL		0							0				
200435313	2	7/17/2004	1.1	200190498	H498	Control	200338103	A	14	1	7/17/2004	STLB		0							0				
200437401	2	7/22/2004	2.9	200390007	Duroc	Clone	200338203	C	12	1			2	1	8/6/04	15	4	0.07	9/7/2004	1	47	22.0	0.56	0.41	
200437402	2	7/22/2004	3.0	200390007	Duroc	Clone	200338203	C	14	1			2	1	8/6/04	15	9.2	0.41	9/9/2004	1	49	35.0	0.76	0.65	
200437403	1	7/22/2004	3.6	200390007	Duroc	Clone	200338203	C	12	1			3	1	8/6/04	15	10.5	0.46	9/9/2004	1	49	41.0	0.90	0.76	
200437404	1	7/22/2004	3.5	200390007	Duroc	Clone	200338203	C	15	1			3	1	8/6/04	15	8.2	0.31	9/9/2004	1	49	37.0	0.85	0.68	
200437405	1	7/22/2004	3.4	200390007	Duroc	Clone	200338203	C	14	1			3	1	8/6/04	15	10.9	0.50	9/9/2004	1	49	37.8	0.79	0.70	
200437406	1	7/22/2004	3.4	200390007	Duroc	Clone	200338203	C	14	1			3	1	8/6/04	15	11	0.51	9/9/2004	1	49	34.8	0.70	0.64	
200437407	1	7/22/2004	2.9	200390007	Duroc	Clone	200338203	C	14	1			3	1	8/6/04	15	8.9	0.40	9/9/2004	1	49	37.4	0.84	0.70	
200437408	1	7/22/2004	2.9	200390007	Duroc	Clone	200338203	C	14	1			3	1	8/6/04	15	9.3	0.43	9/9/2004	1	49	34.0	0.73	0.63	
200437409	1	7/22/2004	2.9	200390007	Duroc	Clone	200338203	C	13	1			3	1	8/6/04	15	8	0.34	9/9/2004	1	49	30.6	0.66	0.57	
200437410	1	7/22/2004	2.7	200390007	Duroc	Clone	200338203	C	13	1			3	1	8/6/04	15	7.8	0.34	9/9/2004	1	49	32.8	0.74	0.61	
200437411	1	7/22/2004	2.7	200390007	Duroc	Clone	200338203	C	12	1			3	1	8/6/04	15	7.2	0.30	9/9/2004	1	49	29.0	0.64	0.54	
200437412	1	7/22/2004	2.5	200390007	Duroc	Clone	200338203	C	13	1	7/22/2004	STLB		0							0				
200437413	2	7/22/2004	2.2	200390007	Duroc	Clone	200338203	C	15	1	7/22/2004	STLB		0							0				
200430701	2	7/7/2004	3.2	200390002	H498	Clone	200338205	C	12	1			2	1	7/27/04	20	10	0.34	9/1/2004	1	56	33.4	0.65	0.54	
200430702	2	7/7/2004	3.2	200390002	H498	Clone	200338205	C	15	1			2	1	7/27/04	20	13.9	0.54	9/1/2004	1	56	35.4	0.60	0.58	
200430703	2	7/7/2004	3.2	200390002	H498	Clone	200338205	C	14	1			2	1	7/27/04	20	12.5	0.47	9/1/2004	1	56	38.0	0.71	0.62	
200430704	2	7/7/2004	2.9	200390002	H498	Clone	200338205	C	14	1			2	1	7/27/04	20	11.7	0.44	9/1/2004	1	56	37.2	0.71	0.61	
200430705	2	7/7/2004	2.8	200390002	H498	Clone	200338205	C	13	1			2	1	7/27/04	20	9.9	0.36	9/1/2004	1	56	38.6	0.80	0.64	
200430706	1	7/7/2004	4.0	200390002	H498	Clone	200338205	C	15	1			3	1	7/27/04	20	14.4	0.52	9/1/2004	1	56	40.0	0.71	0.64	
200430707	1	7/7/2004	3.4	200390002	H498	Clone	200338205	C	12	1			3	1	7/27/04	20	16	0.63	9/1/2004	1	56	49.8	0.94	0.83	
200430708	1	7/7/2004	3.4	200390002	H498	Clone	200338205	C	14	1			3	1	7/27/04	20	13.4	0.50	9/1/2004	1	56	41.0	0.77	0.67	
200430709	1	7/7/2004	3.6	200390002	H498	Clone	200338205	C	14	1			3	1	7/27/04	20	14.5	0.55	9/1/2004	1	56	45.6	0.86	0.75	
200430710	1	7/7/2004	3.6	200390002	H498	Clone	200338205	C	13	1			3	1	7/27/04	20	14.7	0.56	9/1/2004	1	56	44.4	0.83	0.73	
200430711	1	7/7/2004	3.3	200390002	H498	Clone	200338205	C	15	1			3	1	7/27/04	20	11.2	0.40	9/1/2004	1	56	36.8	0.71	0.60	
200439901	1	7/24/2004	3.4	200395515	Duroc	Control	200338501	G	14	1			3	1	8/12/04	19	14.4	0.58	9/14/2004	1	52	43.6	0.88	0.77	
200439902	1	7/24/2004	3.1	200395515	Duroc	Control	200338501	G	14	1			3	1	8/12/04	19	15.8	0.67	9/14/2004	1	52	40.2	0.74	0.71	
200439903	1	7/24/2004	2.8	200395515	Duroc	Control	200338501	G	12	1			3	1	8/12/04	19	11.1	0.44	9/14/2004	1	52	39.6	0.86	0.71	
200439904	1	7/24/2004	1.9	200395515	Duroc	Control	200338501	G	12	1			3	1	8/12/04	19	10.7	0.46	9/14/2004	1	52	37.0	0.80	0.68	
200439905	1	7/24/2004	3.6	200395515	Duroc	Control	200338501	G	12	1	7/24/2004	STLB		0							0				
200437801	2	7/22/2004	4.1	200390007	Duroc	Clone	200339102	N	14	1			2	1	8/6/04	15	8.9	0.32	9/9/2004	1	49	50.6	1.23	0.95	
200437802	1	7/22/2004	4.8	200390007	Duroc	Clone	200339102	N	13	1			3	1	8/6/04	15	9.5	0.31	9/9/2004	1	49	43.6	1.00	0.79	
200437803	1	7/22/2004	4.2	200390007	Duroc	Clone	200339102	N	14	1			3	1	8/6/04	15	8.9	0.31	9/9/2004	1	49	43.4	1.01	0.80	
200437804	1	7/22/2004	3.6	200390007	Duroc	Clone	200339102	N	13	1			3	1	8/6/04	15	8.1	0.30	9/9/2004	1	49	39.8	0.93	0.74	
200437805	1	7/22/2004	4.0	200390007	Duroc	Clone	200339102	N	13	1	7/22/2004	STLB		0							0				
200436401	2	7/19/2004	4.7	200390005	H498	Clone	200339104	N	14	1			2	1	8/5/04	17	13	0.49	9/9/2004	1	52	35.4	0.64	0.59	
200436402	1	7/19/2004	4.3	200390005	H498	Clone	200339104	N	14	1			3	1	8/5/04	17	12.5	0.48	9/9/2004	1	52	45.2	0.93	0.79	
200436403	1	7/19/2004	4.2	200390005	H498	Clone	200339104	N	14	1			3	1	8/5/04	17	11.2	0.41	9/9/2004	1	52	43.4	0.92	0.75	
200436404	1	7/19/2004	3.9	200390005	H498	Clone	200339104	N	14	1			3	1	8/5/04	17	12.4	0.50	9/9/2004	1	52	39.0	0.76	0.68	
200436405	1	7/19/2004	3.5	200390005	H498	Clone	200339104	N	16	1			3	1	8/5/04	17	11	0.44	9/9/2004	1	52	39.0	0.80	0.68	
200438701	2	7/23/2004	3.7	200395515	Duroc	Control	200339105	N	14	1			2	1	8/12/04	20	13.2	0.48	9/14/2004	1	53	44.8	0.96	0.78	
200438702	2	7/23/2004	3.5	200395515	Duroc	Control	200339105	N	16	1			2	1	8/12/04	20	12.1	0.43	9/14/2004	1	53	43.0	0.94	0.75	
200438703	2	7/23/2004	2.8	200395515	Duroc	Control	200339105	N	16	1			2	1	8/12/04	20	11.5	0.44	9/14/2004	1	53	41.8	0.92	0.74	
200438704	2	7/23/2004	2.9	200395515	Duroc	Control	200339105	N	14	1			2	1	8/12/04	20	11.7	0.44	9/14/2004	1	53	40.6	0.88	0.71	
200438705	2	7/23/2004	2.7	200395515	Duroc	Control	200339105	N	14	1			2	1	8/12/04	20	13.4	0.54	9/14/2004	1	53	48.8	1.07	0.87	
200438706	1	7/23/2004	3.9	200395515	Duroc	Control	200339105	N	12	1			3	1	8/12/04	20	12.5	0.43	9/14/2004	1	53	38.0	0.77	0.64	
200438707	1	7/23/2004	3.3	200395515	Duroc	Control	200339105	N	16	1			3	1	8/12/04	20	12.3	0.45	9/14/2004	1	53	43.2	0.94	0.75	
200438708	1	7/23/2004	3.3	200395515	Duroc	Control	200339105	N	16	1			0								0				
200438709	1	7/23/2004	2.8	200395515	Duroc	Control	200339105	N	13	1			3	1	8/12/04	20	9.8	0.35	9/14/2004	1	53	37.2	0.83	0.65	

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA	
200438710	2	7/23/2004	2.6	200395515	Duroc	Control	200339105	N	14	1	7/23/2004	OVRL		0							0				
200438711	1	7/23/2004	3.2	200395515	Duroc	Control	200339105	N	14	1	7/23/2004	STLB		0								0			
200438712	1	7/23/2004	3.5	200395515	Duroc	Control	200339105	N	13	1	7/23/2004	STLB		0								0			
200438714	2	7/23/2004	2.8	200395515	Duroc	Control	200339105	N	14	1	7/23/2004	STLB		0								0			
200432701	2	7/13/2004	4.7	200390003	H498	Clone	200339303	D	12	1			2	1	7/30/04	17	12.7	0.47	9/2/2004	1	51	42.4	0.87	0.74	
200432702	2	7/13/2004	4.4	200390003	H498	Clone	200339303	D	13	1			2	1	7/30/04	17	12.4	0.47	9/2/2004	1	51	37.6	0.74	0.65	
200432703	1	7/13/2004	4.9	200390003	H498	Clone	200339303	D	14	1			3	1	7/30/04	17	13.9	0.53	9/2/2004	1	51	37.3	0.69	0.64	
200432704	1	7/13/2004	4.7	200390003	H498	Clone	200339303	D	14	1			3	1	7/30/04	17	12	0.43	9/2/2004	1	51	31.8	0.58	0.53	
200432705	1	7/13/2004	4.4	200390003	H498	Clone	200339303	D	13	1			3	1	7/30/04	17	12.4	0.47	9/2/2004	1	51	37.2	0.73	0.64	
200432706	1	7/13/2004	3.9	200390003	H498	Clone	200339303	D	13	1			3	1	7/30/04	17	11	0.42	9/2/2004	1	51	38.8	0.82	0.68	
200432707	1	7/13/2004	3.7	200390003	H498	Clone	200339303	D	14	1	7/13/2004	OVRL		0							0				
200432708	2	7/13/2004	3.5	200390003	H498	Clone	200339303	D	13	1	7/13/2004	OVRL		0							0				
200432709	2	7/13/2004	3.1	200390003	H498	Clone	200339303	D	14	1	7/13/2004	STLB		0							0				
200432710	1	7/13/2004	4.0	200390003	H498	Clone	200339303	D	14	1	7/13/2004	STLB		0							0				
200434801	2	7/17/2004	3.9	200395515	Duroc	Clone	200339403	H	16	1			2	1	8/5/04	19	14.1	0.54	9/7/2004	1	52	45.0	0.94	0.79	
200434802	2	7/17/2004	3.8	200395515	Duroc	Control	200339403	H	12	1			2	1	8/5/04	19	14.1	0.54	9/7/2004	1	52	36.6	0.68	0.63	
200434803	2	7/17/2004	3.2	200395515	Duroc	Control	200339403	H	15	1			2	1	8/5/04	19	14.1	0.57	9/7/2004	1	52	42.2	0.85	0.75	
200434804	2	7/17/2004	2.8	200395515	Duroc	Control	200339403	H	14	1			2	1	8/5/04	19	12.6	0.52	9/7/2004	1	52	38.8	0.79	0.69	
200434805	2	7/17/2004	2.5	200395515	Duroc	Control	200339403	H	14	1			2	1	8/5/04	19	10.5	0.42	9/7/2004	1	52	31.0	0.62	0.55	
200434806	1	7/17/2004	4.2	200395515	Duroc	Control	200339403	H	13	1			3	1	8/5/04	19	13.7	0.50	9/7/2004	1	52	40.8	0.82	0.70	
200434807	1	7/17/2004	3.8	200395515	Duroc	Control	200339403	H	13	1			3	1	8/5/04	19	12.8	0.47	9/7/2004	1	52	43.6	0.93	0.77	
200434808	1	7/17/2004	3.5	200395515	Duroc	Control	200339403	H	16	1			3	1	8/5/04	19	13.6	0.53	9/7/2004	1	52	44.0	0.92	0.78	
200434809	1	7/17/2004	3.3	200395515	Duroc	Control	200339403	H	14	1			3	1	8/5/04	19	11	0.41	9/7/2004	1	52	37.4	0.80	0.66	
200434810	2	7/17/2004	3.5	200395515	Duroc	Control	200339403	H	14	1	7/17/2004	OVRL		0							0				
200438401	2	7/23/2004	3.9	200398128	Duroc	Control	200339603	N	14	1			2	1	8/6/04	14	6.9	0.21	9/9/2004	1	48	35.6	0.84	0.66	
200438402	2	7/23/2004	3.9	200398128	Duroc	Control	200339603	N	13	1			2	1	8/6/04	14	4.2	0.02	9/7/2004	1	46	32.4	0.88	0.62	
200438403	2	7/23/2004	3.5	200398128	Duroc	Control	200339603	N	14	1			2	1	8/6/04	14	10.1	0.47	9/9/2004	1	48	32.8	0.67	0.61	
200438404	1	7/23/2004	4.4	200398128	Duroc	Control	200339603	N	14	1			3	1	8/12/04	20	13	0.43	9/14/2004	1	53	48.8	1.08	0.84	
200438405	1	7/23/2004	3.4	200398128	Duroc	Control	200339603	N	12	6			3	1	8/12/04	20	12.6	0.46	9/14/2004	1	53	38.6	0.79	0.66	
200438406	1	7/23/2004	3.5	200398128	Duroc	Control	200339603	N	13	1			3	1	8/6/04	14	11.2	0.55	9/9/2004	1	48	37.0	0.76	0.70	
200438407	1	7/23/2004	3.7	200398128	Duroc	Control	200339603	N	13	1			3	1	8/12/04	20	13.4	0.49	9/14/2004	1	53	49.0	1.08	0.85	
200438408	1	7/23/2004	3.7	200398128	Duroc	Control	200339603	N	14	1			3	1	8/12/04	20	12.6	0.45	9/14/2004	1	53	42.6	0.91	0.73	
200438409	1	7/23/2004	3.4	200398128	Duroc	Control	200339603	N	14	1			3	1	8/12/04	20	14	0.53	9/14/2004	1	53	47.4	1.01	0.83	
200438410	1	7/23/2004	3.3	200398128	Duroc	Control	200339603	N	11	1			3	1	8/12/04	20	11	0.39	9/14/2004	1	53	35.6	0.75	0.61	
200438411	1	7/23/2004	3.4	200398128	Duroc	Control	200339603	N	14	1			3	1	8/12/04	20	10.8	0.37	9/14/2004	1	53	39.0	0.85	0.67	
200438412	1	7/23/2004	3.5	200398128	Duroc	Control	200339603	N	13	1			3	1	8/12/04	20	10.4	0.35	9/14/2004	1	53	38.0	0.84	0.65	
200438413	1	7/23/2004	3.5	200398128	Duroc	Control	200339603	N	14	1			3	1	8/12/04	20	8.4	0.25	9/14/2004	1	53	35.8	0.83	0.61	
200438414	1	7/23/2004	3.0	200398128	Duroc	Control	200339603	N	12	1			3	1	8/12/04	20	9.9	0.35	9/14/2004	1	53	37.8	0.85	0.66	
200438101	2	7/23/2004	3.8	200390007	Duroc	Clone	200339704	J	14	1			2	1	8/6/04	14	10.4	0.47	9/9/2004	1	48	39.0	0.84	0.73	
200438102	2	7/23/2004	3.5	200390007	Duroc	Clone	200339704	J	12	1			2	1	8/6/04	14	10.9	0.53	9/9/2004	1	48	40.8	0.88	0.78	
200438103	2	7/23/2004	3.5	200390007	Duroc	Clone	200339704	J	12	6			2	1	8/6/04	14	8.8	0.38	9/9/2004	1	48	37.4	0.84	0.71	
200438104	2	7/23/2004	3.4	200390007	Duroc	Clone	200339704	J	14	1			2	1	8/6/04	14	11.2	0.56	9/9/2004	1	48	39.2	0.82	0.75	
200438105	2	7/23/2004	3.3	200390007	Duroc	Clone	200339704	J	14	1			2	1	8/6/04	14	9.3	0.43	9/9/2004	1	48	37.8	0.84	0.72	
200438106	1	7/23/2004	4.1	200390007	Duroc	Clone	200339704	J	12	1			3	1	8/6/04	14	11.1	0.50	9/9/2004	1	48	44.6	0.99	0.84	
200438107	1	7/23/2004	3.6	200390007	Duroc	Clone	200339704	J	13	1			3	1	8/6/04	14	10.9	0.52	9/9/2004	1	48	37.0	0.77	0.70	
200438108	1	7/23/2004	3.6	200390007	Duroc	Clone	200339704	J	16	1	7/23/2004	STLB		0							0				
200438109	2	7/23/2004	3.4	200390007	Duroc	Clone	200339704	J	12	1	7/23/2004	STLB		0							0				
200438110	2	7/23/2004	2.9	200390007	Duroc	Clone	200339704	J	12	1	7/23/2004	STLB		0							0				
200436301	2	7/19/2004	3.7	200395515	Duroc	Control	200340001	A	14	1			2	1	8/5/04	17	10.5	0.40	9/9/2004	1	52	32.4	0.63	0.55	
200436302	2	7/19/2004	3.3	200395515	Duroc	Control	200340001	A	14	1			2	1	8/5/04	17	9.2	0.35	9/9/2004	1	52	37.6	0.81	0.66	
200436303	2	7/19/2004	3.3	200395515	Duroc	Control	200340001	A	14	1			2	1	8/5/04	17	11.7	0.49	9/9/2004	1	52	43.6	0.91	0.78	

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA	
200436306	2	7/19/2004	2.8	200395515	Duroc	Control	200340001	A	12	1			2	1	8/5/04	17	10.3	0.44	9/9/2004	1	52	37.4	0.77	0.67	
200436307	2	7/19/2004	2.7	200395515	Duroc	Control	200340001	A	15	1			2	1	8/5/04	17	10	0.43	9/9/2004	1	52	40.2	0.86	0.72	
200436308	2	7/19/2004	2.5	200395515	Duroc	Control	200340001	A	12	1				0							0				
200436309	1	7/19/2004	3.2	200395515	Duroc	Control	200340001	A	14	1			3	1	8/5/04	17	11.8	0.51	9/9/2004	1	52	38.8	0.77	0.68	
200436310	1	7/19/2004	2.4	200395515	Duroc	Control	200340001	A	13	1			3	1	8/5/04	17	9.7	0.43	9/9/2004	1	52	35.6	0.74	0.64	
200436311	1	7/19/2004	2.3	200395515	Duroc	Control	200340001	A	14	1			3	1	8/5/04	17	8.1	0.34	9/9/2004	1	52	33.2	0.72	0.59	
200436312	1	7/19/2004	2.3	200395515	Duroc	Control	200340001	A	14	1			3	1	8/5/04	17	9.2	0.41	9/9/2004	1	52	27.2	0.51	0.48	
200436313	2	7/19/2004	3.4	200395515	Duroc	Control	200340001	A	13	1	7/19/2004	OVRL		0							0				
200434301	2	7/15/2004	3.7	200395515	Duroc	Control	200340204	M	14	1			2	1	7/30/04	15	11.1	0.49	9/2/2004	1	49	41.2	0.89	0.77	
200434302	2	7/15/2004	3.1	200395515	Duroc	Control	200340204	M	14	1			2	1	7/30/04	15	9.1	0.40	9/2/2004	1	49	31.6	0.66	0.58	
200434303	2	7/15/2004	3.0	200395515	Duroc	Control	200340204	M	13	1			2	1	7/30/04	15	9.7	0.45	9/2/2004	1	49	30.4	0.61	0.56	
200434304	1	7/15/2004	3.6	200395515	Duroc	Control	200340204	M	13	1			3	1	7/30/04	15	8	0.29	9/2/2004	1	49	29.8	0.64	0.53	
200434305	1	7/15/2004	4.0	200395515	Duroc	Control	200340204	M	13	1			3	1	7/30/04	15	12.4	0.56	9/2/2004	1	49	41.4	0.85	0.76	
200434306	1	7/15/2004	3.5	200395515	Duroc	Control	200340204	M	12	1			3	1	7/30/04	15	9.9	0.43	9/2/2004	1	49	32.0	0.65	0.58	
200434307	1	7/15/2004	2.9	200395515	Duroc	Control	200340204	M	14	1	8/13/2004	SEPT	3	1	7/30/04	15	10.3	0.49		0					
200434308	1	7/15/2004	3.2	200395515	Duroc	Control	200340204	M	13	1			3	1	7/30/04	15	9	0.39	9/2/2004	1	49	30.2	0.62	0.55	
200434309	1	7/15/2004	3.0	200395515	Duroc	Control	200340204	M	13	1			3	1	7/30/04	15	10.7	0.51	9/2/2004	1	49	28.6	0.53	0.52	
200434310	1	7/15/2004	2.6	200395515	Duroc	Control	200340204	M	12	1			3	1	7/30/04	15	8.4	0.39	9/2/2004	1	49	28.4	0.59	0.53	
200434311	2	7/15/2004	3.0	200395515	Duroc	Control	200340204	M	13	1	7/15/2004	STLB		0							0				
200436601	2	7/20/2004	3.8	200390007	Duroc	Clone	200340206	M	14	1			2	1	8/5/04	16	12	0.51	9/9/2004	1	51	45.4	0.95	0.82	
200436602	2	7/20/2004	3.3	200390007	Duroc	Clone	200340206	M	15	1			2	1	8/5/04	16	11.2	0.49	9/9/2004	1	51	46.4	1.01	0.85	
200436603	2	7/20/2004	3.0	200390007	Duroc	Clone	200340206	M	14	1			2	1	8/6/04	17	11.8	0.52	9/9/2004	1	51	42.8	0.91	0.78	
200436604	2	7/20/2004	2.9	200390007	Duroc	Clone	200340206	M	13	1			2	1	8/6/04	17	11.5	0.51	9/9/2004	1	51	38.8	0.80	0.70	
200436605	1	7/20/2004	4.5	200390007	Duroc	Clone	200340206	M	14	1			3	1	8/6/04	17	12.2	0.45	9/9/2004	1	51	41.6	0.86	0.73	
200436606	1	7/20/2004	4.0	200390007	Duroc	Clone	200340206	M	12	1			3	1	8/6/04	17	13.1	0.54	9/9/2004	1	51	46.6	0.99	0.84	
200436607	1	7/20/2004	3.8	200390007	Duroc	Clone	200340206	M	15	1			3	1	8/6/04	17	13.1	0.55	9/9/2004	1	51	41.4	0.83	0.74	
200436608	1	7/20/2004	4.1	200390007	Duroc	Clone	200340206	M	14	1			3	1	8/6/04	17	14.5	0.61	9/9/2004	1	51	48.4	1.00	0.87	
200436609	1	7/20/2004	4.0	200390007	Duroc	Clone	200340206	M	15	1			3	1	8/6/04	17	13.8	0.58	9/9/2004	1	51	47.6	0.99	0.85	
200436610	1	7/20/2004	3.5	200390007	Duroc	Clone	200340206	M	14	1			3	1	8/6/04	17	12.4	0.52	9/9/2004	1	51	42.8	0.89	0.77	
200436611	1	7/20/2004	3.5	200390007	Duroc	Clone	200340206	M	13	1			3	1	8/6/04	17	11	0.44	9/9/2004	1	51	40.0	0.85	0.72	
200436612	1	7/20/2004	3.1	200390007	Duroc	Clone	200340206	M	14	1			3	1	8/6/04	17	11.1	0.47	9/9/2004	1	51	45.8	1.02	0.84	
200437301	2	7/22/2004	4.1	200395515	Duroc	Control	200340702	J	14	1			2	1	8/6/04	15	9.3	0.35	9/9/2004	1	49	43.4	1.00	0.80	
200437302	2	7/22/2004	4.0	200395515	Duroc	Control	200340702	J	15	1			2	1	8/6/04	15	8.9	0.33	9/9/2004	1	49	33.0	0.71	0.59	
200437303	2	7/22/2004	4.1	200395515	Duroc	Control	200340702	J	15	1			2	1	8/6/04	15	10.3	0.41	9/9/2004	1	49	41.4	0.91	0.76	
200437304	1	7/22/2004	4.6	200395515	Duroc	Control	200340702	J	14	1			3	1	8/6/04	15	11.4	0.45	9/9/2004	1	49	40.4	0.85	0.73	
200437305	1	7/22/2004	3.9	200395515	Duroc	Control	200340702	J	14	1			3	1	8/6/04	15	9.3	0.36	9/9/2004	1	49	39.2	0.88	0.72	
200437306	1	7/22/2004	4.0	200395515	Duroc	Control	200340702	J	15	1			3	1	8/6/04	15	11	0.47	9/9/2004	1	49	47.8	1.08	0.89	
200437307	1	7/22/2004	4.1	200395515	Duroc	Control	200340702	J	16	1			3	1	8/6/04	15	11.4	0.49	9/9/2004	1	49	41.6	0.89	0.77	
200437308	1	7/22/2004	3.8	200395515	Duroc	Control	200340702	J	15	1			3	1	8/6/04	15	8.8	0.33	9/9/2004	1	49	35.4	0.78	0.64	
200437309	1	7/22/2004	3.7	200395515	Duroc	Control	200340702	J	15	1			3	1	8/6/04	15	8.7	0.33	9/9/2004	1	49	39.4	0.90	0.73	
200437310	2	7/22/2004	3.4	200395515	Duroc	Control	200340702	J	14	1	7/22/2004	OVRL		0							0				
200440601	2	7/25/2004	3.8	200398128	Duroc	Control	200340901	F	14	1			2	1	8/13/04	19	14.3	0.55	9/15/2004	1	52	50.4	1.09	0.90	
200440602	2	7/25/2004	3.3	200398128	Duroc	Control	200340901	F	14	1			2	1	8/13/04	19	13.6	0.54	9/15/2004	1	52	50.4	1.12	0.91	
200440603	1	7/25/2004	4.0	200398128	Duroc	Control	200340901	F	12	1			3	1	8/13/04	19	14.2	0.54	9/15/2004	1	52	41.9	0.84	0.73	
200440604	1	7/25/2004	3.9	200398128	Duroc	Control	200340901	F	14	1			3	1	8/13/04	19	14.2	0.54	9/15/2004	1	52	47.8	1.02	0.84	
200440605	1	7/25/2004	3.7	200398128	Duroc	Control	200340901	F	17	6			3	1	8/13/04	19	11.6	0.42	9/15/2004	1	52	48.8	1.13	0.87	
200440606	1	7/25/2004	3.5	200398128	Duroc	Control	200340901	F	15	1			3	1	8/13/04	19	12.9	0.49	9/15/2004	1	52	41.8	0.88	0.74	
200440607	1	7/25/2004	3.7	200398128	Duroc	Control	200340901	F	15	6			3	1	8/13/04	19	13.3	0.51	9/15/2004	1	52	48.3	1.06	0.86	
200440608	1	7/25/2004	3.4	200398128	Duroc	Control	200340901	F	15	1			3	1	8/13/04	19	11.9	0.45	9/15/2004	1	52	25.2	0.40	0.42	
200440609	1	7/25/2004	2.9	200398128	Duroc	Control	200340901	F	15	1			3	1	8/13/04	19	10.8	0.42	9/15/2004	1	52	39.1	0.86	0.70	
200440610	1	7/25/2004	3.1	200398128	Duroc	Control	200340901	F	14	1	7/25/2004	STLB		0							0				

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA	
200433301	2	7/13/2004	2.7	200190498	H498	Control	200340902	F	14	1			2	1	7/30/04	17	10.3	0.45	9/2/2004	1	51	33.4	0.68	0.60	
200433302	2	7/13/2004	2.7	200190498	H498	Control	200340902	F	14	1			2	1	7/30/04	17	6.8	0.24	9/2/2004	1	51	23.4	0.49	0.41	
200433303	2	7/13/2004	2.6	200190498	H498	Control	200340902	F	14	1			2	1	7/30/04	17	6.8	0.25	9/2/2004	1	51	22.0	0.45	0.38	
200433304	2	7/13/2004	2.3	200190498	H498	Control	200340902	F	14	1			2	1	7/30/04	17	3.2	0.05	9/1/2004	1	50	18.8	0.47	0.33	
200433305	2	7/13/2004	1.9	200190498	H498	Control	200340902	F	14	1			2	1	7/30/04	17	6.6	0.28	9/2/2004	1	51	19.8	0.39	0.35	
200433306	1	7/13/2004	3.1	200190498	H498	Control	200340902	F	14	1			3	1	7/30/04	17	9.8	0.39	9/2/2004	1	51	35.2	0.75	0.63	
200433307	1	7/13/2004	2.8	200190498	H498	Control	200340902	F	14	1			3	1	7/30/04	17	9.3	0.38	9/2/2004	1	51	34.6	0.74	0.62	
200433308	2	7/13/2004	3.3	200190498	H498	Control	200340902	F	15	1	7/13/2004	OVRL		0							0				
200433309	1	7/13/2004	3.3	200190498	H498	Control	200340902	F	14	1	7/13/2004	OVRL		0							0				
200433310	1	7/13/2004	3.5	200190498	H498	Control	200340902	F	15	1	7/13/2004	OVRL		0							0				
200433311	1	7/13/2004	0.9	200190498	H498	Control	200340902	F	13	1	7/13/2004	OVRL		0							0				
200433312	1	7/13/2004	2.5	200190498	H498	Control	200340902	F	14	1	7/13/2004	STLB		0							0				
200431501	2	7/10/2004	4.0	200390002	H498	Clone	200340903	F	14	1			2	1	7/27/04	17	10.6	0.39	9/1/2004	1	53	38.4	0.77	0.65	
200431502	2	7/10/2004	3.7	200390002	H498	Clone	200340903	F	15	1			2	1	7/27/04	17	13.4	0.57	9/1/2004	1	53	46.2	0.91	0.80	
200431503	2	7/10/2004	3.3	200390002	H498	Clone	200340903	F	13	1			2	1	7/27/04	17	14	0.63	9/1/2004	1	53	42.0	0.78	0.73	
200431504	1	7/10/2004	3.9	200390002	H498	Clone	200340903	F	14	1			3	1	7/27/04	17	13.7	0.58	9/1/2004	1	53	46.2	0.90	0.80	
200431505	1	7/10/2004	3.8	200390002	H498	Clone	200340903	F	14	1			8	1	7/27/04	17	8.7	0.29	9/1/2004	1	53	41.0	0.90	0.70	
200431506	1	7/10/2004	3.8	200390002	H498	Clone	200340903	F	15	1			3	1	7/27/04	17	14.6	0.64	9/1/2004	1	53	44.0	0.82	0.76	
200430901	2	7/8/2004	3.9	200390003	H498	Clone	200340904	F	14	1				0							0				
200430902	1	7/8/2004	4.0	200390003	H498	Clone	200340904	F	14	1			3	1	7/27/04	19	11.8	0.41	9/1/2004	1	55	44.2	0.90	0.73	
200430903	1	7/8/2004	3.7	200390003	H498	Clone	200340904	F	13	1			3	1	7/27/04	19	12.7	0.47	9/1/2004	1	55	43.6	0.86	0.73	
200430904	1	7/8/2004	3.6	200390003	H498	Clone	200340904	F	14	1				0							0				
200430905	1	7/8/2004	3.3	200390003	H498	Clone	200340904	F	14	1			3	1	7/27/04	19	11.1	0.41	9/1/2004	1	55	48.8	1.05	0.83	
200430906	1	7/8/2004	3.1	200390003	H498	Clone	200340904	F	14	1			3	1	7/27/04	19	11.7	0.45	9/1/2004	1	55	47.6	1.00	0.81	
200430907	1	7/8/2004	4.0	200390003	H498	Clone	200340904	F	14	1	7/9/2004	WEAK		0							0				
200430908	1	7/8/2004	3.7	200390003	H498	Clone	200340904	F	14	1	7/8/2004	STLB		0							0				
200430909	1	7/8/2004	4.4	200390003	H498	Clone	200340904	F	16	1	7/8/2004	STLB		0							0				
200430910	1	7/8/2004	3.5	200390003	H498	Clone	200340904	F	14	1	7/8/2004	STLB		0							0				
200430911	2	7/8/2004	4.0	200390003	H498	Clone	200340904	F	16	1	7/8/2004	STLB		0							0				
200430912	1	7/8/2004	4.1	200390003	H498	Clone	200340904	F	14	1	7/8/2004	STLB		0							0				
200438601	2	7/23/2004	3.3	200390007	Duroc	Clone	200340906	F	14	1			2	1	8/12/04	20	14.4	0.56	9/14/2004	1	53	45.0	0.93	0.79	
200438602	2	7/23/2004	3.1	200390007	Duroc	Clone	200340906	F	14	1			2	1	8/12/04	20	11.7	0.43	9/14/2004	1	53	41.4	0.90	0.72	
200438603	2	7/23/2004	3.1	200390007	Duroc	Clone	200340906	F	15	1			2	1	8/12/04	20	11.6	0.43	9/14/2004	1	53	40.0	0.86	0.70	
200438604	2	7/23/2004	2.8	200390007	Duroc	Clone	200340906	F	14	1			2	1	8/12/04	20	11.3	0.43	9/14/2004	1	53	41.4	0.91	0.73	
200438605	2	7/23/2004	2.7	200390007	Duroc	Clone	200340906	F	15	1			2	1	8/12/04	20	12.3	0.48	9/14/2004	1	53	42.4	0.91	0.75	
200438606	2	7/23/2004	2.6	200390007	Duroc	Clone	200340906	F	14	1			2	1	8/12/04	20	9.6	0.35	9/14/2004	1	53	36.6	0.82	0.64	
200438607	1	7/23/2004	3.5	200390007	Duroc	Clone	200340906	F	14	1			3	1	8/12/04	20	12	0.43	9/14/2004	1	53	42.0	0.91	0.73	
200438608	1	7/23/2004	3.3	200390007	Duroc	Clone	200340906	F	14	1			3	1	8/12/04	20	11.3	0.40	9/14/2004	1	53	42.2	0.94	0.73	
200438609	1	7/23/2004	2.6	200390007	Duroc	Clone	200340906	F	14	1			3	1	8/12/04	20	11.6	0.45	9/14/2004	1	53	39.2	0.84	0.69	
200438610	1	7/23/2004	2.5	200390007	Duroc	Clone	200340906	F	14	1			3	1	8/12/04	20	10.2	0.39	9/14/2004	1	53	28.8	0.56	0.50	
200438611	1	7/23/2004	2.3	200390007	Duroc	Clone	200340906	F	14	1			3	1	8/12/04	20	9.5	0.36	9/14/2004	1	53	36.8	0.83	0.65	
200438612	1	7/23/2004	3.5	200390007	Duroc	Clone	200340906	F	12	1	7/23/2004	STLB		0							0				
200438613	1	7/23/2004	3.3	200390007	Duroc	Clone	200340906	F	12	1	7/23/2004	STLB		0							0				
200438614	2	7/23/2004	2.7	200390007	Duroc	Clone	200340906	F	14	1	7/23/2004	STLB		0							0				
200433601	2	7/13/2004	4.6	200390003	H498	Clone	200341201	M	14	1			2	1	7/30/04	17	16.9	0.72	9/2/2004	1	51	45.0	0.83	0.79	
200433602	2	7/13/2004	4.0	200390003	H498	Clone	200341201	M	14	1			2	1	7/30/04	17	13.3	0.55	9/2/2004	1	51	44.0	0.90	0.78	
200433603	2	7/13/2004	3.5	200390003	H498	Clone	200341201	M	12	1			2	1	7/30/04	17	12.5	0.53	9/2/2004	1	51	41.8	0.86	0.75	
200433604	2	7/13/2004	3.3	200390003	H498	Clone	200341201	M	14	1			2	1	7/30/04	17	12.4	0.54	9/2/2004	1	51	36.8	0.72	0.66	
200433605	1	7/13/2004	4.8	200390003	H498	Clone	200341201	M	14	1			3	1	7/30/04	17	15.5	0.63	9/2/2004	1	51	51.0	1.04	0.91	
200433606	1	7/13/2004	4.2	200390003	H498	Clone	200341201	M	14	1	7/13/2004	OVRL		0							0				
200433607	2	7/13/2004	3.7	200390003	H498	Clone	200341201	M	12	1	7/13/2004	OVRL		0							0				

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA	
200433608	2	7/13/2004	3.2	200390003	H498	Clone	200341201	M	14	1	7/13/2004	OVRL		0							0				
200433609	2	7/13/2004	3.1	200390003	H498	Clone	200341201	M	15	1	7/13/2004	STLB		0								0			
200433001	2	7/13/2004	3.1	200390003	H498	Clone	200341501	H	14	1				2	1	7/30/04	17	12.9	0.58	9/2/2004	1	51	34.4	0.63	0.61
200433002	2	7/13/2004	3.1	200390003	H498	Clone	200341501	H	14	1				2	1	7/30/04	17	11	0.46	9/2/2004	1	51	32.4	0.63	0.57
200433003	2	7/13/2004	2.7	200390003	H498	Clone	200341501	H	14	1				2	1	7/30/04	17	8.7	0.35	9/2/2004	1	51	29.6	0.61	0.53
200433004	1	7/13/2004	3.3	200390003	H498	Clone	200341501	H	14	1				3	1	7/30/04	17	9.2	0.35	9/2/2004	1	51	34.6	0.75	0.61
200433005	1	7/13/2004	3.1	200390003	H498	Clone	200341501	H	15	1				3	1	7/30/04	17	9.4	0.37	9/2/2004	1	51	33.4	0.71	0.59
200433006	1	7/13/2004	3.1	200390003	H498	Clone	200341501	H	12	1				3	1	7/30/04	17	9.4	0.37	9/2/2004	1	51	28.8	0.57	0.50
200433007	1	7/13/2004	2.9	200390003	H498	Clone	200341501	H	12	1				2	1	7/30/04	17	9.4	0.38	9/2/2004	1	51	31.4	0.65	0.56
200433008	1	7/13/2004	3.2	200390003	H498	Clone	200341501	H	15	1				3	1	7/30/04	17	9	0.34	9/2/2004	1	51	29.0	0.59	0.51
200433009	1	7/13/2004	2.8	200390003	H498	Clone	200341501	H	14	1				0							0				
200433010	1	7/13/2004	2.0	200390003	H498	Clone	200341501	H	12	1				3	1	7/30/04	17	5.8	0.22	9/1/2004	1	50	24.6	0.57	0.45
200433011	1	7/13/2004	1.7	200390003	H498	Clone	200341501	H	16	1				3	1	7/30/04	17	6.6	0.29	9/1/2004	1	50	18.8	0.37	0.34
200433012	2	7/13/2004	2.9	200390003	H498	Clone	200341501	H	15	1	7/13/2004	OVRL		0							0				
200433013	1	7/13/2004	2.9	200390003	H498	Clone	200341501	H	13	1	7/13/2004	OVRL		0							0				
200433014	1	7/13/2004	2.3	200390003	H498	Clone	200341501	H	14	1	7/13/2004	OVRL		0							0				
200433015	1	7/13/2004	2.0	200390003	H498	Clone	200341501	H	13	1	7/13/2004	OVRL		0							0				
200440901	2	7/26/2004	3.2	200398128	Duroc	Control	200341702	K	14	1				2	1	8/13/04	18	9.6	0.36	9/15/2004	1	51	37.7	0.85	0.68
200440902	2	7/26/2004	2.9	200398128	Duroc	Control	200341702	K	14	1				2	1	8/13/04	18	10.4	0.42	9/15/2004	1	51	41.5	0.94	0.76
200440903	2	7/26/2004	2.9	200398128	Duroc	Control	200341702	K	12	1				2	1	8/13/04	18	9.6	0.37	9/15/2004	1	51	34.8	0.76	0.63
200440904	2	7/26/2004	2.7	200398128	Duroc	Control	200341702	K	14	1				0							0				
200440905	2	7/26/2004	2.3	200398128	Duroc	Control	200341702	K	12	1				2	1	8/13/04	18	5.8	0.19	9/15/2004	1	51	27.7	0.66	0.50
200440906	2	7/26/2004	2.3	200398128	Duroc	Control	200341702	K	13	1				2	1	8/13/04	18	9.7	0.41	9/15/2004	1	51	39.2	0.89	0.72
200440907	1	7/26/2004	3.2	200398128	Duroc	Control	200341702	K	14	1				3	1	8/13/04	18	10.5	0.41	9/15/2004	1	51	37.4	0.82	0.67
200440908	1	7/26/2004	3.0	200398128	Duroc	Control	200341702	K	15	1				3	1	8/13/04	18	10.1	0.39	9/15/2004	1	51	38.5	0.86	0.70
200440909	1	7/26/2004	2.8	200398128	Duroc	Control	200341702	K	14	1				3	1	8/13/04	18	9.8	0.39	9/15/2004	1	51	36.6	0.81	0.66
200440910	1	7/26/2004	2.8	200398128	Duroc	Control	200341702	K	13	1				3	1	8/13/04	18	9.7	0.38	9/15/2004	1	51	38.6	0.88	0.70
200440911	1	7/26/2004	2.6	200398128	Duroc	Control	200341702	K	14	1				3	1	8/13/04	18	8.1	0.31	9/15/2004	1	51	29.3	0.64	0.52
200440912	1	7/26/2004	2.5	200398128	Duroc	Control	200341702	K	15	1				3	1	8/13/04	18	8.7	0.34	9/15/2004	1	51	31.7	0.70	0.57
200440913	1	7/26/2004	1.8	200398128	Duroc	Control	200341702	K	15	1				3	1	8/13/04	18	8.4	0.37	9/15/2004	1	51	36.0	0.84	0.67
200432101	2	7/12/2004	4.2	200390003	H498	Clone	200342004	A	16	1				2	1	7/27/04	15	12.3	0.54	9/1/2004	1	51	38.4	0.73	0.67
200432102	2	7/12/2004	3.9	200390003	H498	Clone	200342004	A	14	1				2	1	7/27/04	15	11.9	0.53	9/1/2004	1	51	32.4	0.57	0.56
200432103	2	7/12/2004	3.6	200390003	H498	Clone	200342004	A	15	1				2	1	7/27/04	15	10.1	0.43	9/1/2004	1	51	35.2	0.70	0.62
200432104	1	7/12/2004	4.3	200390003	H498	Clone	200342004	A	14	1				3	1	7/27/04	15	12.3	0.53	9/1/2004	1	51	34.4	0.61	0.59
200432105	1	7/12/2004	3.9	200390003	H498	Clone	200342004	A	15	1				3	1	7/27/04	15	11.7	0.52	9/1/2004	1	51	34.8	0.64	0.61
200432106	1	7/12/2004	4.0	200390003	H498	Clone	200342004	A	14	1				3	1	7/27/04	15	13.9	0.66	9/1/2004	1	51	40.0	0.73	0.71
200432107	1	7/12/2004	3.8	200390003	H498	Clone	200342004	A	14	1				0							0				
200432108	1	7/12/2004	3.9	200390003	H498	Clone	200342004	A	14	1				3	1	7/27/04	15	12.8	0.59	9/1/2004	1	51	38.0	0.70	0.67
200432109	1	7/12/2004	3.9	200390003	H498	Clone	200342004	A	14	1				3	1	7/27/04	15	12.3	0.56	9/1/2004	1	51	32.6	0.56	0.56
200432110	1	7/12/2004	2.7	200390003	H498	Clone	200342004	A	14	1	7/12/2004	OVRL		0							0				
200432111	1	7/12/2004	2.8	200390003	H498	Clone	200342004	A	16	1	7/12/2004	OVRL		0							0				
200432112	1	7/12/2004	2.9	200390003	H498	Clone	200342004	A	15	1	7/12/2004	OVRL		0							0				
200432113	2	7/12/2004	2.5	200390003	H498	Clone	200342004	A	14	1	7/12/2004	OVRL		0							0				
200437501	2	7/22/2004	3.2	200390007	Duroc	Clone	200342201	K	13	1				2	1	8/6/04	15	9.4	0.41	9/9/2004	1	49	40.6	0.92	0.76
200437502	2	7/22/2004	3.0	200390007	Duroc	Clone	200342201	K	15	1				2	1	8/6/04	15	10	0.47	9/9/2004	1	49	39.4	0.86	0.74
200437503	2	7/22/2004	2.7	200390007	Duroc	Clone	200342201	K	15	6				0							0				
200437504	1	7/22/2004	4.0	200390007	Duroc	Clone	200342201	K	14	1				3	1	8/6/04	15	10.6	0.44	9/9/2004	1	49	37.6	0.79	0.69
200437505	1	7/22/2004	3.9	200390007	Duroc	Clone	200342201	K	12	1				3	1	8/6/04	15	12.1	0.55	9/9/2004	1	49	44.2	0.94	0.82
200437506	1	7/22/2004	3.7	200390007	Duroc	Clone	200342201	K	13	1				3	1	8/6/04	15	9.9	0.41	9/9/2004	1	49	37.6	0.81	0.69
200437507	1	7/22/2004	3.5	200390007	Duroc	Clone	200342201	K	13	1				3	1	8/6/04	15	11	0.50	9/9/2004	1	49	47.8	1.08	0.90
200437508	1	7/22/2004	3.3	200390007	Duroc	Clone	200342201	K	14	1				0							0				

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA
200437509	1	7/22/2004	3.0	200390007	Duroc	Clone	200342201	K	14	1			3	1	8/6/04	15	10.1	0.47	9/9/2004	1	49	38.4	0.83	0.72
200437510	1	7/22/2004	3.0	200390007	Duroc	Clone	200342201	K	14	1			3	1	8/6/04	15	9.7	0.45	9/9/2004	1	49	35.2	0.75	0.66
200437511	1	7/22/2004	3.0	200390007	Duroc	Clone	200342201	K	13	1	7/22/2004	OVRL		0						0				
200430501	2	7/7/2004	4.0	200190498	H498	Control	200342304	H	14	1			2	1	7/27/04	20	12.4	0.42	9/1/2004	1	56	38.0	0.71	0.61
200430502	2	7/7/2004	3.7	200190498	H498	Control	200342304	H	13	1			2	1	7/27/04	20	9.9	0.31	9/1/2004	1	56	36.8	0.75	0.59
200430503	2	7/7/2004	3.1	200190498	H498	Control	200342304	H	14	1			2	1	7/27/04	20	8.5	0.27	9/1/2004	1	56	35.8	0.76	0.58
200430504	2	7/7/2004	3.2	200190498	H498	Control	200342304	H	12	1			2	1	7/27/04	20	10.1	0.35	9/1/2004	1	56	28.4	0.51	0.45
200430505	2	7/7/2004	2.9	200190498	H498	Control	200342304	H	15	1			2	1	7/27/04	20	8.4	0.28	9/1/2004	1	56	32.4	0.67	0.53
200430506	2	7/7/2004	2.7	200190498	H498	Control	200342304	H	14	1			2	1	7/27/04	20	8.5	0.29	9/1/2004	1	56	34.2	0.71	0.56
200430507	1	7/7/2004	3.8	200190498	H498	Control	200342304	H	14	1			3	1	7/27/04	20	10.1	0.32	9/1/2004	1	56	32.4	0.62	0.51
200430508	1	7/7/2004	3.7	200190498	H498	Control	200342304	H	14	1			3	1	7/27/04	20	9.7	0.30	9/1/2004	1	56	36.4	0.74	0.58
200430509	1	7/7/2004	3.0	200190498	H498	Control	200342304	H	14	1			3	1	7/27/04	20	7.6	0.23	9/1/2004	1	56	27.2	0.54	0.43
200430510	1	7/7/2004	2.2	200190498	H498	Control	200342304	H	12	1			3	1	7/27/04	20	6.2	0.20	9/1/2004	1	56	36.0	0.83	0.60
200430511	1	7/7/2004	3.8	200190498	H498	Control	200342304	H	14	1	7/7/2004	STLB		0						0				
200430512	1	7/7/2004	3.2	200190498	H498	Control	200342304	H	14	1	7/7/2004	STLB		0						0				
200439501	2	7/24/2004	4.1	200390007	Duroc	Clone	200342502	E	13	1			2	1	8/12/04	19	13.5	0.49	9/14/2004	1	52	44.8	0.95	0.78
200439502	2	7/24/2004	4.2	200390007	Duroc	Clone	200342502	E	14	1			2	1	8/12/04	19	15.6	0.60	9/14/2004	1	52	50.8	1.07	0.90
200439503	2	7/24/2004	3.5	200390007	Duroc	Clone	200342502	E	15	1			2	1	8/12/04	19	13	0.50	9/14/2004	1	52	47.0	1.03	0.84
200439504	2	7/24/2004	2.8	200390007	Duroc	Clone	200342502	E	13	1			2	1	8/12/04	19	12.8	0.53	9/14/2004	1	52	40.4	0.84	0.72
200439505	1	7/24/2004	4.4	200390007	Duroc	Clone	200342502	E	12	1			3	1	8/12/04	19	15.1	0.56	9/14/2004	1	52	44.2	0.88	0.77
200439506	1	7/24/2004	4.2	200390007	Duroc	Clone	200342502	E	12	1	7/24/2004	STLB		0						0				
200439507	1	7/24/2004	3.6	200390007	Duroc	Clone	200342502	E	12	1	7/24/2004	STLB		0						0				
200439508	1	7/24/2004	1.4	200390007	Duroc	Clone	200342502	E	12	1	7/24/2004	STLB		0						0				
200433401	2	7/13/2004	4.1	200190498	H498	Control	200342601	E	15	1			2	1	7/30/04	17	13.4	0.55	9/2/2004	1	51	31.2	0.52	0.53
200433402	2	7/13/2004	3.8	200190498	H498	Control	200342601	E	14	1			2	1	7/30/04	17	13	0.54	9/2/2004	1	51	32.8	0.58	0.57
200433403	2	7/13/2004	3.4	200190498	H498	Control	200342601	E	15	1			2	1	7/30/04	17	11.3	0.46	9/2/2004	1	51	30.0	0.55	0.52
200433404	2	7/13/2004	3.4	200190498	H498	Control	200342601	E	15	1			2	1	7/30/04	17	12.8	0.55	9/2/2004	1	51	32.8	0.59	0.58
200433405	2	7/13/2004	3.5	200190498	H498	Control	200342601	E	15	1			2	1	7/30/04	17	13	0.56	9/2/2004	1	51	33.6	0.61	0.59
200433406	2	7/13/2004	3.3	200190498	H498	Control	200342601	E	15	1			2	1	7/30/04	17	9.4	0.36	9/2/2004	1	51	28.6	0.56	0.50
200433407	2	7/13/2004	2.4	200190498	H498	Control	200342601	E	14	1			2	1	7/30/04	17	4.4	0.12	9/1/2004	1	50	23.2	0.57	0.42
200433408	1	7/13/2004	4.6	200190498	H498	Control	200342601	E	14	1			3	1	7/30/04	17	13.8	0.54	9/2/2004	1	51	38.9	0.74	0.67
200433409	1	7/13/2004	4.3	200190498	H498	Control	200342601	E	13	1			3	1	7/30/04	17	14.1	0.58	9/2/2004	1	51	44.6	0.90	0.79
200433410	2	7/13/2004	3.6	200190498	H498	Control	200342601	E	14	1	7/13/2004	STLB		0						0				
200433201	2	7/13/2004	4.4	200390005	H498	Clone	200342902	C	13	1			2	1	7/30/04	17	12.7	0.49	9/2/2004	1	51	39.6	0.79	0.69
200433202	2	7/13/2004	4.0	200390005	H498	Clone	200342902	C	13	1			2	1	7/30/04	17	10.8	0.40	9/2/2004	1	51	31.2	0.60	0.53
200433203	2	7/13/2004	3.4	200390005	H498	Clone	200342902	C	13	1			2	1	7/30/04	17	12.7	0.55	9/2/2004	1	51	32.6	0.59	0.57
200433204	2	7/13/2004	3.2	200390005	H498	Clone	200342902	C	15	1			2	1	7/30/04	17	10.2	0.41	9/2/2004	1	51	27.6	0.51	0.48
200433205	1	7/13/2004	4.5	200390005	H498	Clone	200342902	C	14	1			3	1	7/30/04	17	14.4	0.58	9/2/2004	1	51	36.4	0.65	0.63
200433206	1	7/13/2004	4.1	200390005	H498	Clone	200342902	C	14	1			3	1	7/30/04	17	10.3	0.36	9/2/2004	1	51	34.2	0.70	0.59
200433207	1	7/13/2004	3.7	200390005	H498	Clone	200342902	C	13	1			3	1	7/30/04	17	10.4	0.39	9/2/2004	1	51	30.6	0.59	0.53
200433208	1	7/13/2004	3.5	200390005	H498	Clone	200342902	C	14	1			3	1	7/30/04	17	10.6	0.42	9/2/2004	1	51	31.8	0.62	0.55
200433209	1	7/13/2004	3.0	200390005	H498	Clone	200342902	C	14	1	7/13/2004	OVRL		0						0				
200433210	2	7/13/2004	3.8	200390005	H498	Clone	200342902	C	14	1	7/13/2004	STLB		0						0				
200431801	2	7/12/2004	3.2	200398128	Duroc	Control	200342905	C	14	1			2	1	7/27/04	15	11.2	0.53	9/1/2004	1	51	37.4	0.73	0.67
200431802	2	7/12/2004	3.3	200398128	Duroc	Control	200342905	C	14	1			2	1	7/27/04	15	10.2	0.46	9/1/2004	1	51	44.0	0.94	0.80
200431803	2	7/12/2004	2.3	200398128	Duroc	Control	200342905	C	13	1			2	1	7/27/04	15	6.6	0.29	9/1/2004	1	51	24.4	0.49	0.43
200431804	1	7/12/2004	3.0	200398128	Duroc	Control	200342905	C	12	1			3	1	7/27/04	15	10.2	0.48	9/1/2004	1	51	28.2	0.50	0.49
200431805	1	7/12/2004	3.0	200398128	Duroc	Control	200342905	C	12	1			3	1	7/27/04	15	10.3	0.49	9/1/2004	1	51	32.6	0.62	0.58
200431806	2	7/12/2004	3.2	200398128	Duroc	Control	200342905	C	14	1	7/12/2004	OVRL		0						0				
200431807	1	7/12/2004	3.0	200398128	Duroc	Control	200342905	C	12	1	7/12/2004	OVRL		0						0				
200431808	1	7/12/2004	2.3	200398128	Duroc	Control	200342905	C	13	1	7/12/2004	OVRL		0						0				
200431809	2	7/12/2004	2.3	200398128	Duroc	Control	200342905	C	12	1	7/12/2004	OVRL		0						0				
200431810	1	7/12/2004	3.5	200398128	Duroc	Control	200342905	C	14	1	7/12/2004	STLB		0						0				

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA	
200431811	1	7/12/2004	3.4	200398128	Duroc	Control	200342905	C	14	1	7/12/2004	STLB		0							0				
200431812	1	7/12/2004	2.2	200398128	Duroc	Control	200342905	C	14	1	7/12/2004	STLB		0								0			
200431813	1	7/12/2004	3.5	200398128	Duroc	Control	200342905	C	13	1	7/12/2004	STLB		0								0			
200431814	2	7/12/2004	2.5	200398128	Duroc	Control	200342905	C	13	1	7/12/2004	STLB		0								0			
200431601	2	7/10/2004	4.0	200390002	H498	Clone	200343201	A	14	1			2	1	7/27/04	17	11.2	0.42	9/1/2004	1	53	33.6	0.62	0.56	
200431602	2	7/10/2004	3.9	200390002	H498	Clone	200343201	A	14	1			2	1	7/27/04	17	13.1	0.54	9/1/2004	1	53	38.0	0.69	0.64	
200431603	2	7/10/2004	3.5	200390002	H498	Clone	200343201	A	14	1			2	1	7/27/04	17	12.2	0.51	9/1/2004	1	53	35.8	0.66	0.61	
200431604	2	7/10/2004	3.4	200390002	H498	Clone	200343201	A	14	1			2	1	7/27/04	17	11.5	0.48	9/1/2004	1	53	36.2	0.69	0.62	
200431605	2	7/10/2004	2.9	200390002	H498	Clone	200343201	A	12	1			2	1	7/27/04	17	9.3	0.38	9/1/2004	1	53	31.4	0.61	0.54	
200431606	1	7/10/2004	4.4	200390002	H498	Clone	200343201	A	15	1			3	1	7/27/04	17	13.8	0.55	9/1/2004	1	53	40.8	0.75	0.69	
200431607	1	7/10/2004	3.6	200390002	H498	Clone	200343201	A	15	1			3	1	7/27/04	17	13.3	0.57	9/1/2004	1	53	36.0	0.63	0.61	
200431608	1	7/10/2004	3.9	200390002	H498	Clone	200343201	A	14	1			3	1	7/27/04	17	13.9	0.59	9/1/2004	1	53	45.4	0.88	0.78	
200431609	1	7/10/2004	3.3	200390002	H498	Clone	200343201	A	14	1			3	0							0				
200431610	1	7/10/2004	2.9	200390002	H498	Clone	200343201	A	15	1			3	1	7/27/04	17	11.4	0.50	9/1/2004	1	53	36.0	0.68	0.62	
200431611	1	7/10/2004	3.0	200390002	H498	Clone	200343201	A	11	1	7/10/2004	OVRL		0							0				
200435401	2	7/18/2004	3.7	200395515	Duroc	Control	200343605	F	14	1			2	1	8/5/04	18	8	0.24	9/7/2004	1	51	36.2	0.85	0.64	
200435402	2	7/18/2004	3.4	200395515	Duroc	Control	200343605	F	15	1			2	1	8/5/04	18	7.6	0.23	9/7/2004	1	51	33.6	0.79	0.59	
200435403	2	7/18/2004	3.1	200395515	Duroc	Control	200343605	F	14	1			2	1	8/5/04	18	12	0.49	9/7/2004	1	51	40.4	0.86	0.73	
200435404	2	7/18/2004	2.9	200395515	Duroc	Control	200343605	F	15	1			2	1	8/5/04	18	11.6	0.48	9/7/2004	1	51	45.8	1.04	0.84	
200435405	2	7/18/2004	2.6	200395515	Duroc	Control	200343605	F	17	1			2	1	8/5/04	18	11.3	0.48	9/7/2004	1	51	37.2	0.78	0.68	
200435406	2	7/18/2004	2.0	200395515	Duroc	Control	200343605	F	12	1			2	0							0				
200435407	1	7/18/2004	3.5	200395515	Duroc	Control	200343605	F	14	1			3	1	8/5/04	18	12.4	0.49	9/7/2004	1	51	44.6	0.98	0.81	
200435408	1	7/18/2004	3.3	200395515	Duroc	Control	200343605	F	14	1			3	1	8/5/04	18	10.9	0.42	9/7/2004	1	51	43.0	0.97	0.78	
200435409	1	7/18/2004	3.0	200395515	Duroc	Control	200343605	F	14	1			3	1	8/5/04	18	10.4	0.41	9/7/2004	1	51	37.2	0.81	0.67	
200435410	1	7/18/2004	3.0	200395515	Duroc	Control	200343605	F	14	1			3	1	8/5/04	18	8.2	0.29	9/7/2004	1	51	31.2	0.70	0.55	
200435411	1	7/18/2004	2.8	200395515	Duroc	Control	200343605	F	14	1			3	1	8/5/04	18	10.6	0.43	9/7/2004	1	51	34.4	0.72	0.62	
200435412	1	7/18/2004	2.4	200395515	Duroc	Control	200343605	F	14	1			3	1	8/5/04	18	10.3	0.44	9/7/2004	1	51	35.0	0.75	0.64	
200435413	1	7/18/2004	1.6	200395515	Duroc	Control	200343605	F	14	1			3	1	8/5/04	18	8	0.36	9/7/2004	1	51	29.4	0.65	0.55	
200439401	2	7/24/2004	3.8	200398128	Duroc	Control	200343802	C	14	1			2	1	8/12/04	19	12.5	0.46	9/14/2004	1	52	47.6	1.06	0.84	
200439402	1	7/24/2004	3.8	200398128	Duroc	Control	200343802	C	8	1			3	0							0				
200439403	1	7/24/2004	3.3	200398128	Duroc	Control	200343802	C	13	1			3	1	8/12/04	19	11.8	0.45	9/14/2004	1	52	34.6	0.69	0.60	
200439404	1	7/24/2004	3.2	200398128	Duroc	Control	200343802	C	12	1			3	1	8/12/04	19	11.7	0.45	9/14/2004	1	52	41.8	0.91	0.74	
200439405	1	7/24/2004	3.1	200398128	Duroc	Control	200343802	C	13	1			3	1	8/12/04	19	12.5	0.49	9/14/2004	1	52	42.8	0.92	0.76	
200439406	1	7/24/2004	3.1	200398128	Duroc	Control	200343802	C	12	1			3	1	8/12/04	19	12	0.47	9/14/2004	1	52	42.8	0.93	0.76	
200439407	1	7/24/2004	3.0	200398128	Duroc	Control	200343802	C	13	1			3	1	8/12/04	19	10.5	0.39	9/14/2004	1	52	35.4	0.75	0.62	
200439408	1	7/24/2004	2.6	200398128	Duroc	Control	200343802	C	13	1			3	1	8/12/04	19	11.3	0.46	9/14/2004	1	52	38.6	0.83	0.69	
200439409	1	7/24/2004	2.7	200398128	Duroc	Control	200343802	C	12	1			3	1	8/12/04	19	11.7	0.47	9/14/2004	1	52	41.8	0.91	0.75	
200439410	1	7/24/2004	2.2	200398128	Duroc	Control	200343802	C	14	1	7/24/2004	OVRL		0							0				
200432401	2	7/12/2004	4.3	200390002	H498	Clone	200344002	N	15	1			2	1	7/27/04	15	9.5	0.35	9/2/2004	1	52	33.8	0.66	0.57	
200432402	2	7/12/2004	4.0	200390002	H498	Clone	200344002	N	14	1			2	1	7/27/04	15	9.8	0.39	9/2/2004	1	52	37.0	0.74	0.63	
200432403	2	7/12/2004	3.8	200390002	H498	Clone	200344002	N	16	1			2	1	7/27/04	15	9.5	0.38	9/2/2004	1	52	32.8	0.63	0.56	
200432404	2	7/12/2004	3.6	200390002	H498	Clone	200344002	N	13	1			2	1	7/27/04	15	8.9	0.35	9/2/2004	1	52	37.9	0.78	0.66	
200432405	1	7/12/2004	4.4	200390002	H498	Clone	200344002	N	12	1			3	1	7/27/04	15	11.4	0.47	9/2/2004	1	52	43.6	0.87	0.75	
200432406	1	7/12/2004	4.0	200390002	H498	Clone	200344002	N	13	1			3	1	7/27/04	15	11.5	0.50	9/2/2004	1	52	42.8	0.85	0.75	
200432407	1	7/12/2004	4.0	200390002	H498	Clone	200344002	N	14	1			3	1	7/27/04	15	9.3	0.35	9/2/2004	1	52	33.8	0.66	0.57	
200432408	1	7/12/2004	3.0	200390002	H498	Clone	200344002	N	13	1			3	0							0				
200432409	1	7/12/2004	3.0	200390002	H498	Clone	200344002	N	14	1	7/12/2004	OVRL		0							0				
200434601	2	7/16/2004	4.1	200390002	H498	Clone	200344102	H	14	1			2	1	8/5/04	20	13.8	0.49	9/7/2004	1	53	42.2	0.86	0.72	
200434602	2	7/16/2004	3.1	200390002	H498	Clone	200344102	H	14	1			2	1	8/5/04	20	12.9	0.49	9/7/2004	1	53	38.2	0.77	0.66	
200434603	2	7/16/2004	3.0	200390002	H498	Clone	200344102	H	14	1			2	1	8/5/04	20	9.1	0.31	9/7/2004	1	53	29.0	0.60	0.49	
200434604	2	7/16/2004	3.1	200390002	H498	Clone	200344102	H	14	1			2	1	8/5/04	20	12.7	0.48	9/7/2004	1	53	33.8	0.64	0.58	

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA	
200434605	2	7/16/2004	2.8	200390002	H498	Clone	200344102	H	14	1			2	1	8/5/04	20	9.1	0.32	9/7/2004	1	53	31.8	0.69	0.55	
200434606	2	7/16/2004	2.7	200390002	H498	Clone	200344102	H	13	1			2	1	8/5/04	20	10.1	0.37	9/7/2004	1	53	32.4	0.68	0.56	
200434607	1	7/16/2004	2.6	200390002	H498	Clone	200344102	H	12	1			3	1	8/5/04	20	9.6	0.35	9/7/2004	1	53	29.0	0.59	0.50	
200434608	1	7/16/2004	2.6	200390002	H498	Clone	200344102	H	12	1			3	1	8/5/04	20	10.7	0.41	9/7/2004	1	53	33.2	0.68	0.58	
200434609	2	7/16/2004	3.4	200390002	H498	Clone	200344102	H	13	1	7/16/2004	OVRL		0							0				
200434610	1	7/16/2004	2.9	200390002	H498	Clone	200344102	H	14	1	7/16/2004	STLB		0							0				
200434611	2	7/16/2004	2.8	200390002	H498	Clone	200344102	H	14	1	7/16/2004	STLB		0							0				
200434612	1	7/16/2004	3.3	200390002	H498	Clone	200344102	H	13	1	7/16/2004	STLB		0							0				
200436001	2	7/19/2004	3.3	200390005	H498	Clone	200344201	K	14	1			2	1	8/5/04	17	11.6	0.49	9/7/2004	1	50	34.8	0.70	0.63	
200436002	2	7/19/2004	3.2	200390005	H498	Clone	200344201	K	14	1			2	1	8/5/04	17	13.4	0.60	9/7/2004	1	50	34.8	0.65	0.63	
200436003	2	7/19/2004	3.0	200390005	H498	Clone	200344201	K	15	1			2	1	8/5/04	17	10.3	0.43	9/7/2004	1	50	35.6	0.77	0.65	
200436004	2	7/19/2004	2.9	200390005	H498	Clone	200344201	K	14	1			2	1	8/5/04	17	6.5	0.21	9/7/2004	1	50	33.6	0.82	0.61	
200436005	2	7/19/2004	2.8	200390005	H498	Clone	200344201	K	14	1			2	1	8/5/04	17	8.9	0.36	9/9/2004	1	52	35.0	0.75	0.62	
200436006	2	7/19/2004	2.8	200390005	H498	Clone	200344201	K	16	1			2	1	8/5/04	17	7.8	0.29	9/9/2004	1	52	39.8	0.91	0.71	
200436007	2	7/19/2004	2.6	200390005	H498	Clone	200344201	K	16	1			2	1	8/5/04	17	9.6	0.41	9/9/2004	1	52	40.0	0.87	0.72	
200436008	2	7/19/2004	2.5	200390005	H498	Clone	200344201	K	14	1			2	1	8/5/04	17	10.6	0.48	9/9/2004	1	52	38.4	0.79	0.69	
200436009	1	7/19/2004	3.4	200390005	H498	Clone	200344201	K	13	1			3	1	8/5/04	17	11	0.45	9/9/2004	1	52	47.8	1.05	0.85	
200436010	1	7/19/2004	3.1	200390005	H498	Clone	200344201	K	15	1			3	1	8/5/04	17	11.3	0.48	9/9/2004	1	52	43.8	0.93	0.78	
200436011	1	7/19/2004	2.9	200390005	H498	Clone	200344201	K	16	1			3	1	8/5/04	17	11.2	0.49	9/9/2004	1	52	37.2	0.74	0.66	
200436012	1	7/19/2004	2.4	200390005	H498	Clone	200344201	K	15	1			3	1	8/5/04	17	9.6	0.42	9/9/2004	1	52	44.6	1.00	0.81	
200436013	1	7/19/2004	2.1	200390005	H498	Clone	200344201	K	14	1			3	1	8/5/04	17	8.8	0.39	9/9/2004	1	52	32.6	0.68	0.59	
200436014	1	7/19/2004	1.3	200390005	H498	Clone	200344201	K	15	1				0							0				
200436015	1	7/19/2004	3.2	200390005	H498	Clone	200344201	K	14	4	7/20/2004	DDFR		0							0				
200436016	1	7/19/2004	3.5	200390005	H498	Clone	200344201	K	14	1	7/19/2004	STLB		0							0				
200434501	2	7/16/2004	2.8	200390003	H498	Clone	200344402	B	14	1			2	1	8/5/04	20	11.9	0.46	9/7/2004	1	53	34.8	0.69	0.60	
200434502	2	7/16/2004	2.7	200390003	H498	Clone	200344402	B	13	1			2	1	8/5/04	20	11.7	0.45	9/7/2004	1	53	34.6	0.69	0.60	
200434503	2	7/16/2004	2.4	200390003	H498	Clone	200344402	B	14	1			2	1	8/5/04	20	12	0.48	9/7/2004	1	53	34.4	0.68	0.60	
200434504	2	7/16/2004	2.6	200390003	H498	Clone	200344402	B	14	1			2	1	8/5/04	20	12.6	0.50	9/7/2004	1	53	41.2	0.87	0.73	
200434505	2	7/16/2004	1.5	200390003	H498	Clone	200344402	B	13	1				0							0				
200434506	1	7/16/2004	3.4	200390003	H498	Clone	200344402	B	14	1			3	1	8/5/04	20	13.2	0.49	9/7/2004	1	53	39.2	0.79	0.68	
200434507	1	7/16/2004	3.4	200390003	H498	Clone	200344402	B	12	1			3	1	8/5/04	20	16.6	0.66	9/7/2004	1	53	46.2	0.90	0.81	
200434508	1	7/16/2004	3.0	200390003	H498	Clone	200344402	B	12	1			3	1	8/5/04	20	13.5	0.53	9/7/2004	1	53	39.2	0.78	0.68	
200434509	1	7/16/2004	2.5	200390003	H498	Clone	200344402	B	15	1			3	1	8/5/04	20	11.8	0.47	9/7/2004	1	53	30.8	0.58	0.53	
200434510	1	7/16/2004	2.5	200390003	H498	Clone	200344402	B	13	1			3	1	8/5/04	20	13.1	0.53	9/7/2004	1	53	39.0	0.78	0.69	
200434511	1	7/16/2004	2.5	200390003	H498	Clone	200344402	B	14	1			3	1	8/5/04	20	9.3	0.34	9/7/2004	1	53	39.2	0.91	0.69	
200434512	1	7/16/2004	2.4	200390003	H498	Clone	200344402	B	13	1			3	1	8/5/04	20	13	0.53	9/7/2004	1	53	37.0	0.73	0.65	
200434901	2	7/17/2004	3.8	200398128	Duroc	Control	200344403	B	15	1			2	1	8/5/04	19	8.9	0.27	9/7/2004	1	52	37.2	0.86	0.64	
200434902	2	7/17/2004	3.5	200398128	Duroc	Control	200344403	B	12	1			2	1	8/5/04	19	11.4	0.42	9/7/2004	1	52	43.4	0.97	0.77	
200434903	2	7/17/2004	2.9	200398128	Duroc	Control	200344403	B	14	1			2	1	8/5/04	19	9.8	0.36	9/7/2004	1	52	38.6	0.87	0.69	
200434904	2	7/17/2004	2.3	200398128	Duroc	Control	200344403	B	13	1			2	1	8/5/04	19	8.4	0.32	9/7/2004	1	52	35.0	0.81	0.63	
200434905	1	7/17/2004	3.5	200398128	Duroc	Control	200344403	B	15	1			3	1	8/5/04	19	11.5	0.42	9/7/2004	1	52	41.2	0.90	0.73	
200434906	1	7/17/2004	3.0	200398128	Duroc	Control	200344403	B	14	1				0							0				
200434907	1	7/17/2004	3.4	200398128	Duroc	Control	200344403	B	13	1	7/17/2004	OVRL		0							0				
200434908	2	7/17/2004	3.5	200398128	Duroc	Control	200344403	B	14	1	7/17/2004	STLB		0							0				
200434909	1	7/17/2004	3.2	200398128	Duroc	Control	200344403	B	14	1	7/17/2004	STLB		0							0				
200434910	1	7/17/2004	3.5	200398128	Duroc	Control	200344403	B	15	1	7/17/2004	STLB		0							0				
200434911	2	7/17/2004	3.0	200398128	Duroc	Control	200344403	B	14	1	7/17/2004	STLB		0							0				
200434912	2	7/17/2004	4.1	200398128	Duroc	Control	200344403	B	14	1	7/17/2004	STLB		0							0				
200431401	2	7/10/2004	4.3	200390005	H498	Clone	200344902	D	14	1			2	1	7/27/04	17	12.5	0.48	9/1/2004	1	53	41.2	0.80	0.70	
200431402	2	7/10/2004	4.3	200390005	H498	Clone	200344902	D	14	1			2	1	7/27/04	17	12.4	0.48	9/1/2004	1	53	45.2	0.91	0.77	
200431403	2	7/10/2004	3.8	200390005	H498	Clone	200344902	D	12	1			2	1	7/27/04	17	10.6	0.40	9/1/2004	1	53	33.4	0.63	0.56	

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA
200431404	2	7/10/2004	4.1	200390005	H498	Clone	200344902	D	12	1			2	1	7/27/04	17	10.8	0.39	9/1/2004	1	53	39.0	0.78	0.66
200431405	2	7/10/2004	3.7	200390005	H498	Clone	200344902	D	12	1			2	1	7/27/04	17	12	0.49	9/1/2004	1	53	43.8	0.88	0.76
200431406	2	7/10/2004	3.7	200390005	H498	Clone	200344902	D	12	1			2	1	7/27/04	17	11.8	0.48	9/1/2004	1	53	32.4	0.57	0.54
200431407	2	7/10/2004	3.6	200390005	H498	Clone	200344902	D	14	1			2	1	7/27/04	17	12.2	0.51	9/1/2004	1	53	39.8	0.77	0.68
200431408	2	7/10/2004	3.5	200390005	H498	Clone	200344902	D	13	1			2	1	7/27/04	17	10.5	0.41	9/1/2004	1	53	41.4	0.86	0.72
200431409	1	7/10/2004	4.7	200390005	H498	Clone	200344902	D	13	1			3	1	7/27/04	17	12.1	0.44	9/1/2004	1	53	46.4	0.95	0.79
200431410	1	7/10/2004	3.9	200390005	H498	Clone	200344902	D	14	1			3	1	7/27/04	17	10	0.36	9/1/2004	1	53	32.4	0.62	0.54
200431411	1	7/10/2004	4.1	200390005	H498	Clone	200344902	D	12	1			3	1	7/27/04	17	11.8	0.45	9/1/2004	1	53	38.6	0.74	0.65
200431412	1	7/10/2004	3.4	200390005	H498	Clone	200344902	D	13	1	7/11/2004	OVRL		0						0				
200431413	2	7/10/2004	3.7	200390005	H498	Clone	200344902	D	13	1	7/10/2004	OVRL		0						0				
200431414	2	7/10/2004	2.3	200390005	H498	Clone	200344902	D	14	1	7/10/2004	OVRL		0						0				
200431001	2	7/8/2004	4.4	200390002	H498	Clone	200344904	D	14	1			2	1	7/27/04	19	13.3	0.47	9/1/2004	1	55	45.6	0.90	0.75
200431002	2	7/8/2004	3.6	200390002	H498	Clone	200344904	D	14	1			2	1	7/27/04	19	9.5	0.31	9/1/2004	1	55	33.4	0.66	0.54
200431003	2	7/8/2004	3.3	200390002	H498	Clone	200344904	D	13	1			2	1	7/27/04	19	6.8	0.18	9/1/2004	1	55	39.8	0.92	0.66
200431004	2	7/8/2004	3.2	200390002	H498	Clone	200344904	D	14	1			2	1	7/27/04	19	9.3	0.32	9/1/2004	1	55	29.6	0.56	0.48
200431005	2	7/8/2004	3.1	200390002	H498	Clone	200344904	D	14	1			2	1	7/27/04	19	9.7	0.35	9/1/2004	1	55	35.4	0.71	0.59
200431006	1	7/8/2004	4.5	200390002	H498	Clone	200344904	D	13	1			3	1	7/27/04	19	12.9	0.44	9/1/2004	1	55	50.8	1.05	0.84
200431007	1	7/8/2004	4.2	200390002	H498	Clone	200344904	D	14	1			3	1	7/27/04	19	11.6	0.39	9/1/2004	1	55	36.0	0.68	0.58
200431008	1	7/8/2004	4.2	200390002	H498	Clone	200344904	D	14	1			3	1	7/27/04	19	12.3	0.43	9/1/2004	1	55	40.6	0.79	0.66
200431009	1	7/8/2004	4.1	200390002	H498	Clone	200344904	D	14	1			3	1	7/27/04	19	11	0.36	9/1/2004	1	55	46.4	0.98	0.77
200431010	1	7/8/2004	4.1	200390002	H498	Clone	200344904	D	15	1			3	1	7/27/04	19	7.6	0.18	9/1/2004	1	55	32.0	0.68	0.51
200431011	1	7/8/2004	3.1	200390002	H498	Clone	200344904	D	14	99			3	1	7/27/04	19	11.4	0.44	9/1/2004	1	55	40.6	0.81	0.68
200431012	1	7/8/2004	3.1	200390002	H498	Clone	200344904	D	14	99			3	1	7/27/04	19	8.7	0.29	9/1/2004	1	55	40.4	0.88	0.68
200431013	1	7/8/2004	2.2	200390002	H498	Clone	200344904	D	14	99				0						0				
200431014	2	7/8/2004	3.8	200390002	H498	Clone	200344904	D	14	1	7/8/2004	OVRL		0						0				
200431015	1	7/8/2004	4.1	200390002	H498	Clone	200344904	D	14	1	7/8/2004	OVRL		0						0				
200431016	2	7/8/2004	2.8	200390002	H498	Clone	200344904	D	13	1	7/8/2004	STLB		0						0				
200431017	2	7/8/2004	3.9	200390002	H498	Clone	200344904	D	14	1	7/8/2004	STLB		0						0				
200431018	1	7/8/2004	3.4	200390002	H498	Clone	200344904	D	13	1	7/8/2004	STLB		0						0				
200432501	2	7/13/2004	3.8	200390003	H498	Clone	200345003	J	14	1			2	1	7/30/04	17	12.9	0.54	9/2/2004	1	51	47.6	1.02	0.86
200432502	2	7/13/2004	3.7	200390003	H498	Clone	200345003	J	14	1			2	1	7/30/04	17	12.4	0.51	9/2/2004	1	51	41.2	0.85	0.74
200432503	2	7/13/2004	3.1	200390003	H498	Clone	200345003	J	14	1			2	1	7/30/04	17	12.8	0.57	9/2/2004	1	51	43.9	0.91	0.80
200432504	2	7/13/2004	3.5	200390003	H498	Clone	200345003	J	14	1			2	1	7/30/04	17	12.7	0.54	9/2/2004	1	51	41.0	0.83	0.74
200432505	2	7/13/2004	2.9	200390003	H498	Clone	200345003	J	14	1				0						0				
200432506	2	7/13/2004	2.5	200390003	H498	Clone	200345003	J	14	1			2	1	7/30/04	17	9.1	0.39	9/2/2004	1	51	25.0	0.47	0.44
200432507	1	7/13/2004	3.4	200390003	H498	Clone	200345003	J	15	1			3	1	7/30/04	17	10.5	0.42	9/2/2004	1	51	39.2	0.84	0.70
200432508	1	7/13/2004	3.2	200390003	H498	Clone	200345003	J	14	1			3	1	7/30/04	17	10.5	0.43		0				
200432509	2	7/13/2004	3.2	200390003	H498	Clone	200345003	J	13	1	7/13/2004	OVRL		0						0				
200432510	2	7/13/2004	2.5	200390003	H498	Clone	200345003	J	14	1	7/13/2004	STLB		0						0				
200432511	2	7/13/2004	2.6	200390003	H498	Clone	200345003	J	14	1	7/13/2004	STLB		0						0				
200432512	2	7/13/2004	2.3	200390003	H498	Clone	200345003	J	13	1	7/13/2004	STLB		0						0				
200431701	2	7/11/2004	3.5	200390005	H498	Clone	200345103	A	15	1			2	1	7/27/04	16	10.3	0.43	9/1/2004	1	52	33.8	0.65	0.58
200431702	1	7/11/2004	3.5	200390005	H498	Clone	200345103	A	16	1			3	1	7/27/04	16	11.9	0.53	9/1/2004	1	52	36.4	0.68	0.63
200431703	1	7/11/2004	3.2	200390005	H498	Clone	200345103	A	13	1			3	1	7/27/04	16	11.5	0.52	9/1/2004	1	52	37.8	0.73	0.67
200431704	1	7/11/2004	3.4	200390005	H498	Clone	200345103	A	15	1			3	1	7/27/04	16	12.1	0.54	9/1/2004	1	52	38.0	0.72	0.67
200431705	1	7/11/2004	3.4	200390005	H498	Clone	200345103	A	14	1				0						0				
200431706	1	7/11/2004	3.2	200390005	H498	Clone	200345103	A	14	1			3	1	7/27/04	16	11.8	0.54	9/1/2004	1	52	39.0	0.76	0.69
200431707	1	7/11/2004	3.2	200390005	H498	Clone	200345103	A	14	1				0						0				
200431708	1	7/11/2004	1.9	200390005	H498	Clone	200345103	A	14	1			3	1	7/27/04	16	8.5	0.41	9/1/2004	1	52	27.6	0.53	0.49
200431709	1	7/11/2004	2.1	200390005	H498	Clone	200345103	A	14	1	7/11/2004	OVRL		0						0				
200434401	2	7/15/2004	4.0	200390005	H498	Clone	200345302	H	14	1			2	1	7/30/04	15	9.8	0.39	9/2/2004	1	49	36.0	0.77	0.65

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA
200434402	2	7/15/2004	3.9	200390005	H498	Clone	200345302	H	13	1			2	1	7/30/04	15	11.7	0.52	9/2/2004	1	49	37.8	0.77	0.69
200434403	1	7/15/2004	3.8	200390005	H498	Clone	200345302	H	14	1			3	1	7/30/04	15	9.7	0.39	9/2/2004	1	49	36.0	0.77	0.66
200430801	2	7/7/2004	4.3	200390005	H498	Clone	200345501	F	13	1			2	1	7/27/04	20	11.5	0.36	9/1/2004	1	56	37.6	0.73	0.59
200430802	2	7/7/2004	3.9	200390005	H498	Clone	200345501	F	14	1			2	1	7/27/04	20	12.1	0.41	9/1/2004	1	56	45.0	0.91	0.73
200430803	1	7/7/2004	4.0	200390005	H498	Clone	200345501	F	13	1			3	1	7/27/04	20	8.3	0.22	9/1/2004	1	56	35.2	0.75	0.56
200430804	1	7/7/2004	4.0	200390005	H498	Clone	200345501	F	13	1			3	1	7/27/04	20	13.9	0.50	9/1/2004	1	56	50.4	1.01	0.83
200430805	1	7/7/2004	3.7	200390005	H498	Clone	200345501	F	14	1			3	1	7/27/04	20	12.9	0.46	9/1/2004	1	56	43.6	0.85	0.71
200434001	2	7/15/2004	3.4	200398128	Duroc	Control	200345603	M	14	1			2	1	7/30/04	15	7.3	0.26	9/2/2004	1	49	36.6	0.86	0.68
200434002	2	7/15/2004	3.0	200398128	Duroc	Control	200345603	M	14	1			2	1	7/30/04	15	10.9	0.53	9/2/2004	1	49	42.6	0.93	0.81
200434003	2	7/15/2004	3.1	200398128	Duroc	Control	200345603	M	13	1			2	1	7/30/04	15	9.7	0.44	9/2/2004	1	49	35.0	0.74	0.65
200434004	2	7/15/2004	3.0	200398128	Duroc	Control	200345603	M	13	1			2	1	7/30/04	15	8.2	0.35	9/2/2004	1	49	40.0	0.94	0.76
200434005	2	7/15/2004	2.7	200398128	Duroc	Control	200345603	M	12	1			2	1	7/30/04	15	8.3	0.37	9/2/2004	1	49	32.4	0.71	0.61
200434006	1	7/15/2004	3.4	200398128	Duroc	Control	200345603	M	15	1			3	1	7/30/04	15	8.8	0.36	9/2/2004	1	49	34.2	0.75	0.63
200434007	1	7/15/2004	3.4	200398128	Duroc	Control	200345603	M	13	1			3	1	7/30/04	15	12.2	0.59	9/2/2004	1	49	35.0	0.67	0.64
200434008	1	7/15/2004	2.7	200398128	Duroc	Control	200345603	M	13	1			3	1	7/30/04	15	9.1	0.43	9/2/2004	1	49	40.4	0.92	0.77
200434009	1	7/15/2004	1.7	200398128	Duroc	Control	200345603	M	12	1			3	1	7/30/04	15	8.1	0.43	9/2/2004	1	49	37.6	0.87	0.73
200434010	2	7/15/2004	1.5	200398128	Duroc	Control	200345603	M	13	1	7/15/2004	STLB		0						0				
200434011	1	7/15/2004	2.5	200398128	Duroc	Control	200345603	M	13	1	7/15/2004	STLB		0						0				
200433501	2	7/13/2004	5.1	200190498	H498	Control	200345803	D	13	1			2	1	7/30/04	17	12	0.41	9/2/2004	1	51	28.8	0.49	0.46
200433502	2	7/13/2004	4.7	200190498	H498	Control	200345803	D	14	1			2	1	7/30/04	17	12.4	0.45	9/2/2004	1	51	35.8	0.69	0.61
200433503	2	7/13/2004	4.1	200190498	H498	Control	200345803	D	14	1			2	1	7/30/04	17	8.7	0.27	9/2/2004	1	51	26.4	0.52	0.44
200433504	2	7/13/2004	3.9	200190498	H498	Control	200345803	D	12	1			2	1	7/30/04	17	10.4	0.38	9/2/2004	1	51	32.9	0.66	0.57
200433505	2	7/13/2004	3.4	200190498	H498	Control	200345803	D	12	1			2	1	7/30/04	17	11.4	0.47	9/2/2004	1	51	33.9	0.66	0.60
200433506	2	7/13/2004	3.3	200190498	H498	Control	200345803	D	14	1			2	1	7/30/04	17	9.5	0.36	9/2/2004	1	51	28.8	0.57	0.50
200433507	2	7/13/2004	2.6	200190498	H498	Control	200345803	D	14	1			2	1	7/30/04	17	9.4	0.40	9/2/2004	1	51	25.8	0.48	0.45
200433508	1	7/13/2004	4.6	200190498	H498	Control	200345803	D	14	1	7/14/2004	DINJ		0						0				
200433509	1	7/13/2004	4.3	200190498	H498	Control	200345803	D	13	1			3	1	7/30/04	17	13.6	0.55	9/2/2004	1	51	37.6	0.71	0.65
200433510	1	7/13/2004	4.0	200190498	H498	Control	200345803	D	14	1			3	1	7/30/04	17	12.3	0.49	9/2/2004	1	51	35.4	0.68	0.62
200433511	1	7/13/2004	3.9	200190498	H498	Control	200345803	D	14	1			3	1	7/30/04	17	12.1	0.48	9/2/2004	1	51	31.2	0.56	0.54
200433512	1	7/13/2004	3.6	200190498	H498	Control	200345803	D	13	1			3	1	7/30/04	17	9.8	0.36	9/2/2004	1	51	30.4	0.61	0.53
200433701	2	7/14/2004	4.2	200390007	Duroc	Clone	200346101	D	11	1			2	1	7/30/04	16	13.2	0.56	9/2/2004	1	50	35.0	0.64	0.62
200433702	2	7/14/2004	4.1	200390007	Duroc	Clone	200346101	D	13	1			2	1	7/30/04	16	13	0.56	9/2/2004	1	50	38.4	0.75	0.69
200433703	2	7/14/2004	4.1	200390007	Duroc	Clone	200346101	D	14	1			2	1	7/30/04	16	13.6	0.59	9/2/2004	1	50	38.6	0.74	0.69
200433704	2	7/14/2004	3.4	200390007	Duroc	Clone	200346101	D	12	1			2	1	7/30/04	16	11.1	0.48	9/2/2004	1	50	36.8	0.76	0.67
200433705	2	7/14/2004	3.5	200390007	Duroc	Clone	200346101	D	12	1			2	1	7/30/04	16	11.8	0.52	9/2/2004	1	50	34.6	0.67	0.62
200433706	2	7/14/2004	3.2	200390007	Duroc	Clone	200346101	D	12	1			2	1	7/30/04	16	11	0.49	9/2/2004	1	50	34.8	0.70	0.63
200433707	2	7/14/2004	3.1	200390007	Duroc	Clone	200346101	D	12	1			2	1	7/30/04	16	11.3	0.51	9/2/2004	1	50	34.0	0.67	0.62
200433708	2	7/14/2004	2.5	200390007	Duroc	Clone	200346101	D	11	1			2	1	7/30/04	16	10.1	0.48	9/2/2004	1	50	30.2	0.59	0.55
200433709	2	7/14/2004	2.2	200390007	Duroc	Clone	200346101	D	13	1			2	1	7/30/04	16	9.5	0.46	9/2/2004	1	50	32.2	0.67	0.60
200433710	1	7/14/2004	3.7	200390007	Duroc	Clone	200346101	D	11	1			3	1	7/30/04	16	12	0.52	9/2/2004	1	50	39.0	0.79	0.71
200433711	1	7/14/2004	3.5	200390007	Duroc	Clone	200346101	D	13	1			3	1	7/30/04	16	12.2	0.54	9/2/2004	1	50	35.8	0.69	0.65
200433712	1	7/14/2004	3.1	200390007	Duroc	Clone	200346101	D	13	1			3	1	7/30/04	16	12.6	0.59	9/2/2004	1	50	35.6	0.68	0.65
200433713	1	7/14/2004	4.4	200390007	Duroc	Clone	200346101	D	14	1	7/14/2004	OVRL		0						0				
200434201	2	7/15/2004	1.9	200398128	Duroc	Control	200346102	D	11	1			2	1	7/30/04	15	5.1	0.21	9/1/2004	1	48	23.6	0.56	0.45
200434202	1	7/15/2004	3.4	200398128	Duroc	Control	200346102	D	13	1			3	1	7/30/04	15	9.4	0.40	9/2/2004	1	49	36.4	0.79	0.67
200434203	1	7/15/2004	2.8	200398128	Duroc	Control	200346102	D	12	1			3	1	7/30/04	15	7.5	0.31	9/2/2004	1	49	27.4	0.59	0.50
200434204	1	7/15/2004	3.0	200398128	Duroc	Control	200346102	D	12	1			3	1	7/30/04	15	8.4	0.36	9/2/2004	1	49	26.6	0.54	0.48
200434205	1	7/15/2004	2.8	200398128	Duroc	Control	200346102	D	11	1			3	1	7/30/04	15	8.7	0.39	9/2/2004	1	49	33.6	0.73	0.63
200434206	1	7/15/2004	2.7	200398128	Duroc	Control	200346102	D	12	1			3	1	7/30/04	15	8.9	0.41	9/2/2004	1	49	36.2	0.80	0.68
200434207	1	7/15/2004	2.6	200398128	Duroc	Control	200346102	D	12	1			3	1	7/30/04	15	8.7	0.41	9/2/2004	1	49	30.6	0.64	0.57
200434208	1	7/15/2004	2.7	200398128	Duroc	Control	200346102	D	11	1			3	1	7/30/04	15	7.4	0.31	9/2/2004	1	49	35.2	0.82	0.66

Animal	Sex	Birth Date	Weight	Sire	Sire Genetics	Sire Treatment	Dam	Dam Sire Code	Nipples	Abnormality	Disposal Date	Disposal Reason	Sex (after Castration)	Live at weaning	Weaning Date	Age at Weaning	Weaning Weight	ADG at Weaning	Week 8 Weigh Date	Live at 8 weeks	Age at Week 8	Week 8 Weight	Week 8 ADG	Week 8 WDA
200434209	1	7/15/2004	2.3	200398128	Duroc	Control	200346102	D	13	1			3	1	7/30/04	15	7	0.31	9/2/2004	1	49	23.6	0.49	0.43
200434210	1	7/15/2004	2.0	200398128	Duroc	Control	200346102	D	12	1			3	1	7/30/04	15	7.1	0.34	9/2/2004	1	49	28.0	0.61	0.53
200434211	1	7/15/2004	2.4	200398128	Duroc	Control	200346102	D	12	1	7/15/2004	STLB		0						0				
200434212	2	7/15/2004	2.9	200398128	Duroc	Control	200346102	D	12	1	7/15/2004	STLB		0						0				
200434213	2	7/15/2004	2.2	200398128	Duroc	Control	200346102	D	12	1	7/15/2004	STLB		0						0				
200432901	2	7/13/2004	4.1	200395515	Duroc	Control	200346104	D	14	1			2	1	7/30/04	17	13	0.52	9/2/2004	1	51	39.8	0.79	0.70
200432902	2	7/13/2004	4.1	200395515	Duroc	Control	200346104	D	12	1			2	1	7/30/04	17	13.2	0.54	9/2/2004	1	51	39.0	0.76	0.68
200432903	2	7/13/2004	3.9	200395515	Duroc	Control	200346104	D	13	1			2	1	7/30/04	17	13.2	0.55	9/2/2004	1	51	44.0	0.91	0.79
200432904	2	7/13/2004	3.8	200395515	Duroc	Control	200346104	D	14	1			2	1	7/30/04	17	11.9	0.48	9/2/2004	1	51	35.8	0.70	0.63
200432905	2	7/13/2004	3.8	200395515	Duroc	Control	200346104	D	14	1			2	1	7/30/04	17	11.6	0.46	9/2/2004	1	51	35.6	0.71	0.62
200432906	2	7/13/2004	3.3	200395515	Duroc	Control	200346104	D	12	1			2	1	7/30/04	17	12.7	0.55	9/2/2004	1	51	39.0	0.77	0.70
200432907	1	7/13/2004	4.2	200395515	Duroc	Control	200346104	D	14	1			3	1	7/30/04	17	12.8	0.51	9/2/2004	1	51	36.6	0.70	0.64
200432908	1	7/13/2004	4.3	200395515	Duroc	Control	200346104	D	14	1			3	1	7/30/04	17	15.6	0.66	9/2/2004	1	51	43.8	0.83	0.77
200432909	1	7/13/2004	3.9	200395515	Duroc	Control	200346104	D	13	1			3	1	7/30/04	17	13.7	0.58	9/2/2004	1	51	34.2	0.60	0.59
200432910	1	7/13/2004	3.4	200395515	Duroc	Control	200346104	D	12	1			3	1	7/30/04	17	11.6	0.48	9/2/2004	1	51	43.6	0.94	0.79
200432911	1	7/13/2004	3.6	200395515	Duroc	Control	200346104	D	14	1			3	1	7/30/04	17	10.2	0.39	9/2/2004	1	51	37.2	0.79	0.66
200432912	1	7/13/2004	3.2	200395515	Duroc	Control	200346104	D	12	1			3	1	7/30/04	17	12.7	0.56	9/2/2004	1	51	35.6	0.67	0.64
200432913	1	7/13/2004	3.0	200395515	Duroc	Control	200346104	D	14	1			3	1	7/30/04	17	11.1	0.48	9/2/2004	1	51	32.6	0.63	0.58

Abnormality Codes:	
1	= None
4	= Atresia ani
6	= Spraddle Legs
99	= Other

Disposal Codes:	
DDFR	= Destroyed Deformed
DINJ	= Destroyed Injury
OVRL	= Overlay
STLB	= Still Born
UNK	= Unknown
WEAK	= Weak

Treatment	Genetics	Boar ID	# Piglets Born	Live at Birth	# Mummies	# Stillborn	Males Weaned	Females Weaned	Weaned in Litter	Weaned in Litter by Dam	8 weeks of Age
Clone	TX1	2	81	71	3	7	30	31	61	57	58
Clone	TX1	3	128	113	4	11	50	41	91	80	99
Clone	TX1	5	75	71	1	3	33	29	62	55	61
Clone	TX2	7	118	103	0	15	43	38	81	76	85
Control	TX1	H498	61	54	2	5	19	29	48	45	55
Control	TX2	25515	105	95	4	6	51	36	87	80	75
Control	TX2	18128	134	118	0	16	64	44	108	88	102
		Total	702	625	14	63	290	248	538	481	535

Treatment	Genetics	Boar ID	# Obs	Birth WT (lbs)	Nipple Count	Weaning WT (lbs)	ADG at Weaning (lbs)	8 wk weight	ADG at 8 wk (lbs/day)	WDA at 8 wk (lbs/day)	BW (kg)
Clone	TX1	2	78	3.41	13.77	11.50	0.44	38.62	0.77	0.65	1.55128
Clone	TX1	3	124	3.14	13.89	10.51	0.44	33.74	0.67	0.60	1.42595
Clone	TX1	5	74	3.39	13.82	11.05	0.43	37.93	0.76	0.65	1.54177
Clone	TX2	7	118	3.35	13.25	10.96	0.45	39.48	0.84	0.72	1.52388
Control	TX1	H498	59	3.33	14.00	10.68	0.40	33.00	0.65	0.57	1.51541
Control	TX2	25515	101	3.26	13.94	11.08	0.45	37.67	0.78	0.67	1.4829
Control	TX2	18128	134	3.09	13.14	10.70	0.42	38.92	0.85	0.69	1.40636

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
1	200430501	2	200190498	1/6/05	6	9/1/04	38.0	10/5/04	96.0	11/2/04	142.6	11/30/04	200.2	12/28/04	254.0	
2	200430502	2	200190498	1/3/05	6	9/1/04	36.8	10/5/04	95.6	11/2/04	156.6	11/30/04	206.6	12/28/04	270.8	
3	200430503	2	200190498	12/13/04	6	9/1/04	35.8	10/5/04	94.2	11/2/04	158.0	11/30/04	226.0			
4	200430504	2	200190498	1/12/05	6	9/1/04	28.4	10/5/04	72.6	11/2/04	128.4	11/30/04	183.2	12/28/04	242.0	
5	200430505	2	200190498	2/10/05	8	9/1/04	32.4	10/5/04	77.8	11/2/04	123.8	11/30/04	178.6	12/28/04	223.8	1/25/C
6	200430506	2	200190498	1/10/05	6	9/1/04	34.2	10/5/04	87.4	11/2/04	140.0	11/30/04	193.8	12/28/04	248.4	
7	200430507	1	200190498	1/20/05	6	9/1/04	32.4	10/5/04	84.4	11/2/04	130.2	11/30/04	183.2	12/28/04	235.6	
8	200430508	1	200190498	12/13/04	6	9/1/04	36.4	10/5/04	96.8	11/2/04	163.4	11/30/04	234.4			
9	200430509	1	200190498	1/3/05	6	9/1/04	27.2	10/5/04	70.4	11/2/04	133.2	11/30/04	195.4	12/28/04	258.8	
10	200430510	1	200190498	12/21/04	6	9/1/04	36.0	10/5/04	89.8	11/2/04	145.6	11/30/04	207.6			
11	200430601	2	200390005	12/14/04	3	9/1/04	33.8	10/5/04	63.2	11/2/04	98.0	11/30/04	132.0			
12	200430602	2	200390005	1/20/05	6	9/1/04	39.8	10/5/04	91.4	11/2/04	144.8	11/30/04	197.4	12/28/04	241.0	
13	200430603	2	200390005	2/2/05	6	9/1/04	35.6	10/5/04	87.4	11/2/04	139.0	11/30/04	182.4	12/28/04	196.2	1/25/C
14	200430604	1	200390005	12/13/04	6	9/1/04	48.4	10/5/04	112.2	11/2/04	181.2	11/30/04	252.6			
15	200430605	1	200390005	12/15/04	6	9/1/04	41.4	10/5/04	102.0	11/2/04	163.2	11/30/04	222.2			
16	200430606	1	200390005	12/29/04	6	9/1/04	32.6	10/5/04	83.2	11/2/04	133.2	11/30/04	196.4			
17	200430607	1	200390005	12/15/04	6	9/1/04	37.8	10/5/04	104.0	11/2/04	169.4	11/30/04	230.0			
18	200430608	1	200390005	1/3/05	6	9/1/04	40.8	10/5/04	97.8	11/2/04	153.8	11/30/04	203.6	12/28/04	268.0	
19	200430609	1	200390005	12/15/04	6	9/1/04	37.2	10/5/04	100.6	11/2/04	155.6	11/30/04	220.0			
20	200430610	1	200390005	1/10/05	6	9/1/04	37.8	10/5/04	86.2	11/2/04	131.8	11/30/04	189.6	12/28/04	246.4	
21	200430611	1	200390005	1/12/05	6	9/1/04	34.8	10/5/04	92.8	11/2/04	145.4	11/30/04	198.0	12/28/04	245.4	
22	200430701	2	200390002	1/10/05	6	9/1/04	33.4	10/5/04	81.2	11/2/04	132.6	11/30/04	187.2	12/28/04	248.6	
23	200430702	2	200390002	1/20/05	6	9/1/04	35.4	10/5/04	87.8	11/2/04	146.6	11/30/04	185.2	12/28/04	231.4	
24	200430703	2	200390002	12/20/04	6	9/1/04	38.0	10/5/04	97.4	11/2/04	154.6	11/30/04	213.0			
25	200430704	2	200390002	12/27/04	6	9/1/04	37.2	10/5/04	94.2	11/2/04	151.0	11/30/04	209.0			
26	200430705	2	200390002	12/15/04	6	9/1/04	38.6	10/5/04	100.0	11/2/04	157.4	11/30/04	221.0			
27	200430706	1	200390002	12/13/04	6	9/1/04	40.0	10/5/04	111.8	11/2/04	177.0	11/30/04	244.2			
28	200430707	1	200390002	12/13/04	6	9/1/04	49.8	10/5/04	108.6	11/2/04	175.0	11/30/04	239.8			
29	200430708	1	200390002	12/15/04	6	9/1/04	41.0	10/5/04	96.0	11/2/04	150.4	11/30/04	220.4			
30	200430709	1	200390002	12/13/04	6	9/1/04	45.6	10/5/04	110.0	11/2/04	175.0	11/30/04	239.4			
31	200430710	1	200390002	12/13/04	6	9/1/04	44.4	10/5/04	104.4	11/2/04	171.2	11/30/04	252.2			
32	200430711	1	200390002	12/13/04	6	9/1/04	36.8	10/5/04	99.6	11/2/04	168.8	11/30/04	237.0			
33	200430801	2	200390005	1/20/05	6	9/1/04	37.6	10/5/04	90.8	11/2/04	133.4	11/30/04	179.0	12/28/04	233.8	
34	200430802	2	200390005	12/27/04	6	9/1/04	45.0	10/5/04	95.2	11/2/04	152.2	11/30/04	209.8			
35	200430803	1	200390005	12/29/04	6	9/1/04	35.2	10/5/04	79.4	11/2/04	137.0	11/30/04	198.6			
36	200430804	1	200390005	12/13/04	6	9/1/04	50.4	10/5/04	98.0	11/2/04	165.8	11/30/04	238.6			
37	200430805	1	200390005	12/13/04	6	9/1/04	43.6	10/5/04	93.4	11/2/04	152.8	11/30/04	231.8			
38	200430902	1	200390003	12/13/04	6	9/1/04	44.2	10/5/04	105.0	11/2/04	168.4	11/30/04	230.4			
39	200430903	1	200390003	12/20/04	6	9/1/04	43.6	10/5/04	97.6	11/2/04	153.6	11/30/04	217.0			

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
40	200430905	1	200390003	12/13/04	6	9/1/04	48.8	10/5/04	113.6	11/2/04	175.2	11/30/04	239.0			
41	200430906	1	200390003	12/13/04	6	9/1/04	47.6	10/5/04	101.4	11/2/04	164.6	11/30/04	234.2			
42	200431001	2	200390002	11/5/04	3	9/1/04	45.6	10/5/04	103.4							
43	200431002	2	200390002	1/12/05	6	9/1/04	33.4	10/5/04	76.8	11/2/04	129.0	11/30/04	179.8	12/28/04	240.8	
44	200431003	2	200390002	12/20/04	6	9/1/04	39.8	10/5/04	93.2	11/2/04	146.2	11/30/04	210.4			
45	200431004	2	200390002	1/12/05	6	9/1/04	29.6	10/5/04	71.2	11/2/04	115.2	11/30/04	172.8	12/28/04	236.8	
46	200431005	2	200390002	1/20/05	6	9/1/04	35.4	10/5/04	77.4	11/2/04	128.6	11/30/04	184.6	12/28/04	233.0	
47	200431006	1	200390002	12/13/04	6	9/1/04	50.8	10/5/04	110.6	11/2/04	179.0	11/30/04	243.8			
48	200431007	1	200390002	1/3/05	6	9/1/04	36.0	10/5/04	82.0	11/2/04	134.8	11/30/04	189.2	12/28/04	256.2	
49	200431008	1	200390002	12/15/04	6	9/1/04	40.6	10/5/04	100.8	11/2/04	163.0	11/30/04	222.2			
50	200431009	1	200390002	12/13/04	6	9/1/04	46.4	10/5/04	106.4	11/2/04	171.0	11/30/04	236.8			
51	200431010	1	200390002	1/12/05	6	9/1/04	32.0	10/5/04	73.8	11/2/04	126.2	11/30/04	185.2	12/28/04	241.4	
52	200431011	1	200390002	12/20/04	6	9/1/04	40.6	10/5/04	100.4	11/2/04	164.6	11/30/04	222.2			
53	200431012	1	200390002	1/3/05	6	9/1/04	40.4	10/5/04	93.8	11/2/04	152.6	11/30/04	204.0	12/28/04	274.0	
54	200431401	2	200390005	12/27/04	6	9/1/04	41.2	10/5/04	86.0	11/2/04	135.6	11/30/04	202.6			
55	200431402	2	200390005	1/3/05	6	9/1/04	45.2	10/5/04	99.8	11/2/04	152.2	11/30/04	206.0	12/28/04	260.2	
56	200431403	2	200390005	12/27/04	6	9/1/04	33.4	10/5/04	78.4	11/2/04	134.2	11/30/04	199.6			
57	200431404	2	200390005	12/13/04	6	9/1/04	39.0	10/5/04	91.4	11/2/04	152.0	11/30/04	231.0			
58	200431405	2	200390005	12/15/04	6	9/1/04	43.8	10/5/04	99.4	11/2/04	150.2	11/30/04	218.2			
59	200431406	2	200390005	12/20/04	6	9/1/04	32.4	10/5/04	79.2	11/2/04	136.8	11/30/04	207.6			
60	200431407	2	200390005	12/15/04	6	9/1/04	39.8	10/5/04	101.4	11/2/04	164.2	11/30/04	226.6			
61	200431408	2	200390005	12/13/04	6	9/1/04	41.4	10/5/04	106.4	11/2/04	167.2	11/30/04	234.4			
62	200431409	5	200390005	1/3/05	6	9/1/04	46.4	10/5/04	88.2	11/2/04	153.2	12/7/04	232.4			
63	200431410	1	200390005	1/6/05	6	9/1/04	32.4	10/5/04	72.2	11/2/04	135.0	11/30/04	186.8	12/28/04	251.2	
64	200431411	1	200390005	12/13/04	6	9/1/04	38.6	10/5/04	93.4	11/2/04	158.0	11/30/04	226.2			
65	200431501	2	200390002	12/15/04	6	9/1/04	38.4	10/5/04	95.4	11/2/04	156.8	11/30/04	224.8			
66	200431502	2	200390002	12/13/04	6	9/1/04	46.2	10/5/04	108.2	11/2/04	166.6	11/30/04	238.8			
67	200431503	2	200390002	1/18/05	6	9/1/04	42.0	10/5/04	89.0	11/2/04	132.6	11/30/04	192.6	12/28/04	240.2	
68	200431504	1	200390002	12/15/04	6	9/1/04	46.2	10/5/04	100.0	11/2/04	159.0	11/30/04	220.0			
69	200431505	1	200390002	12/13/04	6	9/1/04	41.0	10/5/04	99.4	11/2/04	158.8	11/30/04	231.2			
70	200431506	1	200390002	1/6/05	6	9/1/04	44.0	10/5/04	86.8	11/2/04	136.8	11/30/04	184.6	12/28/04	249.4	
71	200431601	2	200390002	1/12/05	6	9/1/04	33.6	10/5/04	81.4	11/2/04	129.2	11/30/04	188.2	12/28/04	242.0	
72	200431602	2	200390002	1/20/05	6	9/1/04	38.0	10/5/04	92.2	11/2/04	142.8	11/30/04	194.2	12/28/04	239.6	
73	200431603	2	200390002	1/6/05	6	9/1/04	35.8	10/5/04	85.4	11/2/04	139.2	11/30/04	191.8	12/28/04	251.2	
74	200431604	2	200390002	12/20/04	6	9/1/04	36.2	10/5/04	94.6	11/2/04	155.4	11/30/04	215.4			
75	200431605	2	200390002	1/12/05	6	9/1/04	31.4	10/5/04	80.2	11/2/04	137.2	11/30/04	190.2	12/28/04	243.4	
76	200431606	1	200390002	12/15/04	6	9/1/04	40.8	10/5/04	96.6	11/2/04	160.6	11/30/04	224.4			
77	200431607	1	200390002	12/29/04	6	9/1/04	36.0	10/5/04	92.6	11/2/04	151.6	11/30/04	207.2			
78	200431608	1	200390002	12/13/04	6	9/1/04	45.4	10/5/04	94.4	11/2/04	162.2	11/30/04	230.4			

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
79	200431610	1	200390002	12/20/04	6	9/1/04	36.0	10/5/04	91.0	11/2/04	149.6	11/30/04	212.8			
80	200431701	2	200390005	1/10/05	6	9/1/04	33.8	10/5/04	81.6	11/2/04	137.0	11/30/04	195.8	12/28/04	249.8	
81	200431702	1	200390005	1/12/05	6	9/1/04	36.4	10/5/04	84.0	11/2/04	136.8	11/30/04	188.2	12/28/04	245.4	
82	200431703	1	200390005	12/27/04	6	9/1/04	37.8	10/5/04	90.6	11/2/04	135.6	11/30/04	199.4			
83	200431704	1	200390005	12/27/04	6	9/1/04	38.0	10/5/04	93.8	11/2/04	149.4	11/30/04	208.2			
84	200431706	1	200390005	12/27/04	6	9/1/04	39.0	10/5/04	92.0	11/2/04	145.6	11/30/04	206.0			
85	200431708	1	200390005	1/18/05	6	9/1/04	27.6	10/5/04	77.4	11/2/04	119.4	11/30/04	168.6	12/28/04	232.0	
86	200431801	2	200398128	12/27/04	6	9/1/04	37.4	10/5/04	88.6	11/2/04	136.0	11/30/04	202.2			
87	200431802	2	200398128	12/15/04	6	9/1/04	44.0	10/5/04	96.8	11/2/04	156.2	11/30/04	222.8			
88	200431803	2	200398128	1/20/05	6	9/1/04	24.4	10/5/04	68.8	11/2/04	116.4	11/30/04	174.8	12/28/04	226.8	
89	200431804	1	200398128	2/2/05	6	9/1/04	28.2	10/5/04	59.6	11/2/04	104.8	11/30/04	145.0	12/28/04	169.4	1/25/C
90	200431805	1	200398128	12/21/04	6	9/1/04	32.6	10/5/04	77.8	11/2/04	142.6	11/30/04	212.0			
91	200432001	2	200390002	1/6/05	6	9/2/04	42.2	10/5/04	87.6	11/2/04	140.0	11/30/04	195.2	12/28/04	253.2	
92	200432002	2	200390002	12/29/04	6	9/2/04	42.6	10/5/04	86.6	11/2/04	129.4	11/30/04	195.0			
93	200432003	2	200390002	1/18/05	6	9/2/04	40.6	10/5/04	86.2	11/2/04	133.2	11/30/04	183.8	12/28/04	238.0	
94	200432004	1	200390002	12/20/04	6	9/2/04	45.8	10/5/04	101.8	11/2/04	158.2	11/30/04	218.4			
95	200432005	1	200390002	1/6/05	6	9/2/04	39.8	10/5/04	90.6	11/2/04	141.8	11/30/04	197.0			
96	200432006	1	200390002	12/29/04	6	9/2/04	40.4	10/5/04	97.4	11/2/04	154.0	11/30/04	208.8			
97	200432007	1	200390002	1/3/05	6	9/2/04	41.4	10/5/04	91.4	11/2/04	138.6	11/30/04	195.2	12/28/04	262.8	
98	200432008	1	200390002	1/18/05	6	9/2/04	30.0	10/5/04	71.0	11/2/04	124.2	11/30/04	181.4	12/28/04	236.2	
99	200432101	2	200390003	12/29/04	6	9/1/04	38.4	10/5/04	85.0	11/2/04	138.2	11/30/04	199.2			
100	200432102	2	200390003	1/6/05	6	9/1/04	32.4	10/5/04	87.6	11/2/04	136.6	11/30/04	190.2	12/28/04	250.4	
101	200432103	2	200390003	12/20/04	6	9/1/04	35.2	10/5/04	94.6	11/2/04	158.4	11/30/04	218.6			
102	200432104	1	200390003	1/3/05	6	9/1/04	34.4	10/5/04	89.8	11/2/04	146.8	11/30/04	200.8	12/28/04	261.0	
103	200432105	1	200390003	12/27/04	6	9/1/04	34.8	10/5/04	89.4	11/2/04	148.4	11/30/04	208.4			
104	200432106	1	200390003	12/20/04	7	9/1/04	40.0	10/5/04	94.6	11/2/04	148.8	11/30/04	213.4			
105	200432108	1	200390003	12/20/04	6	9/1/04	38.0	10/5/04	90.6	11/2/04	147.6	11/30/04	210.8			
106	200432109	1	200390003	1/3/05	7	9/1/04	32.6	10/5/04	83.4	11/2/04	140.6	11/30/04	196.0	12/28/04	257.2	
107	200432301	2	200390003	1/12/05	7	9/2/04	32.8	10/5/04	79.4	11/2/04	132.8	11/30/04	190.6	12/28/04	245.8	
108	200432302	2	200390003	12/29/04	6	9/2/04	28.6	10/5/04	74.4	11/2/04	131.2	11/30/04	196.4			
109	200432303	2	200390003	1/12/05	7	9/2/04	35.8	10/5/04	87.6	11/2/04	141.4	11/30/04	194.2	12/28/04	244.6	
110	200432304	2	200390003	1/18/05	6	9/2/04	37.2	10/5/04	87.8	11/2/04	144.8	11/30/04	193.0	12/28/04	240.8	
111	200432305	2	200390003	2/10/05	8	9/2/04	28.2	10/5/04	72.0	11/2/04	129.4	11/30/04	171.8	12/28/04	216.4	1/25/C
112	200432306	2	200390003	2/2/05	6	9/2/04	26.8	10/5/04	73.6	11/2/04	125.8	11/30/04	182.6	12/28/04	221.0	1/25/C
113	200432307	1	200390003	1/6/05	7	9/2/04	30.6	10/5/04	72.4	11/2/04	123.2	11/30/04	187.6	12/28/04	250.2	
114	200432308	1	200390003	12/20/04	6	9/2/04	38.6	10/5/04	85.8	11/2/04	149.0	11/30/04	214.4			
115	200432309	1	200390003	2/10/05	8	9/2/04	22.0	10/5/04	59.4	11/2/04	108.6	11/30/04	170.0	12/28/04	226.2	
116	200432310	1	200390003	12/20/04	6	9/2/04	34.4	10/5/04	90.6	11/2/04	153.0	11/30/04	211.6			
117	200432311	1	200390003	1/6/05	7	9/2/04	29.8	10/5/04	80.4	11/2/04	136.0	11/30/04	187.4	12/28/04	252.4	

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
118	200432312	1	200390003	1/6/05	6	9/2/04	25.0	10/5/04	70.2	11/2/04	117.2	11/30/04	180.6	12/28/04	249.4	
119	200432401	2	200390002	1/6/05	6	9/2/04	33.8	10/5/04	78.2	11/2/04	130.2	11/30/04	182.8	12/28/04	248.8	
120	200432402	2	200390002	12/27/04	6	9/2/04	37.0	10/5/04	81.0	11/2/04	131.4	11/30/04	197.6			
121	200432403	2	200390002	1/12/05	6	9/2/04	32.8	10/5/04	73.4	11/2/04	117.6	11/30/04	176.2	12/28/04	239.8	
122	200432404	2	200390002	12/20/04	6	9/2/04	37.9	10/5/04	93.8	11/2/04	151.4	11/30/04	210.8			
123	200432405	1	200390002	12/13/04	6	9/2/04	43.6	10/5/04	101.8	11/2/04	157.0	11/30/04	226.6			
124	200432406	1	200390002	12/20/04	6	9/2/04	42.8	10/5/04	102.2	11/2/04	165.0	11/30/04	220.8			
125	200432407	1	200390002	1/10/05	6	9/2/04	33.8	10/5/04	80.4	11/2/04	137.6	11/30/04	191.6	12/28/04	248.4	
126	200432501	2	200390003	12/29/04	6	9/2/04	47.6	10/5/04	106.4	11/2/04	160.2	11/30/04	211.2			
127	200432502	2	200390003	1/6/05	7	9/2/04	41.2	10/5/04	88.4	11/2/04	138.4	11/30/04	196.8	12/28/04	255.6	
128	200432503	2	200390003	12/29/04	6	9/2/04	43.9	10/5/04	97.4	11/2/04	152.6	11/30/04	207.8			
129	200432504	2	200390003	1/6/05	6	9/2/04	41.0	10/5/04	91.2	11/2/04	143.0	11/30/04	197.0	12/28/04	252.0	
130	200432506	2	200390003	1/26/05	6	9/2/04	25.0	10/5/04	59.8	11/2/04	105.4	11/30/04	156.0	12/28/04	211.6	
131	200432507	1	200390003	1/18/05	6	9/2/04	39.2	10/5/04	91.6	11/2/04	143.4	11/30/04	194.2	12/28/04	243.8	
132	200432508	1	200390003	9/3/04	3	9/2/04	14.4									
133	200432601	2	200398128	12/21/04	6	9/2/04	38.6	10/5/04	84.2	11/2/04	143.8	11/30/04	207.2			
134	200432602	2	200398128	12/21/04	6	9/2/04	34.8	10/5/04	88.2	11/2/04	135.8	11/30/04	208.6			
135	200432603	2	200398128	1/4/05	4	9/2/04	39.0	10/5/04	85.0	11/2/04	143.6	11/30/04	204.2			
136	200432604	2	200398128	12/29/04	6	9/2/04	40.0	10/5/04	85.8	11/2/04	143.6	11/30/04	202.8			
137	200432605	2	200398128	1/14/05	4	9/2/04	31.6	10/5/04	74.4	11/2/04	130.4	11/30/04	190.0	12/28/04	249.8	
138	200432607	1	200398128	12/21/04	6	9/2/04	37.2	10/5/04	94.8	11/2/04	158.8	11/30/04	219.4			
139	200432608	1	200398128	1/4/05	4	9/2/04	32.2	10/5/04	73.2	11/2/04	134.6	11/30/04	203.2			
140	200432609	1	200398128	12/15/04	6	9/2/04	37.8	10/5/04	89.6	11/2/04	143.4	11/30/04	216.6			
141	200432610	1	200398128	12/21/04	6	9/2/04	32.0	10/5/04	82.4	11/2/04	142.8	11/30/04	210.4			
142	200432701	2	200390003	1/11/05	3	9/2/04	42.4	10/5/04	89.6	11/2/04	145.4	11/30/04	179.2	12/28/04	170.2	
143	200432702	2	200390003	12/27/04	6	9/2/04	37.6	10/5/04	89.0	11/2/04	149.8	11/30/04	209.6			
144	200432703	1	200390003	2/10/05	8	9/2/04	37.3	10/5/04	82.8	11/2/04	137.8	11/30/04	194.6	12/28/04	243.0	
145	200432704	1	200390003	12/20/04	6	9/2/04	31.8	10/5/04	87.8	11/2/04	143.0	11/30/04	208.2			
146	200432705	1	200390003	12/20/04	6	9/2/04	37.2	10/5/04	88.0	11/2/04	149.0	11/30/04	214.0			
147	200432706	1	200390003	12/20/04	6	9/2/04	38.8	10/5/04	85.0	11/2/04	147.6	11/30/04	211.0			
148	200432901	2	200395515	1/12/05	6	9/2/04	39.8	10/5/04	91.4	11/2/04	135.4	11/30/04	188.0	12/28/04	243.6	
149	200432902	2	200395515	10/20/04	3	9/2/04	39.0	10/5/04	87.2							
150	200432903	2	200395515	1/4/05	4	9/2/04	44.0	10/5/04	104.2	11/2/04	151.6	11/30/04	217.0			
151	200432904	2	200395515	1/10/05	6	9/2/04	35.8	10/5/04	84.6	11/2/04	134.8	11/30/04	185.8	12/28/04	247.6	
152	200432905	2	200395515	1/4/05	4	9/2/04	35.6	10/5/04	92.2	11/2/04	140.8	11/30/04	207.4			
153	200432906	2	200395515	1/10/05	6	9/2/04	39.0	10/5/04	84.2	11/2/04	141.4	11/30/04	190.8	12/28/04	249.2	
154	200432907	1	200395515	12/21/04	6	9/2/04	36.6	10/5/04	86.2	11/2/04	146.4	11/30/04	212.0			
155	200432908	1	200395515	1/4/05	4	9/2/04	43.8	10/5/04	99.2	11/2/04	155.2	11/30/04	219.6			
156	200432909	1	200395515	12/29/04	6	9/2/04	34.2	10/5/04	90.0	11/2/04	132.6	11/30/04	196.0			

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
157	200432910	1	200395515	1/4/05	4	9/2/04	43.6	10/5/04	98.2	11/2/04	158.0	11/30/04	232.6			
158	200432911	1	200395515	12/15/04	6	9/2/04	37.2	10/5/04	87.0	11/2/04	146.8	11/30/04	217.8			
159	200432912	1	200395515	1/4/05	4	9/2/04	35.6	10/5/04	80.8	11/2/04	141.6	11/30/04	206.4			
160	200432913	1	200395515	12/21/04	6	9/2/04	32.6	10/5/04	79.4	11/2/04	129.4	11/30/04	200.4			
161	200433001	2	200390003	1/20/05	6	9/2/04	34.4	10/5/04	97.8	11/2/04	135.2	11/30/04	183.6	12/28/04	234.6	
162	200433002	2	200390003	1/10/05	6	9/2/04	32.4	10/5/04	76.4	11/2/04	127.4	11/30/04	183.4	12/28/04	244.4	
163	200433003	2	200390003	1/6/05	6	9/2/04	29.6	10/5/04	68.2	11/2/04	122.2	11/30/04	182.8	12/28/04	248.8	
164	200433004	1	200390003	2/10/05	8	9/2/04	34.6	10/5/04	83.4	11/2/04	139.6	11/30/04	186.8	12/28/04	237.4	
165	200433005	1	200390003	1/3/05	7	9/2/04	33.4	10/5/04	80.6	11/2/04	141.8	11/30/04	194.6	12/28/04	259.2	
166	200433006	1	200390003	1/12/05	7	9/2/04	28.8	10/5/04	82.0	11/2/04	136.0	11/30/04	184.8	12/28/04	242.0	
167	200433007	2	200390003	1/18/05	6	9/2/04	31.4	10/5/04	75.6	11/2/04	123.2	11/30/04	177.8	12/28/04	238.4	
168	200433008	1	200390003	1/12/05	7	9/2/04	29.0	10/5/04	80.6	11/2/04	136.8	11/30/04	192.2	12/28/04	245.6	
169	200433010	1	200390003	1/20/05	6	9/1/04	24.6	10/12/04	77.8	11/9/04	130.8	12/7/04	183.2	1/4/05	237.2	
170	200433011	1	200390003	1/26/05	6	9/1/04	18.8	10/12/04	67.8	11/9/04	108.0	12/7/04	166.8	1/4/05	221.8	
171	200433201	2	200390005	12/15/04	6	9/2/04	39.6	10/5/04	93.0	11/2/04	157.0	11/30/04	223.6			
172	200433202	2	200390005	1/10/05	6	9/2/04	31.2	10/5/04	70.8	11/2/04	124.4	11/30/04	181.4	12/28/04	228.2	
173	200433203	2	200390005	1/3/05	6	9/2/04	32.6	10/5/04	79.0	11/2/04	140.8	11/30/04	199.4	12/28/04	267.6	
174	200433204	2	200390005	1/26/05	6	9/2/04	27.6	10/5/04	71.6	11/2/04	122.6	11/30/04	179.0	12/28/04	224.0	
175	200433205	1	200390005	12/20/04	6	9/2/04	36.4	10/5/04	89.4	11/2/04	153.8	11/30/04	215.4			
176	200433206	1	200390005	12/27/04	6	9/2/04	34.2	10/5/04	83.6	11/2/04	144.4	11/30/04	204.4			
177	200433207	1	200390005	12/29/04	6	9/2/04	30.6	10/5/04	79.2	11/2/04	138.2	11/30/04	198.8			
178	200433208	1	200390005	1/20/05	6	9/2/04	31.8	10/5/04	84.4	11/2/04	135.0	11/30/04	185.4	12/28/04	232.8	
179	200433301	2	200190498	12/15/04	6	9/2/04	33.4	10/5/04	92.4	11/2/04	153.8	11/30/04	218.2			
180	200433302	2	200190498	1/18/05	6	9/2/04	23.4	10/5/04	83.2	11/2/04	128.8	11/30/04	190.6	12/28/04	246.6	
181	200433303	2	200190498	1/18/05	6	9/2/04	22.0	10/5/04	70.8	11/2/04	123.8	11/30/04	183.2	12/28/04	239.0	
182	200433304	2	200190498	2/10/05	8	9/1/04	18.8	10/12/04	69.4	11/9/04	115.0	12/7/04	182.6	1/4/05	232.4	
183	200433305	2	200190498	2/10/05	8	9/2/04	19.8	10/5/04	65.2	11/2/04	115.4	11/30/04	167.0	12/28/04	219.8	
184	200433306	1	200190498	12/21/04	6	9/2/04	35.2	10/5/04	87.8	11/2/04	154.4	11/30/04	215.0			
185	200433307	1	200190498	12/21/04	6	9/2/04	34.6	10/5/04	97.0	11/2/04	154.2	11/30/04	216.0			
186	200433401	2	200190498	1/20/05	6	9/2/04	31.2	10/5/04	81.6	11/2/04	130.4	11/30/04	176.2	12/28/04	228.6	
187	200433402	2	200190498	1/3/05	6	9/2/04	32.8	10/5/04	89.6	11/2/04	144.8	11/30/04	195.0	12/28/04	260.8	
188	200433403	2	200190498	1/3/05	6	9/2/04	30.0	10/5/04	82.2	11/2/04	139.0	11/30/04	193.2	12/28/04	264.4	
189	200433404	2	200190498	12/29/04	6	9/2/04	32.8	10/5/04	88.4	11/2/04	136.8	11/30/04	198.8			
190	200433405	2	200190498	12/21/04	6	9/2/04	33.6	10/5/04	95.4	11/2/04	146.8	11/30/04	210.0			
191	200433406	2	200190498	1/18/05	6	9/2/04	28.6	10/5/04	86.0	11/2/04	132.2	11/30/04	183.2	12/28/04	240.2	
192	200433407	2	200190498	2/10/05	8	9/1/04	23.2	10/12/04	74.4	11/9/04	123.6	12/7/04	174.6	1/4/05	218.2	
193	200433408	1	200190498	12/21/04	6	9/2/04	38.9	10/5/04	96.0	11/2/04	156.4	11/30/04	215.0			
194	200433409	1	200190498	12/13/04	6	9/2/04	44.6	10/5/04	98.4	11/2/04	164.4	11/30/04	231.0			
195	200433501	2	200190498	1/3/05	6	9/2/04	28.8	10/12/04	81.0	11/9/04	129.8	12/7/04	201.4			

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
196	200433502	2	200190498	1/6/05	6	9/2/04	35.8	10/5/04	84.0	11/2/04	128.6	11/30/04	180.0	12/28/04	251.6	
197	200433503	2	200190498	1/6/05	6	9/2/04	26.4	10/5/04	70.8	11/2/04	121.0	11/30/04	176.8	12/28/04	246.6	
198	200433504	2	200190498	1/3/05	6	9/2/04	32.9	10/5/04	86.0	11/2/04	130.6	11/30/04	184.4	12/28/04	256.8	
199	200433505	2	200190498	12/21/04	6	9/2/04	33.9	10/5/04	93.0	11/2/04	148.6	11/30/04	215.0			
200	200433506	2	200190498	1/10/05	6	9/2/04	28.8	10/5/04	76.8	11/2/04	127.2	11/30/04	189.2	12/28/04	248.6	
201	200433507	2	200190498	1/6/05	6	9/2/04	25.8	10/5/04	72.8	11/2/04	120.8	11/30/04	184.8	12/28/04	252.4	
202	200433509	1	200190498	12/13/04	6	9/2/04	37.6	10/5/04	103.2	11/2/04	169.6	11/30/04	242.2			
203	200433510	1	200190498	12/13/04	6	9/2/04	35.4	10/5/04	96.8	11/2/04	166.6	11/30/04	243.6			
204	200433511	1	200190498	12/21/04	6	9/2/04	31.2	10/5/04	80.6	11/2/04	139.4	11/30/04	209.2			
205	200433512	1	200190498	12/21/04	6	9/2/04	30.4	10/5/04	85.2	11/2/04	146.4	11/30/04	208.6			
206	200433601	2	200390003	12/15/04	6	9/2/04	45.0	10/5/04	109.4	11/2/04	156.2	11/30/04	219.8			
207	200433602	2	200390003	12/27/04	6	9/2/04	44.0	10/5/04	106.4	11/2/04	157.8	11/30/04	213.0			
208	200433603	2	200390003	12/20/04	6	9/2/04	41.8	10/5/04	100.8	11/2/04	157.8	11/30/04	215.6			
209	200433604	2	200390003	12/29/04	6	9/2/04	36.8	10/5/04	100.8	11/2/04	145.6	11/30/04	204.6			
210	200433605	1	200390003	12/13/04	6	9/2/04	51.0	10/5/04	111.4	11/2/04	174.6	11/30/04	238.4			
211	200433701	2	200390007	1/3/05	7	9/2/04	35.0	10/5/04	89.8	11/2/04	141.2	11/30/04	196.4	12/28/04	260.0	
212	200433702	2	200390007	12/20/04	7	9/2/04	38.4	10/5/04	91.2	11/2/04	151.6	11/30/04	215.6			
213	200433703	2	200390007	12/29/04	7	9/2/04	38.6	10/5/04	95.6	11/2/04	148.2	11/30/04	205.6			
214	200433704	2	200390007	12/29/04	7	9/2/04	36.8	10/5/04	89.4	11/2/04	146.4	11/30/04	204.6			
215	200433705	2	200390007	2/10/05	8	9/2/04	34.6	10/5/04	80.8	11/2/04	130.2	11/30/04	178.2	12/28/04	223.6	1/25/C
216	200433706	2	200390007	1/12/05	7	9/2/04	34.8	10/5/04	85.0	11/2/04	134.4	11/30/04	183.0	12/28/04	243.4	
217	200433707	2	200390007	1/20/05	7	9/2/04	34.0	10/5/04	81.2	11/2/04	131.0	11/30/04	190.0	12/28/04	241.8	
218	200433708	2	200390007	2/2/05	6	9/2/04	30.2	10/5/04	69.4	11/2/04	98.6	11/30/04	143.0	12/28/04	189.2	1/25/C
219	200433709	2	200390007	1/18/05	6	9/2/04	32.2	10/5/04	75.8	11/2/04	124.6	11/30/04	179.2	12/28/04	238.0	
220	200433710	1	200390007	12/29/04	6	9/2/04	39.0	10/5/04	92.4	11/2/04	151.8	11/30/04	206.6			
221	200433711	1	200390007	12/20/04	6	9/2/04	35.8	10/5/04	90.4	11/2/04	147.8	11/30/04	210.8			
222	200433712	1	200390007	1/3/05	6	9/2/04	35.6	10/5/04	90.0	11/2/04	139.6	11/30/04	195.8	12/28/04	260.6	
223	200433801	2	200390007	1/20/05	6	9/2/04	43.0	10/5/04	102.2	11/2/04	156.8	11/30/04	191.2	12/28/04	236.0	
224	200433802	2	200390007	1/20/05	6	9/2/04	33.4	10/5/04	75.2	11/2/04	126.8	11/30/04	182.8	12/28/04	234.0	
225	200433803	2	200390007	12/27/04	6	9/2/04	37.6	10/5/04	91.0	11/2/04	148.0	11/30/04	206.8			
226	200433804	2	200390007	12/29/04	6	9/2/04	37.4	10/5/04	91.4	11/2/04	145.6	11/30/04	203.6			
227	200433806	1	200390007	1/3/05	6	9/2/04	35.4	10/5/04	61.6	11/2/04	112.8	11/30/04	178.6	12/28/04	254.8	
228	200433807	1	200390007	12/13/04	6	9/2/04	47.6	10/5/04	103.8	11/2/04	170.6	11/30/04	234.2			
229	200433808	1	200390007	12/29/04	6	9/2/04	39.8	10/5/04	73.6	11/2/04	119.0	11/30/04	189.4			
230	200433809	1	200390007	12/27/04	6	9/2/04	28.2	10/5/04	80.4	11/2/04	144.2	11/30/04	207.2			
231	200434001	2	200398128	12/15/04	6	9/2/04	36.6	10/5/04	89.0	11/2/04	145.4	11/30/04	215.0			
232	200434002	2	200398128	1/4/05	4	9/2/04	42.6	10/5/04	89.0	11/2/04	150.2	11/30/04	214.4			
233	200434003	2	200398128	12/21/04	6	9/2/04	35.0	10/12/04	93.4	11/9/04	149.2	12/7/04	217.8			
234	200434004	2	200398128	12/29/04	6	9/2/04	40.0	10/12/04	102.4	11/9/04	160.4	12/7/04	219.0			

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
235	200434005	2	200398128	12/21/04	6	9/2/04	32.4	10/12/04	99.0	11/9/04	156.0	12/7/04	221.8			
236	200434006	1	200398128	1/12/05	6	9/2/04	34.2	10/5/04	71.4	11/2/04	126.0	11/30/04	187.6	12/28/04	245.2	
237	200434007	1	200398128	12/27/04	6	9/2/04	35.0	10/5/04	88.2	11/2/04	135.8	11/30/04	201.8			
238	200434008	1	200398128	1/6/05	6	9/2/04	40.4	10/5/04	84.8	11/2/04	132.6	11/30/04	192.0	12/28/04	253.8	
239	200434009	1	200398128	1/3/05	6	9/2/04	37.6	10/5/04	71.2	11/2/04	117.0	11/30/04	181.8	12/28/04	254.2	
240	200434101	2	200390003	1/18/05	6	9/2/04	22.0	10/5/04	68.2	11/2/04	120.4	11/30/04	175.2	12/28/04	233.2	
241	200434102	2	200390003	2/2/05	6	9/2/04	25.4	10/5/04	71.0	11/2/04	115.8	11/30/04	166.6	12/28/04	209.8	1/25/C
242	200434103	2	200390003	2/10/05	8	9/2/04	20.8	10/5/04	63.4	11/2/04	106.6	11/30/04	146.6	12/28/04	200.4	1/25/C
243	200434104	2	200390003	1/20/05	6	9/2/04	29.4	10/5/04	79.8	11/2/04	132.6	11/30/04	179.2	12/28/04	230.8	
244	200434105	2	200390003	2/2/05	6	9/2/04	27.0	10/5/04	66.0	11/2/04	114.0	11/30/04	161.0	12/28/04	207.8	1/25/C
245	200434106	1	200390003	1/12/05	7	9/2/04	30.6	10/5/04	73.6	11/2/04	123.4	11/30/04	183.6	12/28/04	242.6	
246	200434107	1	200390003	1/20/05	6	9/2/04	27.9	10/5/04	72.8	11/2/04	117.0	11/30/04	170.0	12/28/04	224.6	
247	200434108	1	200390003	1/12/05	7	9/2/04	21.4	10/5/04	64.2	11/2/04	113.0	11/30/04	173.8	12/28/04	237.8	
248	200434109	1	200390003	1/20/05	6	9/2/04	30.9	10/5/04	79.4	11/2/04	131.4	11/30/04	173.8	12/28/04	226.2	
249	200434110	1	200390003	1/12/05	7	9/2/04	27.2	10/5/04	73.2	11/2/04	133.0	11/30/04	188.6	12/28/04	243.0	
250	200434111	1	200390003	1/26/05	6	9/2/04	25.4	10/5/04	62.2	11/2/04	102.8	11/30/04	162.0	12/28/04	217.4	
251	200434112	1	200390003	1/26/05	7	9/2/04	22.2	10/5/04	61.8	11/2/04	106.6	11/30/04	164.2	12/28/04	222.6	
252	200434113	1	200390003	1/26/05	6	9/1/04	16.2	10/12/04	63.4	11/9/04	100.8	12/7/04	162.8	1/4/05	222.0	
253	200434114	1	200390003	1/26/05	7	9/2/04	20.2	10/5/04	57.6	11/2/04	94.4	11/30/04	144.4	12/28/04	202.6	
254	200434201	2	200398128	12/29/04	6	9/1/04	23.6	10/12/04	91.4	11/9/04	138.4	12/7/04	207.4			
255	200434202	1	200398128	12/15/04	6	9/2/04	36.4	10/5/04	88.4	11/2/04	145.2	11/30/04	219.8			
256	200434203	1	200398128	12/21/04	6	9/2/04	27.4	10/5/04	75.6	11/2/04	132.2	11/30/04	205.2			
257	200434204	1	200398128	11/5/04	3	9/2/04	26.6	10/5/04	58.4	11/2/04	100.4					
258	200434205	1	200398128	1/4/05	4	9/2/04	33.6	10/5/04	83.6	11/2/04	124.6	11/30/04	193.2			
259	200434206	1	200398128	12/21/04	6	9/2/04	36.2	10/5/04	93.6	11/2/04	139.0	11/30/04	209.6			
260	200434207	1	200398128	1/4/05	4	9/2/04	30.6	10/5/04	84.6	11/2/04	136.0	11/30/04	205.6			
261	200434208	1	200398128	1/14/05	4	9/2/04	35.2	10/5/04	79.2	11/2/04	118.4	11/30/04	179.4	12/28/04	242.8	
262	200434209	1	200398128	1/14/05	4	9/2/04	23.6	10/5/04	71.8	11/2/04	109.0	11/30/04	170.6	12/28/04	237.2	
263	200434210	1	200398128	1/4/05	4	9/2/04	28.0	10/5/04	78.6	11/2/04	124.8	11/30/04	191.4	12/28/04	261.8	
264	200434301	2	200395515	12/15/04	6	9/2/04	41.2	10/5/04	92.0	11/2/04	150.8	11/30/04	217.6			
265	200434302	2	200395515	1/10/05	6	9/2/04	31.6	10/5/04	80.0	11/2/04	128.0	11/30/04	192.8	12/28/04	250.6	
266	200434303	2	200395515	1/6/05	6	9/2/04	30.4	10/5/04	78.2	11/2/04	117.6	11/30/04	185.0			
267	200434304	1	200395515	1/12/05	6	9/2/04	29.8	10/5/04	65.4	11/2/04	106.8	11/30/04	179.8	12/28/04	240.4	
268	200434305	1	200395515	1/4/05	4	9/2/04	41.4	10/5/04	96.6	11/2/04	146.0	11/30/04	210.2			
269	200434306	1	200395515	1/6/05	6	9/2/04	32.0	10/5/04	79.6	11/2/04	121.0	11/30/04	177.6	12/28/04	246.8	
270	200434308	1	200395515	1/4/05	4	9/2/04	30.2	10/5/04	85.0	11/2/04	126.8	11/30/04	197.2			
271	200434309	1	200395515	1/4/05	4	9/2/04	28.6	10/5/04	82.0	11/2/04	129.2	11/30/04	203.6			
272	200434310	1	200395515	12/29/04	6	9/2/04	28.4	10/5/04	73.4	11/2/04	134.0	11/30/04	197.2			
273	200434401	2	200390005	12/20/04	6	9/2/04	36.0	10/12/04	96.0	11/9/04	157.8	12/7/04	227.0			

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
274	200434402	2	200390005	12/20/04	6	9/2/04	37.8	10/12/04	106.2	11/9/04	168.6	12/7/04	231.6			
275	200434403	1	200390005	12/20/04	6	9/2/04	36.0	10/5/04	78.2	11/2/04	143.8	11/30/04	209.8			
276	200434501	2	200390003	1/10/05	6	9/7/04	34.8	10/12/04	83.6	11/9/04	133.0	12/7/04	194.2	1/4/05	263.2	
277	200434502	2	200390003	1/12/05	6	9/7/04	34.6	10/12/04	84.2	11/9/04	136.0	12/7/04	192.0	1/4/05	254.6	
278	200434503	2	200390003	1/10/05	6	9/7/04	34.4	10/12/04	84.2	11/9/04	137.4	12/7/04	197.0	1/4/05	265.2	
279	200434504	2	200390003	1/10/05	6	9/7/04	41.2	10/12/04	94.8	11/9/04	147.4	12/7/04	203.2	1/4/05	273.4	
280	200434506	1	200390003	1/12/05	7	9/7/04	39.2	10/5/04	76.2	11/2/04	128.4	11/30/04	183.8	12/28/04	243.8	
281	200434507	1	200390003	12/15/04	6	9/7/04	46.2	10/5/04	94.4	11/2/04	158.8	11/30/04	222.6			
282	200434508	1	200390003	12/20/04	7	9/7/04	39.2	10/5/04	78.0	11/2/04	136.2	11/30/04	205.4			
283	200434509	1	200390003	1/10/05	6	9/7/04	30.8	10/5/04	68.8	11/2/04	113.2	11/30/04	174.6	12/28/04	244.0	
284	200434510	1	200390003	1/3/05	7	9/7/04	39.0	10/5/04	83.4	11/2/04	142.0	11/30/04	197.4	12/28/04	262.2	
285	200434511	1	200390003	1/12/05	6	9/7/04	39.2	10/5/04	87.2	11/2/04	143.0	11/30/04	193.0	12/28/04	246.2	
286	200434512	1	200390003	12/20/04	6	9/7/04	37.0	10/5/04	80.0	11/2/04	131.4	11/30/04	203.2			
287	200434601	2	200390002	12/20/04	6	9/7/04	42.2	10/12/04	100.4	11/9/04	158.0	12/7/04	228.4			
288	200434602	2	200390002	1/20/05	6	9/7/04	38.2	10/12/04	81.4	11/9/04	130.6	12/7/04	187.0	1/4/05	242.0	
289	200434603	2	200390002	1/26/05	6	9/7/04	29.0	10/12/04	72.0	11/9/04	113.0	12/7/04	158.2	1/4/05	218.0	
290	200434604	2	200390002	2/2/05	6	9/7/04	33.8	10/12/04	75.2	11/9/04	113.8	12/7/04	163.0	1/4/05	214.0	1/25/C
291	200434605	2	200390002	1/26/05	6	9/7/04	31.8	10/12/04	80.0	11/9/04	131.4	12/7/04	185.8	1/4/05	237.4	
292	200434606	2	200390002	1/18/05	6	9/7/04	32.4	10/12/04	79.8	11/9/04	133.8	12/7/04	184.8	1/4/05	247.4	
293	200434607	1	200390002	1/10/05	6	9/7/04	29.0	10/5/04	63.8	11/2/04	111.8	11/30/04	178.6	12/28/04	244.0	
294	200434608	1	200390002	1/3/05	6	9/7/04	33.2	10/5/04	70.4	11/2/04	122.8	11/30/04	185.4	12/28/04	256.6	
295	200434801	2	200395515	1/4/05	4	9/7/04	45.0	10/12/04	102.4	11/9/04	162.0	12/7/04	217.8			
296	200434802	2	200395515	12/29/04	6	9/7/04	36.6	10/12/04	87.6	11/9/04	143.8	12/7/04	211.6			
297	200434803	2	200395515	1/10/05	6	9/7/04	42.2	10/12/04	88.6	11/9/04	138.8	12/7/04	201.4	1/4/05	267.0	
298	200434804	2	200395515	1/10/05	6	9/7/04	38.8	10/12/04	92.8	11/9/04	148.4	12/7/04	203.4	1/4/05	264.2	
299	200434805	2	200395515	1/20/05	6	9/7/04	31.0	10/12/04	77.6	11/9/04	130.8	12/7/04	178.6	1/4/05	235.4	
300	200434806	1	200395515	12/21/04	6	9/7/04	40.8	10/5/04	83.0	11/2/04	143.2	11/30/04	208.0			
301	200434807	1	200395515	12/21/04	6	9/7/04	43.6	10/5/04	87.0	11/2/04	146.6	11/30/04	213.6			
302	200434808	1	200395515	12/21/04	6	9/7/04	44.0	10/12/04	105.2	11/9/04	170.6	12/7/04	229.2			
303	200434809	1	200395515	12/27/04	6	9/7/04	37.4	10/12/04	92.8	11/9/04	154.2	12/7/04	219.2			
304	200434901	2	200398128	12/21/04	6	9/7/04	37.2	10/12/04	101.4	11/9/04	155.4	12/7/04	222.6			
305	200434902	2	200398128	12/27/04	6	9/7/04	43.4	10/12/04	97.8	11/9/04	156.8	12/7/04	219.4			
306	200434903	2	200398128	1/10/05	6	9/7/04	38.6	10/12/04	93.6	11/9/04	151.4	12/7/04	198.0	1/4/05	261.6	
307	200434904	2	200398128	1/10/05	6	9/7/04	35.0	10/12/04	89.8	11/9/04	140.4	12/7/04	200.0	1/4/05	259.4	
308	200434905	1	200398128	1/10/05	6	9/7/04	41.2	10/12/04	98.4	11/9/04	153.2	12/7/04	206.4	1/4/05	267.4	
309	200435301	2	200190498	1/6/05	6	9/7/04	43.4	10/12/04	92.2	11/9/04	155.2	12/7/04	210.6			
310	200435302	2	200190498	12/21/04	6	9/7/04	38.2	10/12/04	99.0	11/9/04	158.2	12/7/04	222.8			
311	200435303	2	200190498	12/15/04	6	9/7/04	40.0	10/12/04	98.8	11/9/04	162.2	12/7/04	233.6			
312	200435304	2	200190498	1/20/05	6	9/7/04	29.2	10/12/04	76.8	11/9/04	137.2	12/7/04	183.0	1/4/05	243.2	

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
313	200435305	1	200190498	1/6/05	6	9/7/04	33.8	10/12/04	85.6	11/9/04	152.2	12/7/04	211.0			
314	200435306	1	200190498	12/29/04	6	9/7/04	38.2	10/12/04	93.2	11/9/04	157.0	12/7/04	217.2			
315	200435307	1	200190498	1/20/05	6	9/7/04	39.6	10/12/04	100.4	11/9/04	167.0	12/7/04	183.0	1/4/05	242.2	
316	200435308	1	200190498	12/27/04	6	9/7/04	41.0	10/12/04	98.4	11/9/04	164.4	12/7/04	223.2			
317	200435309	1	200190498	12/13/04	6	9/7/04	45.8	10/12/04	115.0	11/9/04	191.4	12/7/04	266.8			
318	200435310	1	200190498	1/10/05	6	9/7/04	30.0	10/12/04	79.2	11/9/04	145.2	12/7/04	203.0	1/4/05	267.0	
319	200435311	1	200190498	12/21/04	6	9/7/04	37.4	10/12/04	96.6	11/9/04	165.4	12/7/04	228.6			
320	200435401	2	200395515	12/29/04	6	9/7/04	36.2	10/12/04	91.6	11/9/04	147.6	12/7/04	212.8			
321	200435402	2	200395515	12/29/04	6	9/7/04	33.6	10/12/04	88.4	11/9/04	142.4	12/7/04	210.6			
322	200435403	2	200395515	1/10/05	6	9/7/04	40.4	10/12/04	88.8	11/9/04	144.4	12/7/04	203.8	1/4/05	264.4	
323	200435404	2	200395515	12/21/04	6	9/7/04	45.8	10/12/04	104.6	11/9/04	164.0	12/7/04	224.8			
324	200435405	2	200395515	1/10/05	6	9/7/04	37.2	10/12/04	91.2	11/9/04	141.2	12/7/04	202.8	1/4/05	259.8	
325	200435407	1	200395515	12/21/04	6	9/7/04	44.6	10/12/04	102.6	11/9/04	166.2	12/7/04	230.8			
326	200435408	1	200395515	1/4/05	4	9/7/04	43.0	10/12/04	105.6	11/9/04	175.4	12/7/04	248.6			
327	200435409	1	200395515	1/3/05	6	9/7/04	37.2	10/12/04	88.6	11/9/04	147.2	12/7/04	210.6			
328	200435410	1	200395515	1/6/05	6	9/7/04	31.2	10/12/04	88.2	11/9/04	151.2	12/7/04	211.0			
329	200435411	1	200395515	1/6/05	6	9/7/04	34.4	10/12/04	90.6	11/9/04	152.8	12/7/04	210.6			
330	200435412	1	200395515	12/27/04	6	9/7/04	35.0	10/12/04	89.4	11/9/04	149.4	12/7/04	215.8			
331	200435413	1	200395515	12/29/04	6	9/7/04	29.4	10/12/04	80.6	11/9/04	140.2	12/7/04	207.6			
332	200435801	2	200390007	1/10/05	6	9/7/04	43.6	10/12/04	94.0	11/9/04	160.2	12/7/04	211.8	1/4/05	275.8	
333	200435802	1	200390007	12/29/04	6	9/7/04	48.8	10/12/04	113.2	11/9/04	168.8	12/7/04	221.4			
334	200435803	1	200390007	1/12/05	6	9/7/04	42.4	10/12/04	101.6	11/9/04	162.4	12/7/04	207.4	1/4/05	259.6	
335	200436001	2	200390005	1/18/05	6	9/7/04	34.8	10/12/04	90.6	11/9/04	141.4	12/7/04	201.4	1/4/05	254.6	
336	200436002	2	200390005	1/6/05	6	9/7/04	34.8	10/12/04	85.0	11/9/04	142.8	12/7/04	205.0			
337	200436003	2	200390005	1/26/05	6	9/7/04	35.6	10/12/04	89.0	11/9/04	128.4	12/7/04	185.8	1/4/05	234.6	
338	200436004	2	200390005	1/20/05	6	9/7/04	33.6	10/12/04	87.4	11/9/04	142.8	12/7/04	195.4	1/4/05	244.8	
339	200436005	2	200390005	1/12/05	6	9/9/04	35.0	10/12/04	86.4	11/9/04	144.6	12/7/04	201.6	1/4/05	255.6	
340	200436006	2	200390005	1/3/05	6	9/9/04	39.8	10/12/04	93.2	11/9/04	149.8	12/7/04	212.0			
341	200436007	2	200390005	1/10/05	6	9/9/04	40.0	10/12/04	95.2	11/9/04	155.6	12/7/04	207.8	1/4/05	272.8	
342	200436008	2	200390005	1/26/05	6	9/9/04	38.4	10/12/04	83.2	11/9/04	127.0	12/7/04	179.6	1/4/05	234.0	
343	200436009	1	200390005	12/13/04	6	9/9/04	47.8	10/12/04	110.4	11/9/04	184.2	12/7/04	251.2			
344	200436010	1	200390005	12/27/04	6	9/9/04	43.8	10/12/04	100.4	11/9/04	164.4	12/7/04	223.4			
345	200436011	1	200390005	12/15/04	6	9/9/04	37.2	10/12/04	94.2	11/9/04	160.0	12/7/04	235.0			
346	200436012	1	200390005	12/15/04	6	9/9/04	44.6	10/12/04	103.8	11/9/04	166.0	12/7/04	239.8			
347	200436013	1	200390005	1/6/05	6	9/9/04	32.6	10/12/04	80.8	11/9/04	144.6	12/7/04	207.0			
348	200436301	2	200395515	1/26/05	6	9/9/04	32.4	10/12/04	78.4	11/9/04	132.2	12/7/04	186.6	1/4/05	236.0	
349	200436302	2	200395515	2/10/05	8	9/9/04	37.6	10/12/04	90.2	11/9/04	141.2	12/7/04	199.2	1/4/05	253.8	
350	200436303	2	200395515	12/27/04	6	9/9/04	43.6	10/12/04	103.6	11/9/04	162.6	12/7/04	221.4			
351	200436304	2	200395515	2/4/05	4	9/9/04	40.0	10/12/04	90.8	11/9/04	144.4	12/7/04	197.0	1/4/05	237.8	

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
352	200436305	2	200395515	1/3/05	6	9/9/04	36.2	10/12/04	94.0	11/9/04	152.8	12/7/04	212.0			
353	200436306	2	200395515	2/4/05	4	9/9/04	37.4	10/12/04	86.6	11/9/04	138.4	12/7/04	189.8	1/4/05	236.6	
354	200436307	2	200395515	2/4/05	4	9/9/04	40.2	10/12/04	87.0	11/9/04	143.6	12/7/04	195.0	1/4/05	250.8	
355	200436309	1	200395515	12/27/04	6	9/9/04	38.8	10/12/04	93.0	11/9/04	152.8	12/7/04	217.2			
356	200436310	1	200395515	1/3/05	6	9/9/04	35.6	10/12/04	89.4	11/9/04	146.4	12/7/04	209.6			
357	200436311	1	200395515	1/4/05	4	9/9/04	33.2	10/12/04	88.0	11/9/04	150.4	12/7/04	214.6			
358	200436312	1	200395515	1/12/05	6	9/9/04	27.2	10/12/04	76.6	11/9/04	132.4	12/7/04	190.8	1/4/05	254.0	
359	200436401	2	200390005	1/26/05	6	9/9/04	35.4	10/12/04	81.0	11/9/04	126.0	12/7/04	181.0	1/4/05	229.6	
360	200436402	1	200390005	12/27/04	6	9/9/04	45.2	10/12/04	98.0	11/9/04	156.0	12/7/04	217.4			
361	200436403	1	200390005	2/2/05	6	9/9/04	43.4	10/12/04	94.2	11/9/04	143.4	12/7/04	190.2	1/4/05	194.8	1/25/C
362	200436404	1	200390005	1/3/05	6	9/9/04	39.0	10/12/04	96.2	11/9/04	146.8	12/7/04	209.6			
363	200436405	1	200390005	12/29/04	6	9/9/04	39.0	10/12/04	82.8	11/9/04	141.2	12/7/04	209.2			
364	200436601	2	200390007	1/20/05	6	9/9/04	45.4	10/12/04	96.0	11/9/04	148.8	12/7/04	198.6	1/4/05	243.2	
365	200436602	2	200390007	1/12/05	6	9/9/04	46.4	10/12/04	98.6	11/9/04	153.4	12/7/04	204.6	1/4/05	256.8	
366	200436603	2	200390007	1/12/05	6	9/9/04	42.8	10/12/04	97.2	11/9/04	150.6	12/7/04	200.2	1/4/05	256.8	
367	200436604	2	200390007	1/18/05	6	9/9/04	38.8	10/12/04	84.8	11/9/04	133.4	12/7/04	185.0	1/4/05	249.8	
368	200436605	1	200390007	1/12/05	7	9/9/04	41.6	10/12/04	84.6	11/9/04	140.6	12/7/04	195.6	1/4/05	257.4	
369	200436606	1	200390007	1/12/05	6	9/9/04	46.6	10/12/04	103.6	11/9/04	147.8	12/7/04	205.2	1/4/05	256.8	
370	200436607	5	200390007	1/18/05	6	9/9/04	41.4	10/12/04	90.6	11/9/04	142.8	12/7/04	184.0	1/4/05	249.4	
371	200436608	1	200390007	1/10/05	6	9/9/04	48.4	10/12/04	104.8	11/9/04	161.8	12/7/04	212.0	1/4/05	273.8	
372	200436609	1	200390007	12/20/04	7	9/9/04	47.6	10/12/04	106.6	11/9/04	165.6	12/7/04	226.8			
373	200436610	1	200390007	1/3/05	6	9/9/04	42.8	10/12/04	84.0	11/9/04	145.8	12/7/04	208.4			
374	200436611	1	200390007	2/10/05	8	9/9/04	40.0	10/12/04	94.0	11/9/04	124.8	12/7/04	188.0	1/4/05	241.0	
375	200436612	1	200390007	1/26/05	6	9/9/04	45.8	10/12/04	96.0	11/9/04	147.6	12/7/04	187.0	1/4/05	232.2	
376	200436901	2	200390003	1/18/05	6	9/7/04	31.2	10/12/04	73.8	11/9/04	125.4	12/7/04	187.0	1/4/05	248.4	
377	200436902	2	200390003	1/6/05	6	9/7/04	38.2	10/12/04	86.2	11/9/04	140.2	12/7/04	202.6			
378	200436903	2	200390003	1/20/05	6	9/7/04	31.4	10/12/04	82.6	11/9/04	125.2	12/7/04	186.6	1/4/05	238.4	
379	200436904	2	200390003	1/10/05	7	9/9/04	35.2	10/12/04	76.6	11/9/04	126.6	12/7/04	190.6	1/4/05	264.8	
380	200436905	2	200390003	1/18/05	6	9/9/04	29.2	10/12/04	73.2	11/9/04	119.8	12/7/04	175.6	1/4/05	243.8	
381	200436906	2	200390003	1/26/05	7	9/9/04	24.0	10/12/04	60.2	11/9/04	107.2	12/7/04	157.6	1/4/05	227.0	
382	200436907	2	200390003	2/10/05	8	9/9/04	27.4	10/12/04	61.8	11/9/04	115.6	12/7/04	172.0	1/4/05	235.0	
383	200436908	2	200390003	2/10/05	8	9/9/04	28.0	10/12/04	61.8	11/9/04	107.0	12/7/04	165.6	1/4/05	214.6	1/25/C
384	200436909	1	200390003	12/15/04	6	9/9/04	42.8	10/12/04	99.2	11/9/04	169.4	12/7/04	237.8			
385	200436910	1	200390003	1/10/05	7	9/9/04	37.0	10/12/04	86.0	11/9/04	151.6	12/7/04	205.4	1/4/05	273.2	
386	200436911	1	200390003	12/29/04	6	9/9/04	35.8	10/12/04	78.2	11/9/04	141.6	12/7/04	208.8			
387	200436912	1	200390003	1/12/05	6	9/9/04	30.2	10/12/04	70.8	11/9/04	123.4	12/7/04	192.8	1/4/05	258.2	
388	200436913	1	200390003	1/12/05	6	9/9/04	32.0	10/12/04	78.4	11/9/04	137.6	12/7/04	192.0	1/4/05	255.4	
389	200437301	2	200395515	12/13/04	6	9/9/04	43.4	10/12/04	103.6	11/9/04	170.4	12/7/04	244.6			
390	200437302	2	200395515	1/6/05	6	9/9/04	33.0	10/12/04	80.8	11/9/04	139.0	12/7/04	204.8			

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
391	200437303	2	200395515	12/15/04	6	9/9/04	41.4	10/12/04	102.6	11/9/04	169.4	12/7/04	237.8			
392	200437304	1	200395515	1/4/05	4	9/9/04	40.4	10/12/04	97.2	11/9/04	167.6	12/7/04	231.6			
393	200437305	1	200395515	12/15/04	6	9/9/04	39.2	10/12/04	93.4	11/9/04	165.6	12/7/04	234.8			
394	200437306	1	200395515	1/4/05	4	9/9/04	47.8	10/12/04	106.4	11/9/04	177.6	12/7/04	244.8			
395	200437307	1	200395515	12/13/04	6	9/9/04	41.6	10/12/04	98.4	11/9/04	168.4	12/7/04	243.6			
396	200437308	1	200395515	1/4/05	4	9/9/04	35.4	10/12/04	94.4	11/9/04	159.0	12/7/04	230.0			
397	200437309	1	200395515	12/27/04	6	9/9/04	39.4	10/12/04	97.6	11/9/04	158.8	12/7/04	219.8			
398	200437401	2	200390007	2/2/05	6	9/7/04	22.0	10/12/04	71.2	11/9/04	125.8	12/7/04	176.2	1/4/05	222.8	1/25/C
399	200437402	2	200390007	1/26/05	6	9/9/04	35.0	10/12/04	83.6	11/9/04	139.8	12/7/04	188.4	1/4/05	235.4	
400	200437403	1	200390007	1/10/05	7	9/9/04	41.0	10/12/04	97.2	11/9/04	161.8	12/7/04	210.2	1/4/05	273.0	
401	200437404	1	200390007	12/27/04	6	9/9/04	37.0	10/12/04	88.0	11/9/04	147.8	12/7/04	214.8			
402	200437405	1	200390007	1/26/05	7	9/9/04	37.8	10/12/04	58.8	11/9/04	115.2	12/7/04	176.6	1/4/05	234.0	
403	200437406	1	200390007	1/12/05	6	9/9/04	34.8	10/12/04	90.0	11/9/04	147.6	12/7/04	196.4	1/4/05	258.0	
404	200437407	1	200390007	2/10/05	8	9/9/04	37.4	10/12/04	88.6	11/9/04	133.8	12/7/04	187.6	1/4/05	251.0	
405	200437408	1	200390007	1/6/05	6	9/9/04	34.0	10/12/04	83.0	11/9/04	158.6	12/7/04	212.8			
406	200437409	1	200390007	1/10/05	7	9/9/04	30.6	10/12/04	82.8	11/9/04	141.0	12/7/04	194.6	1/4/05	267.8	
407	200437410	1	200390007	1/10/05	7	9/9/04	32.8	10/12/04	84.4	11/9/04	145.0	12/7/04	202.4	1/4/05	268.8	
408	200437411	1	200390007	2/10/05	8	9/9/04	29.0	10/12/04	78.2	11/9/04	135.8	12/7/04	193.6	1/4/05	246.8	
409	200437501	2	200390007	1/3/05	6	9/9/04	40.6	10/12/04	80.6	11/9/04	147.2	12/7/04	210.8			
410	200437502	2	200390007	1/10/05	6	9/9/04	39.4	10/12/04	84.4	11/9/04	143.2	12/7/04	202.0	1/4/05	266.0	
411	200437504	1	200390007	12/20/04	6	9/9/04	37.6	10/12/04	92.6	11/9/04	164.8	12/7/04	226.8			
412	200437505	1	200390007	12/15/04	7	9/9/04	44.2	10/12/04	103.4	11/9/04	183.2	12/7/04	240.2			
413	200437506	1	200390007	12/15/04	6	9/9/04	37.6	10/12/04	96.6	11/9/04	163.6	12/7/04	231.6			
414	200437507	1	200390007	12/15/04	7	9/9/04	47.8	10/12/04	111.4	11/9/04	178.4	12/7/04	243.0			
415	200437509	1	200390007	1/26/05	6	9/9/04	38.4	10/12/04	88.6	11/9/04	151.4	12/7/04	185.2	1/4/05	233.4	
416	200437510	1	200390007	1/3/05	6	9/9/04	35.2	10/12/04	76.8	11/9/04	147.8	12/7/04	209.4			
417	200437801	2	200390007	12/27/04	6	9/9/04	50.6	10/12/04	104.4	11/9/04	163.2	12/7/04	221.8			
418	200437802	1	200390007	12/27/04	6	9/9/04	43.6	10/19/04	120.2	11/16/04	182.4	12/14/04	238.8			
419	200437803	1	200390007	1/18/05	6	9/9/04	43.4	10/19/04	115.8	11/16/04	170.0	12/14/04	218.2	1/11/05	273.2	
420	200437804	1	200390007	1/3/05	6	9/9/04	39.8	10/19/04	110.6	11/16/04	175.2	12/14/04	230.8			
421	200438101	2	200390007	1/18/05	6	9/9/04	39.0	10/12/04	92.6	11/9/04	145.4	12/7/04	197.6	1/4/05	250.0	
422	200438102	2	200390007	2/2/05	6	9/9/04	40.8	10/12/04	95.6	11/9/04	148.8	12/7/04	205.0	1/4/05	235.6	1/25/C
423	200438103	2	200390007	2/10/05	8	9/9/04	37.4	10/12/04	87.6	11/9/04	121.2	12/7/04	169.4	1/4/05	224.8	
424	200438104	2	200390007	1/12/05	6	9/9/04	39.2	10/12/04	90.8	11/9/04	143.6	12/7/04	197.4	1/4/05	257.0	
425	200438105	2	200390007	1/6/05	6	9/9/04	37.8	10/12/04	92.6	11/9/04	146.0	12/7/04	207.2			
426	200438106	1	200390007	1/6/05	6	9/9/04	44.6	10/19/04	105.0	11/16/04	168.6	12/14/04	225.8			
427	200438107	1	200390007	12/27/04	6	9/9/04	37.0	10/19/04	109.8	11/16/04	171.6	12/14/04	234.8			
428	200438401	2	200398128	12/27/04	6	9/9/04	35.6	10/12/04	88.2	11/9/04	147.2	12/7/04	214.0			
429	200438402	2	200398128	1/10/05	6	9/7/04	32.4	10/12/04	88.0	11/9/04	151.4	12/7/04	205.8	1/4/05	273.8	

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
430	200438403	2	200398128	2/2/05	6	9/9/04	32.8	10/12/04	75.2	11/9/04	132.2	12/7/04	188.8	1/4/05	224.0	1/25/C
431	200438404	1	200398128	12/27/04	6	9/14/04	48.8	10/19/04	111.8	11/16/04	181.6	12/14/04	237.0			
432	200438405	1	200398128	2/10/05	8	9/14/04	38.6	10/19/04	87.4	11/16/04	141.2	12/14/04	195.0	1/11/05	250.4	
433	200438406	1	200398128	12/27/04	6	9/9/04	37.0	10/19/04	102.8	11/16/04	170.6	12/14/04	235.6			
434	200438407	1	200398128	1/4/05	4	9/14/04	49.0	10/19/04	109.6	11/16/04	175.4	12/14/04	238.2			
435	200438408	1	200398128	12/21/04	6	9/14/04	42.6	10/19/04	107.2	11/16/04	168.0	12/14/04	240.4			
436	200438409	1	200398128	1/4/05	4	9/14/04	47.4	10/19/04	110.6	11/16/04	185.8	12/14/04	258.0			
437	200438410	1	200398128	1/4/05	4	9/14/04	35.6	10/19/04	100.6	11/16/04	169.2	12/14/04	229.4			
438	200438411	1	200398128	2/4/05	4	9/14/04	39.0	10/19/04	91.6	11/16/04	142.8	12/14/04	201.0	1/11/05	264.2	
439	200438412	1	200398128	1/4/05	4	9/14/04	38.0	10/19/04	98.0	11/16/04	162.0	12/14/04	233.6			
440	200438413	1	200398128	2/4/05	4	9/14/04	35.8	10/19/04	82.0	11/16/04	132.8	12/14/04	181.6	1/11/05	255.8	
441	200438414	1	200398128	1/4/05	4	9/14/04	37.8	10/19/04	91.2	11/16/04	153.0	12/14/04	221.2			
442	200438501	2	200398128	1/10/05	6	9/9/04	30.6	10/12/04	83.4	11/9/04	141.4	12/7/04	202.8	1/4/05	271.2	
443	200438502	2	200398128	1/14/05	4	9/9/04	34.0	10/12/04	85.4	11/9/04	148.8	12/7/04	206.4	1/4/05	271.8	
444	200438503	2	200398128	1/3/05	6	9/9/04	38.2	10/12/04	77.6	11/9/04	131.4	12/7/04	203.0			
445	200438504	2	200398128	1/4/05	4	9/14/04	45.4	10/12/04	84.8	11/9/04	137.2	12/7/04	201.8			
446	200438505	2	200398128	12/27/04	6	9/14/04	45.0	10/12/04	85.8	11/9/04	143.2	12/7/04	212.4			
447	200438506	2	200398128	1/4/05	4	9/14/04	44.0	10/12/04	90.6	11/9/04	151.2	12/7/04	219.8			
448	200438507	2	200398128	12/1/04	3	9/14/04	53.4	10/12/04	97.4	11/9/04	149.6					
449	200438508	2	200398128	2/4/05	4	9/14/04	34.6	10/12/04	68.8	11/9/04	117.4	12/7/04	181.8	1/4/05	243.4	
450	200438509	2	200398128	2/4/05	4	9/14/04	41.6	10/12/04	86.8	11/9/04	141.2	12/7/04	198.8	1/4/05	250.8	
451	200438510	2	200398128	1/4/05	4	9/14/04	39.8	10/12/04	84.0	11/9/04	135.2	12/7/04	205.6			
452	200438511	1	200398128	12/21/04	6	9/14/04	53.6	10/19/04	118.6	11/16/04	193.2	12/14/04	249.4			
453	200438512	1	200398128	11/5/04	3	9/14/04	45.6	10/19/04	86.6							
454	200438513	1	200398128	12/21/04	6	9/14/04	46.0	10/19/04	107.2	11/16/04	174.2	12/14/04	239.4			
455	200438514	1	200398128	11/4/04	3	9/14/04	45.6	10/19/04	101.8							
456	200438601	2	200390007	2/10/05	8	9/14/04	45.0	10/12/04	92.4	11/9/04	149.2	12/7/04	204.2	1/4/05	240.6	1/25/C
457	200438602	2	200390007	1/18/05	6	9/14/04	41.4	10/12/04	88.0	11/9/04	145.6	12/7/04	200.0	1/4/05	251.8	
458	200438603	2	200390007	1/26/05	6	9/14/04	40.0	10/19/04	101.0	11/16/04	163.8	12/14/04	196.6	1/11/05	242.4	
459	200438604	2	200390007	2/2/05	6	9/14/04	41.4	10/19/04	95.8	11/16/04	146.8	12/14/04	189.8	1/11/05	230.6	1/25/C
460	200438605	2	200390007	1/26/05	6	9/14/04	42.4	10/19/04	98.4	11/16/04	157.2	12/14/04	198.4	1/11/05	243.8	
461	200438606	2	200390007	1/26/05	6	9/14/04	36.6	10/19/04	88.6	11/16/04	149.8	12/14/04	196.0	1/11/05	243.4	
462	200438607	1	200390007	1/18/05	6	9/14/04	42.0	10/19/04	95.6	11/16/04	155.4	12/14/04	210.2	1/11/05	281.6	
463	200438608	1	200390007	1/20/05	6	9/14/04	42.2	10/19/04	76.8	11/16/04	118.6	12/14/04	178.6	1/11/05	249.4	
464	200438609	1	200390007	1/18/05	6	9/14/04	39.2	10/19/04	95.6	11/16/04	153.0	12/14/04	206.0	1/11/05	272.0	
465	200438610	1	200390007	1/18/05	6	9/14/04	28.8	10/19/04	81.6	11/16/04	136.2	12/14/04	199.4	1/11/05	271.0	
466	200438611	1	200390007	1/20/05	6	9/14/04	36.8	10/19/04	89.4	11/16/04	148.6	12/14/04	201.2	1/11/05	259.4	
467	200438701	2	200395515	1/26/05	6	9/14/04	44.8	10/19/04	98.8	11/16/04	156.6	12/14/04	202.2	1/11/05	248.5	
468	200438702	2	200395515	2/10/05	8	9/14/04	43.0	10/19/04	96.2	11/16/04	149.4	12/14/04	187.2	1/11/05	246.4	1/25/C

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
469	200438703	2	200395515	2/2/05	6	9/14/04	41.8	10/19/04	94.8	11/16/04	152.0	12/14/04	193.6	1/11/05	229.4	1/25/C
470	200438704	2	200395515	2/15/05	4	9/14/04	40.6	10/19/04	97.0	11/16/04	149.6	12/14/04	196.2	1/11/05	244.2	
471	200438705	2	200395515	1/20/05	6	9/14/04	48.8	10/19/04	104.4	11/16/04	160.6	12/14/04	199.4	1/11/05	256.4	
472	200438706	1	200395515	1/6/05	6	9/14/04	38.0	10/19/04	94.8	11/16/04	162.0	12/14/04	222.8			
473	200438707	1	200395515	1/18/05	6	9/14/04	43.2	10/19/04	101.8	11/16/04	143.6	12/14/04	197.2	1/11/05	270.2	
474	200438709	1	200395515	1/20/05	6	9/14/04	37.2	10/19/04	88.8	11/16/04	143.0	12/14/04	188.8	1/11/05	248.4	
475	200439201	2	200398128	1/14/05	4	9/14/04	49.4	10/19/04	93.0	11/16/04	157.0	12/14/04	214.6			
476	200439202	2	200398128	12/29/04	6	9/14/04	48.4	10/19/04	110.6	11/16/04	173.4	12/14/04	231.2			
477	200439203	2	200398128	2/15/05	4	9/14/04	42.0	10/19/04	98.4	11/16/04	161.6	12/14/04	208.2	1/11/05	249.6	
478	200439204	2	200398128	1/26/05	6	9/15/04	42.3	10/19/04	86.0	11/16/04	137.8	12/14/04	181.2	1/11/05	239.6	
479	200439205	2	200398128	1/18/05	6	9/15/04	41.0	10/12/04	76.8	11/9/04	138.0	12/7/04	190.8	1/4/05	252.4	
480	200439206	1	200398128	12/29/04	6	9/15/04	55.1	10/19/04	86.0	11/16/04	158.8	12/14/04	226.8			
481	200439207	1	200398128	2/10/05	8	9/15/04	42.4	10/19/04	91.0	11/16/04	151.4	12/14/04	206.8	1/11/05	254.4	
482	200439208	1	200398128	1/6/05	6	9/15/04	45.0	10/19/04	85.4	11/16/04	153.6	12/14/04	217.8			
483	200439209	1	200398128	1/14/05	4	9/15/04	40.9	10/19/04	83.4	11/16/04	149.4	12/14/04	213.0			
484	200439210	1	200398128	12/27/04	6	9/15/04	46.7	10/19/04	105.8	11/16/04	170.2	12/14/04	236.2			
485	200439211	1	200398128	1/4/05	4	9/15/04	49.2	10/19/04	107.0	11/16/04	177.0	12/14/04	242.0			
486	200439212	1	200398128	1/4/05	4	9/15/04	44.2	10/19/04	93.6	11/16/04	161.2	12/14/04	231.0			
487	200439215	1	200398128	1/14/05	4	9/15/04	40.2	10/19/04	93.0	11/16/04	152.2	12/14/04	212.0			
488	200439401	2	200398128	2/2/05	6	9/14/04	47.6	10/19/04	105.0	11/16/04	167.2	12/14/04	207.6	1/11/05	244.2	1/25/C
489	200439403	1	200398128	2/4/05	4	9/14/04	34.6	10/19/04	83.6	11/16/04	143.4	12/14/04	197.2	1/11/05	259.2	
490	200439404	1	200398128	2/2/05	6	9/14/04	41.8	10/19/04	92.0	11/16/04	160.6	12/14/04	183.6	1/11/05	226.4	1/25/C
491	200439405	1	200398128	1/4/05	4	9/14/04	42.8	10/19/04	100.2	11/16/04	168.2	12/14/04	238.6			
492	200439406	1	200398128	12/29/04	6	9/14/04	42.8	10/19/04	98.8	11/16/04	166.0	12/14/04	230.0			
493	200439407	1	200398128	2/4/05	4	9/14/04	35.4	10/19/04	82.4	11/16/04	144.0	12/14/04	203.0	1/11/05	270.6	
494	200439408	1	200398128	1/4/05	4	9/14/04	38.6	10/19/04	94.0	11/16/04	160.0	12/14/04	220.6			
495	200439409	1	200398128	1/4/05	4	9/14/04	41.8	10/19/04	98.4	11/16/04	166.4	12/14/04	230.4			
496	200439501	2	200390007	1/26/05	6	9/14/04	44.8	10/19/04	100.6	11/16/04	155.4	12/14/04	193.4	1/11/05	245.6	
497	200439502	2	200390007	1/20/05	6	9/14/04	50.8	10/19/04	114.4	11/16/04	172.2	12/14/04	212.6	1/11/05	259.8	
498	200439503	2	200390007	2/2/05	6	9/14/04	47.0	10/19/04	103.8	11/16/04	158.6	12/14/04	194.2	1/11/05	239.0	1/25/C
499	200439504	2	200390007	1/26/05	6	9/14/04	40.4	10/19/04	97.0	11/16/04	147.4	12/14/04	188.6	1/11/05	239.0	
500	200439505	1	200390007	1/18/05	6	9/14/04	44.2	10/19/04	99.6	11/16/04	153.2	12/14/04	203.0	1/11/05	268.6	
501	200439901	1	200395515	1/20/05	6	9/14/04	43.6	10/19/04	99.6	11/16/04	163.8	12/14/04	215.2	1/11/05	261.8	
502	200439902	1	200395515	1/18/05	6	9/14/04	40.2	10/19/04	99.6	11/16/04	163.4	12/14/04	215.0	1/11/05	290.4	
503	200439903	1	200395515	1/18/05	6	9/14/04	39.6	10/19/04	102.4	11/16/04	161.4	12/14/04	213.0	1/11/05	271.2	
504	200439904	1	200395515	1/26/05	6	9/14/04	37.0	10/19/04	89.0	11/16/04	136.6	12/14/04	161.4	1/11/05	226.4	
505	200440601	2	200398128	1/3/05	6	9/15/04	50.4	10/12/04	94.0	11/9/04	147.4	12/7/04	209.6			
506	200440602	2	200398128	12/29/04	6	9/15/04	50.4	10/12/04	90.8	11/9/04	146.4	12/7/04	212.2			
507	200440603	1	200398128	1/6/05	6	9/15/04	41.9	10/19/04	96.6	11/16/04	167.2	12/14/04	225.0			

	Animal	Sex	Sire	Disposal Date	Disposal Code	8 weeks		12 weeks		16 weeks		20 weeks		24 weeks		28 w
						Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date	Weight (lbs)	Date
508	200440604	1	200398128	1/4/05	4	9/15/04	47.8	10/19/04	108.8	11/16/04	176.4	12/14/04	242.6			
509	200440605	1	200398128	12/27/04	6	9/15/04	48.8	10/19/04	113.4	11/16/04	184.2	12/14/04	240.6			
510	200440606	1	200398128	1/4/05	4	9/15/04	41.8	10/19/04	97.6	11/16/04	167.4	12/14/04	226.4			
511	200440607	1	200398128	1/4/05	4	9/15/04	48.3	10/19/04	103.8	11/16/04	165.8	12/14/04	226.4			
512	200440608	1	200398128	9/24/04	3	9/15/04	25.2									
513	200440609	1	200398128	2/4/05	4	9/15/04	39.1	10/19/04	89.4	11/16/04	148.2	12/14/04	200.8	1/11/05	247.8	
514	200440901	2	200398128	1/3/05	6	9/15/04	37.7	10/12/04	77.6	11/9/04	134.2	12/7/04	204.2			
515	200440902	2	200398128	1/14/05	4	9/15/04	41.5	10/12/04	78.6	11/9/04	138.6	12/7/04	198.6	1/4/05	266.0	
516	200440903	2	200398128	1/12/05	6	9/15/04	34.8	10/12/04	66.2	11/9/04	123.2	12/7/04	191.4	1/4/05	256.2	
517	200440905	2	200398128	2/15/05	4	9/15/04	27.7	10/12/04	58.4	11/9/04	105.0	12/7/04	161.6	1/4/05	226.6	
518	200440906	2	200398128	1/3/05	6	9/15/04	39.2	10/12/04	78.2	11/9/04	147.6	12/7/04	209.6			
519	200440907	1	200398128	1/18/05	6	9/15/04	37.4	10/19/04	96.0	11/16/04	163.2	12/14/04	215.4	1/11/05	277.0	
520	200440908	1	200398128	1/4/05	4	9/15/04	38.5	10/19/04	93.2	11/16/04	163.6	12/14/04	234.0			
521	200440909	1	200398128	1/18/05	6	9/15/04	36.6	10/19/04	79.8	11/16/04	145.0	12/14/04	203.2	1/11/05	269.6	
522	200440910	1	200398128	1/4/05	4	9/15/04	38.6	10/19/04	100.0	11/16/04	169.4	12/14/04	230.8			
523	200440911	1	200398128	1/6/05	6	9/15/04	29.3	10/19/04	76.6	11/16/04	137.8	12/14/04	210.0			
524	200440912	1	200398128	1/14/05	4	9/15/04	31.7	10/19/04	86.0	11/16/04	146.2	12/14/04	209.2			
525	200440913	1	200398128	1/14/05	4	9/15/04	36.0	10/19/04	96.8	11/16/04	155.2	12/14/04	212.4			
526	200441201	2	200395515	1/26/05	6	9/15/04	37.4	10/12/04	81.2	11/9/04	127.6	12/7/04	189.0	1/4/05	235.2	
527	200441202	2	200395515	1/18/05	6	9/15/04	37.8	10/12/04	74.6	11/9/04	128.2	12/7/04	191.4	1/4/05	250.4	
528	200441203	1	200395515	1/26/05	6	9/15/04	33.9	10/19/04	86.2	11/16/04	144.6	12/14/04	200.4	1/11/05	249.2	
529	200441204	1	200395515	2/10/05	8	9/15/04	34.6	10/19/04	91.0	11/16/04	148.0	12/14/04	204.6	1/11/05	260.4	
530	200441205	1	200395515	1/26/05	6	9/15/04	34.1	10/19/04	81.2	11/16/04	141.8	12/14/04	177.0	1/11/05	234.8	
531	200441206	1	200395515	2/4/05	4	9/15/04	40.0	10/19/04	102.8	11/16/04	164.8	12/14/04	215.8	1/11/05	277.8	
532	200441207	1	200395515	1/26/05	6	9/15/04	28.5	10/19/04	79.4	11/16/04	133.4	12/14/04	194.0	1/11/05	244.4	
533	200441208	1	200395515	2/4/05	4	9/15/04	38.8	10/19/04	95.2	11/16/04	156.4	12/14/04	212.4	1/11/05	271.4	
534	200441209	1	200395515	1/26/05	6	9/15/04	28.7	10/19/04	74.2	11/16/04	132.0	12/14/04	185.0	1/11/05	239.2	
535	200441210	1	200395515	2/4/05	4	9/15/04	32.4	10/19/04	76.2	11/16/04	133.4	12/14/04	190.6	1/11/05	243.0	
536	200441211	1	200395515	1/26/05	6	9/15/04	33.9	10/12/04	70.8	11/9/04	126.0	12/7/04	180.4	1/4/05	234.8	

Disposal Codes		
Code	Description	#
3	Dead	10
4	Control Shipped for sale	64
6	Slaughter data	405
7	Tanked at slaughter	35
8	Shipped to Purdue	22
Total=		536

Sex Codes	
1	Male
2	Female

Weights=Lbs.

Total Number	Day Order	Marc Animal ID	Treatment (Control/Clone)	Slaughter Date	Hot Carcass Weight/lbs	USDA CARCASS MUSCLE (1-3)	CARCASS LENGTH (cm)	FIRMNESS (1-3)	COLOR (1-6)	MARBLING (1-6, 10)	Longissimus pH (24 hr)	LOINEYE AREA (in ²)	hunter (L*)	hunter (a*)	hunter (B*)
1	1	200433509	Control	12-Dec-00	183	3	84.9	2	2	2	5.90	6.37	53.98	8.26	13.78
2	2	200437307	Control	12-Dec-00	160	3	83.5	2	3	2	6.00	5.60	55.38	7.88	14.31
3	3	200430508	Control	12-Dec-00	166	3	82.5	2	3	2	5.70	5.96	58.10	5.80	13.95
4	4	200437301	Control	12-Dec-00	168	3	81.0	2	3	2	5.90	7.11	55.26	7.39	13.84
5	5	200433409	Control	12-Dec-00	173	3	79.7	2	3	2	6.50	5.33	52.60	6.32	12.24
6	6	200435309	Control	12-Dec-00	184	3	84.6	2	2	3	6.20	5.40	57.99	8.33	16.16
7	7	200430503	Control	12-Dec-00	168	3	81.8	2	2	1	5.90	6.68	55.06	7.95	13.55
8	8	200433510	Control	12-Dec-00	183	3	81.3	2	3	2	6.20	6.11	55.93	8.62	15.19
9	9	200430709	Clone	12-Dec-00	172	3	84.8	2	2	2	6.10	6.60	58.80	6.53	14.54
10	10	200431505	Clone	12-Dec-00	168	3	81.4	2	2	2	6.20	6.45	59.83	6.21	14.35
11	11	200430706	Clone	12-Dec-00	182	3	81.5	2	2	2	6.10	6.84	60.17	6.35	14.23
12	12	200431408	Clone	12-Dec-00	174	3	80.4	2	2	2	6.10	7.84	54.24	7.77	13.08
13	13	200430906	Clone	12-Dec-00	178	3	78.6	2	3	4	5.60	6.42	60.07	8.41	16.27
14	14	200430707	Clone	12-Dec-00	175	3	79.2	2	3	3	5.80	6.44	55.88	7.58	14.37
15	15	200430604	Clone	12-Dec-00	186	3	80.5	2	3	3	5.70	6.69	55.42	8.37	13.74
16	16	200432405	Clone	12-Dec-00	168	3	79.5	2	3	3	5.70	6.70	58.18	6.19	13.52
17	17	200431411	Clone	12-Dec-00	167	3	78.7	2	3	2	5.80	6.35	54.38	7.02	12.82
18	18	200431006	Clone	12-Dec-00	179	3	81.7	2	3	2	5.60	5.89	53.10	9.19	14.75
19	19	200436009	Clone	12-Dec-00	171	3	79.5	2	3	2	5.70	6.80	57.77	7.11	13.64
20	20	200431502	Clone	12-Dec-00	175	3	83.0	2	3	2	5.60	6.91	54.72	8.10	13.16
21	21	200430711	Clone	12-Dec-00	176	3	84.0	2	3	3	5.70	6.37	56.11	7.61	14.26
22	22	200430710	Clone	12-Dec-00	189	3	84.0	2	3	3	5.70	6.74	57.10	8.22	15.58
23	23	200430805	Clone	12-Dec-00	170	3	84.8	2	3	3	5.90	5.74	55.05	7.72	13.82
24	24	200430902	Clone	12-Dec-00	168	3	76.8	2	3	3	5.60	6.00	57.87	8.19	14.46
25	25	200431608	Clone	12-Dec-00	176	3	80.7	2	3	5	5.60	6.31	59.83	5.05	14.24
26	26	200433605	Clone	12-Dec-00	176	3	81.5	2	3	2	5.80	5.86	53.68	7.47	13.48
27	27	200430905	Clone	12-Dec-00	173	3	77.0	2	3	5	5.70	5.70	62.82	6.02	15.33
28	28	200433807	Clone	12-Dec-00	173	3	80.0	2	3	4	5.70	6.60	55.72	6.36	13.73
29	29	200430804	Clone	12-Dec-00	178	3	84.0	2	3	4	5.80	5.81	56.91	7.41	14.44
30	30	200431404	Clone	12-Dec-00	169	3	82.6	2	3	3	5.70	8.19	55.88	7.38	13.14
31	31	200431009	Clone	12-Dec-00	173	3	83.0	2	3	4	5.70	5.74	56.75	7.10	14.47
32	1	200432911	Control	14-Dec-00	162	3	83.0	2	3	3	5.80	5.43	56.20	8.82	14.51

Total Number	Day Order	Marc Animal ID	Treatment (Control/Clone)	Slaughter Date	Hot Carcass Weight/lbs	USDA CARCASS MUSCLE (1-3)	CARCASS LENGTH (cm)	FIRMNESS (1-3)	COLOR (1-6)	MARBLING (1-6, 10)	Longissimus pH (24 hr)	LOINEYE AREA (in ²)	hunter (L*)	hunter (a*)	hunter (B*)
33	2	200434001	Control	14-Dec-00	163	3	78.4	2	3	3	5.80	6.01	56.43	7.20	13.64
34	3	200431802	Control	14-Dec-00	166	3	81.0	2	3	2	5.70	7.13	54.77	7.10	13.26
35	4	200434301	Control	14-Dec-00	165	3	80.2	2	3	3	5.70	6.03	57.51	7.68	13.69
36	5	200435303	Control	14-Dec-00	165	3	81.7	2	3	2	5.70	6.21	54.95	8.17	14.09
37	6	200437305	Control	14-Dec-00	158	3	81.0	2	3	4	5.70	6.13	57.82	7.09	13.85
38	7	200432609	Control	14-Dec-00	168	3	80.8	2	3	3	5.70	6.16	56.94	7.00	14.72
39	8	200437303	Control	14-Dec-00	169	3	81.0	2	3	3	5.70	6.85	58.51	6.76	13.80
40	9	200434202	Control	14-Dec-00	169	3	76.7	2	3	2	5.70	5.71	60.89	8.88	15.35
41	10	200433301	Control	14-Dec-00	161	3	79.5	2	3	4	5.80	5.93	58.22	6.49	14.55
42	11	200437506	Clone	14-Dec-00	162	3	79.0	2	3	3	5.90	6.26	60.14	6.82	15.30
43	12	200436011	Clone	14-Dec-00	162	3	82.0	2	3	2	5.60	6.48	56.30	7.44	14.15
44	13	200430708	Clone	14-Dec-00	170	3	81.3	2	2	2	5.60	6.87	63.20	7.15	15.11
45	14	200430705	Clone	14-Dec-00	169	3	79.5	2	3	3	5.60	7.00	55.26	7.55	13.67
46	15	200431606	Clone	14-Dec-00	174	3	81.5	2	3	2	5.70	6.33	58.49	5.13	14.05
47	16	200434507	Clone	14-Dec-00	170	3	81.5	2	3	3	5.70	6.85	55.72	9.05	14.51
48	17	200431504	Clone	14-Dec-00	173	3	77.8	2	3	3	5.70	6.59	60.44	5.97	14.94
49	18	200436012	Clone	14-Dec-00	165	3	80.2	2	3	4	5.70	5.60	56.02	8.16	14.96
50	19	200433601	Clone	14-Dec-00	161	3	78.0	2	3	3	5.70	7.27	54.07	7.86	13.79
51	20	200431008	Clone	14-Dec-00	168	3	82.7	2	3	3	5.70	5.14	56.72	8.94	15.69
52	21	200430609	Clone	14-Dec-00	159	3	79.7	2	3	3	5.80	5.60	54.79	7.24	13.26
53	22	200431501	Clone	14-Dec-00	167	3	82.0	2	3	1	5.70	7.06	53.75	9.01	14.31
54	23	200436909	Clone	14-Dec-00	167	3	78.5	2	3	2	5.60	6.47	55.56	7.71	13.72
55	24	200431407	Clone	14-Dec-00	167	3	80.3	2	3	2	5.70	6.79	54.76	7.79	14.25
56	25	200431405	Clone	14-Dec-00	156	3	79.5	2	3	2	5.80	7.37	57.24	7.81	15.37
57	26	200430605	Clone	14-Dec-00	165	3	78.0	2	3	3	5.70	6.66	57.28	9.09	16.22
58	27	200433201	Clone	14-Dec-00	176	3	80.0	2	3	4	5.60	6.98	56.57	7.52	13.94
59	28	200430607	Clone	14-Dec-00	177	3	81.5	2	3	3	5.60	5.71	56.41	7.23	14.44
60	1	200431604	Clone	19-Dec-00	176	3	80.5	2	3	2	5.70	8.64	57.62	6.87	13.93
61	2	200433603	Clone	19-Dec-00	170	3	80.0	2	3	3	5.70	6.67	56.59	8.19	14.81
62	3	200432103	Clone	19-Dec-00	173	3	81.8	2	3	4	5.80	6.15	54.26	7.72	14.54
63	4	200430703	Clone	19-Dec-00	165	3	81.3	2	3	2	5.70	7.93	56.20	7.96	13.92
64	5	200437504	Clone	19-Dec-00	161	3	82.5	2	3	4	5.80	4.99	58.98	6.87	15.00

Total Number	Day Order	Marc Animal ID	Treatment (Control/Clone)	Slaughter Date	Hot Carcass Weight/lbs	USDA CARCASS MUSCLE (1-3)	CARCASS LENGTH (cm)	FIRMNESS (1-3)	COLOR (1-6)	MARBLING (1-6, 10)	Longissimus pH (24 hr)	LOINEYE AREA (in ²)	hunter (L*)	hunter (a*)	hunter (B*)
65	6	200434512	Clone	19-Dec-00	165	3	79.0	2	3	3	5.70	6.12	58.03	5.95	13.71
66	7	200434403	Clone	19-Dec-00	170	3	78.5	2	3	3	5.70	5.87	56.21	6.19	14.00
67	8	200432704	Clone	19-Dec-00	158	3	80.0	2	3	4	5.60	5.67	56.56	6.90	14.32
68	9	200433711	Clone	19-Dec-00	166	3	80.5	2	3	4	5.70	7.16	57.06	8.03	14.76
69	10	200430903	Clone	19-Dec-00	166	3	78.3	2	3	4	5.80	6.87	60.02	6.32	15.62
70	11	200433205	Clone	19-Dec-00	166	3	81.0	2	3	3	5.70	5.53	55.51	8.17	14.92
71	12	200432004	Clone	19-Dec-00	174	3	82.7	2	3	3	5.60	5.81	56.66	6.58	14.13
72	13	200432404	Clone	19-Dec-00	168	3	75.5	2	3	3	5.70	6.74	59.44	7.03	15.29
73	14	200431003	Clone	19-Dec-00	163	3	82.2	2	3	3	5.70	6.56	57.76	6.55	14.33
74	15	200431406	Clone	19-Dec-00	157	3	79.0	2	3	3	5.70	6.89	59.04	6.89	14.12
75	16	200432108	Clone	19-Dec-00	165	3	80.6	2	3	3	5.70	5.91	57.28	7.64	14.10
76	17	200431610	Clone	19-Dec-00	171	3	81.7	2	3	3	5.70	7.38	55.57	6.53	13.14
77	18	200431011	Clone	19-Dec-00	174	3	84.5	2	3	2	5.70	7.06	52.98	9.08	13.94
78	19	200434402	Clone	19-Dec-00	173	3	79.7	2	3	2	5.70	6.85	52.52	7.58	12.06
79	20	200432310	Clone	19-Dec-00	163	3	79.3	2	3	3	5.70	5.79	54.93	7.30	13.24
80	21	200432308	Clone	19-Dec-00	170	3	80.3	2	3	3	5.60	5.85	54.60	6.88	13.50
81	22	200432406	Clone	19-Dec-00	184	3	79.5	2	3	3	5.70	6.27	56.59	7.52	15.48
82	23	200432706	Clone	19-Dec-00	163	3	78.5	2	3	3	5.80	5.72	55.71	7.11	13.24
83	24	200434601	Clone	19-Dec-00	163	3	80.0	2	3	3	5.70	6.85	50.60	10.48	12.22
84	25	200432705	Clone	19-Dec-00	173	3	77.3	2	3	3	5.80	6.10	56.72	8.31	15.46
85	26	200434401	Clone	19-Dec-00	167	3	79.0	2	3	3	5.60	6.82	56.92	7.94	15.62
86	1	200433505	Control	20-Dec-00	170	3	83.3	2	3	3	5.80	7.04	52.01	7.60	12.66
87	2	200434206	Control	20-Dec-00	171	3	80.8	2	3	4	5.80	6.39	57.63	8.09	14.77
88	3	200432602	Control	20-Dec-00	170	3	82.0	2	3	2	6.00	6.55	53.67	6.82	12.69
89	4	200438511	Control	20-Dec-00	181	3	82.0	2	3	3	5.70	6.45	58.36	7.50	14.77
90	5	200438408	Control	20-Dec-00	173	3	84.0	2	3	3	5.70	6.87	57.83	6.85	13.37
91	6	200438513	Control	20-Dec-00	172	3	79.2	2	3	4	6.00	6.20	62.82	4.70	14.05
92	7	200433405	Control	20-Dec-00	166	3	83.5	2	3	2	5.60	6.53	55.53	6.27	13.30
93	8	200435302	Control	20-Dec-00	164	3	81.4	2	3	3	5.70	6.51	56.56	7.15	14.30
94	9	200435311	Control	20-Dec-00	177	3	81.2	2	3	3	5.80	6.31	58.09	7.87	14.69
95	10	200435407	Control	20-Dec-00	164	3	81.3	2	3	2	5.70	5.27	57.60	8.27	14.53
96	11	200433512	Control	20-Dec-00	161	3	81.3	2	3	2	5.60	6.05	54.39	7.39	12.64

Total Number	Day Order	Marc Animal ID	Treatment (Control/Clone)	Slaughter Date	Hot Carcass Weight/lbs	USDA CARCASS MUSCLE (1-3)	CARCASS LENGTH (cm)	FIRMNESS (1-3)	COLOR (1-6)	MARBLING (1-6, 10)	Longissimus pH (24 hr)	LOINEYE AREA (in ²)	hunter (L*)	hunter (a*)	hunter (B*)
97	12	200434808	Control	20-Dec-00	168	3	80.5	2	3	3	5.60	5.47	54.68	8.27	14.06
98	13	200432607	Control	20-Dec-00	179	3	81.0	2	3	3	5.70	8.05	58.68	5.47	14.51
99	14	200430510	Control	20-Dec-00	168	3	80.4	2	3	3	5.80	6.55	54.71	8.31	13.67
100	15	200431805	Control	20-Dec-00	159	3	77.5	2	3	2	5.80	5.80	55.26	6.84	13.31
101	16	200434807	Control	20-Dec-00	166	3	83.0	2	3	3	5.90	6.08	53.50	7.89	13.31
102	17	200433306	Control	20-Dec-00	176	3	79.7	2	3	4	6.10	6.40	58.04	7.83	15.93
103	18	200434005	Control	20-Dec-00	165	3	81.0	2	3	4	5.70	5.59	58.23	7.01	13.79
104	19	200432610	Control	20-Dec-00	172	3	81.5	2	3	3	6.00	6.16	54.60	6.96	13.51
105	20	200435404	Control	20-Dec-00	166	3	84.8	2	3	1	5.60	6.38	56.91	6.58	12.85
106	21	200433511	Control	20-Dec-00	164	3	80.6	2	3	2	5.60	6.46	56.26	6.61	13.02
107	22	200433307	Control	20-Dec-00	171	3	80.0	2	3	4	5.70	5.12	58.14	6.40	14.58
108	23	200434003	Control	20-Dec-00	160	3	80.5	2	3	3	5.70	5.79	57.48	7.57	13.58
109	24	200433408	Control	20-Dec-00	173	3	81.7	2	3	2	5.90	6.61	49.95	10.30	12.72
110	25	200432907	Control	20-Dec-00	170	3	82.0	2	3	3	5.70	6.31	54.33	8.64	13.77
111	26	200432913	Control	20-Dec-00	157	3	81.2	2	3	3	5.90	5.36	54.01	8.07	14.40
112	27	200432601	Control	20-Dec-00	172	3	82.0	2	3	3	5.80	7.54	55.38	7.75	12.92
113	28	200434203	Control	20-Dec-00	169	3	80.0	2	3	3	5.80	6.11	58.95	6.23	14.01
114	29	200434901	Control	20-Dec-00	163	3	80.6	2	3	2	5.90	7.22	53.17	7.45	12.20
115	30	200434806	Control	20-Dec-00	166	3	82.0	2	3	3	5.80	5.42	53.91	7.43	12.97
116	1	200431801	Control	26-Dec-00	168	3	81.7	2	3	3	5.70	7.23	55.22	8.50	13.71
117	2	200435308	Control	26-Dec-00	164	3	82.0	2	3	3	5.70	5.89	56.10	6.99	13.97
118	3	200434902	Control	26-Dec-00	166	3	80.3	2	3	3	5.70	6.19	53.34	8.34	12.89
119	4	200436303	Control	26-Dec-00	156	3	78.8	2	3	3	5.80	7.76	56.94	7.62	14.19
120	5	200438401	Control	26-Dec-00	163	3	82.0	2	3	2	5.70	7.04	55.90	6.37	12.76
121	6	200435412	Control	26-Dec-00	159	3	79.7	2	3	3	5.70	6.31	56.88	8.05	14.57
122	7	200438404	Control	26-Dec-00	167	3	82.5	2	3	3	5.90	7.00	59.66	5.13	13.37
123	8	200436309	Control	26-Dec-00	169	3	80.5	2	3	2	5.70	5.78	57.45	7.25	13.99
124	9	200438406	Control	26-Dec-00	173	3	83.0	2	3	2	5.70	6.79	53.71	7.95	13.73
125	10	200437309	Control	26-Dec-00	171	3	83.0	2	3	3	6.00	5.66	57.80	10.07	16.92
126	11	200434007	Control	26-Dec-00	180	3	81.8	2	3	3	6.00	5.56	56.30	6.83	13.81
127	12	200440605	Control	26-Dec-00	170	3	83.5	2	3	2	5.70	6.09	56.90	5.19	13.35
128	13	200438505	Control	26-Dec-00	169	3	80.7	2	3	3	5.70	7.70	56.85	6.45	13.11

Total Number	Day Order	Marc Animal ID	Treatment (Control/Clone)	Slaughter Date	Hot Carcass Weight/lbs	USDA CARCASS MUSCLE (1-3)	CARCASS LENGTH (cm)	FIRMNESS (1-3)	COLOR (1-6)	MARBLING (1-6, 10)	Longissimus pH (24 hr)	LOINEYE AREA (in ²)	hunter (L*)	hunter (a*)	hunter (B*)
129	14	200434809	Control	26-Dec-00	164	3	82.0	2	3	4	5.70	5.15	53.86	7.98	14.65
130	15	200439210	Control	26-Dec-00	171	3	80.5	2	3	3	5.60	8.27	56.39	5.59	13.20
131	16	200431704	Clone	26-Dec-00	173	3	79.5	2	3	3	5.70	6.26	58.11	6.28	14.26
132	17	200436010	Clone	26-Dec-00	173	3	83.0	2	3	2	5.70	7.01	56.83	7.60	14.76
133	18	200431703	Clone	26-Dec-00	174	3	82.5	2	3	3	5.70	6.54	54.53	8.33	14.18
134	19	200431706	Clone	26-Dec-00	175	3	81.0	2	3	4	5.60	4.98	57.62	10.79	17.80
135	20	200431403	Clone	26-Dec-00	156	3	79.5	2	3	3	5.60	6.42	55.46	6.02	12.61
136	21	200431401	Clone	26-Dec-00	161	3	83.5	2	3	2	5.80	7.98	54.95	8.40	13.85
137	22	200430802	Clone	26-Dec-00	175	3	84.0	2	3	2	5.60	6.82	57.59	7.62	14.70
138	23	200432402	Clone	26-Dec-00	164	3	79.3	2	3	2	5.60	6.84	56.92	6.82	14.06
139	24	200433803	Clone	26-Dec-00	171	3	77.7	2	3	3	5.70	6.90	57.14	7.04	13.93
140	25	200437404	Clone	26-Dec-00	166	3	79.3	2	3	4	5.80	6.49	55.51	6.17	13.45
141	26	200433602	Clone	26-Dec-00	175	3	81.0	2	3	2	5.60	6.47	56.20	7.70	14.41
142	27	200430704	Clone	26-Dec-00	167	3	79.6	2	3	2	5.70	7.18	56.48	6.47	13.79
143	28	200436402	Clone	26-Dec-00	166	3	80.0	2	3	3	5.70	6.49	59.67	5.98	14.76
144	29	200437802	Clone	26-Dec-00	175	3	79.0	2	3	3	5.90	7.43	52.47	8.83	14.27
145	30	200433206	Clone	26-Dec-00	170	3	79.5	2	3	2	5.60	6.50	55.14	8.20	14.82
146	31	200432105	Clone	26-Dec-00	154	3	79.5	2	3	2	5.70	5.01	55.03	7.87	14.50
147	32	200432702	Clone	26-Dec-00	176	3	81.7	2	3	3	5.80	6.51	51.79	10.01	14.32
148	33	200437801	Clone	26-Dec-00	178	3	82.5	2	3	3	5.80	7.61	57.21	7.09	13.99
149	34	200438107	Clone	26-Dec-00	171	3	78.5	2	3	6	5.70	6.33	62.81	6.99	15.57
150	35	200433809	Clone	26-Dec-00	179	3	80.0	2	3	3	5.70	6.83	58.90	6.22	14.56
151	1	200435401	Control	28-Dec-00	165	3	81.2	2	3	3	5.80	6.23	55.48	7.77	13.98
152	2	200433404	Control	28-Dec-00	179	3	83.5	2	3	3	5.70	7.76	56.01	6.55	14.08
153	3	200439206	Control	28-Dec-00	178	3	83.0	2	3	3	5.80	7.46	57.50	6.85	14.01
154	4	200432604	Control	28-Dec-00	182	3	83.2	2	3	3	5.90	7.23	55.84	6.62	13.57
155	5	200434310	Control	28-Dec-00	176	3	80.0	2	3	4	5.80	6.36	59.71	5.82	14.84
156	6	200434004	Control	28-Dec-00	178	3	82.0	2	3	3	5.80	6.71	57.19	7.78	14.04
157	7	200435413	Control	28-Dec-00	163	3	79.7	2	3	3	5.70	6.03	57.39	7.49	14.88
158	8	200440602	Control	28-Dec-00	178	3	81.5	2	3	4	5.90	5.93	55.79	7.11	14.19
159	9	200432909	Control	28-Dec-00	166	3	82.5	2	3	3	5.80	5.89	54.76	8.15	13.88
160	10	200439202	Control	28-Dec-00	164	3	81.5	2	3	3	5.70	7.63	51.91	8.93	13.67

Total Number	Day Order	Marc Animal ID	Treatment (Control/Clone)	Slaughter Date	Hot Carcass Weight/lbs	USDA CARCASS MUSCLE (1-3)	CARCASS LENGTH (cm)	FIRMNESS (1-3)	COLOR (1-6)	MARBLING (1-6, 10)	Longissimus pH (24 hr)	LOINEYE AREA (in ²)	hunter (L*)	hunter (a*)	hunter (B*)
161	11	200434802	Control	28-Dec-00	169	3	82.4	2	3	3	5.70	6.26	55.42	8.96	15.25
162	12	200434201	Control	28-Dec-00	163	3	82.3	2	3	3	5.80	6.43	53.68	8.62	12.71
163	13	200439406	Control	28-Dec-00	178	3	79.0	2	3	3	5.80	8.05	57.12	6.77	14.26
164	14	200435402	Control	28-Dec-00	174	3	83.2	2	3	3	5.80	6.85	53.25	8.28	13.76
165	15	200435306	Control	28-Dec-00	173	3	80.5	2	3	3	5.80	5.55	56.01	8.99	15.43
166	16	200432002	Clone	28-Dec-00	164	3	79.0	2	3	2	5.70	7.01	52.34	7.63	11.93
167	17	200432101	Clone	28-Dec-00	169	3	81.5	2	3	2	5.80	6.68	53.63	8.55	13.51
168	18	200432503	Clone	28-Dec-00	175	3	84.0	2	3	3	6.10	6.19	58.90	7.23	14.78
169	19	200431607	Clone	28-Dec-00	185	3	81.8	2	3	2	6.10	6.65	57.22	7.64	15.19
170	20	200433710	Clone	28-Dec-00	186	3	79.7	2	3	4	5.70	7.42	57.77	6.55	14.43
171	21	200436405	Clone	28-Dec-00	166	3	82.4	2	3	3	5.70	6.93	58.08	6.24	14.59
172	22	200432006	Clone	28-Dec-00	176	3	81.0	2	3	2	5.70	5.95	56.75	6.51	14.52
173	23	200430803	Clone	28-Dec-00	170	3	81.3	2	3	3	5.80	5.91	53.88	9.63	14.77
174	24	200436911	Clone	28-Dec-00	171	3	80.0	2	3	4	5.80	5.91	57.32	6.81	15.01
175	25	200433604	Clone	28-Dec-00	176	3	82.5	2	3	3	6.10	6.50	54.68	10.11	15.08
176	26	200432501	Clone	28-Dec-00	177	3	81.0	2	3	2	6.10	7.18	54.12	8.83	14.29
177	27	200433207	Clone	28-Dec-00	175	3	79.3	2	3	3	5.70	5.11	55.93	8.97	16.17
178	28	200432302	Clone	28-Dec-00	149	3	78.3	2	3	3	5.70	6.23	52.13	7.50	13.36
179	29	200433804	Clone	28-Dec-00	168	3	78.0	2	3	3	5.60	6.79	57.35	7.31	14.46
180	30	200435802	Clone	28-Dec-00	176	3	77.5	2	3	4	5.80	6.40	56.25	7.63	14.12
181	31	200433808	Clone	28-Dec-00	155	3	79.5	2	3	3	5.80	6.90	56.90	6.65	14.67
182	32	200430606	Clone	28-Dec-00	174	3	80.0	2	3	3	5.80	5.22	54.57	6.99	13.80
183	1	200433402	Control	02-Jan-01	177	3	85.5	2	3	2	5.70	7.46	56.54	7.47	13.86
184	2	200433504	Control	02-Jan-01	176	3	81.0	2	3	3	5.80	8.44	55.13	7.14	13.80
185	3	200430502	Control	02-Jan-01	185	3	84.2	2	3	3	5.70	6.56	57.36	7.21	14.78
186	4	200433403	Control	02-Jan-01	187	3	81.7	2	3	3	5.90	6.64	57.49	7.16	14.16
187	5	200436310	Control	02-Jan-01	175	3	80.0	2	3	3	6.00	5.61	58.27	6.42	14.75
188	6	200438503	Control	02-Jan-01	177	3	81.3	2	3	3	5.70	7.15	56.27	6.97	13.30
189	7	200440601	Control	02-Jan-01	181	3	83.2	2	3	5	5.80	7.60	55.74	7.67	13.84
190	8	200435409	Control	02-Jan-01	184	3	83.5	2	3	3	5.80	6.07	55.15	7.11	13.75
191	9	200434009	Control	02-Jan-01	177	3	78.2	2	3	5	5.90	6.50	57.36	7.47	13.67
192	10	200440901	Control	02-Jan-01	179	3	85.0	2	3	2	5.90	8.76	54.89	6.94	13.15

Total Number	Day Order	Marc Animal ID	Treatment (Control/Clone)	Slaughter Date	Hot Carcass Weight/lbs	USDA CARCASS MUSCLE (1-3)	CARCASS LENGTH (cm)	FIRMNESS (1-3)	COLOR (1-6)	MARBLING (1-6, 10)	Longissimus pH (24 hr)	LOINEYE AREA (in ²)	hunter (L*)	hunter (a*)	hunter (B*)
193	11	200440906	Control	02-Jan-01	162	3	83.5	2	3	2	5.80	7.49	56.18	7.71	15.06
194	12	200433501	Control	02-Jan-01	172	3	82.0	2	3	1	5.90	7.37	56.63	8.17	14.52
195	13	200430509	Control	02-Jan-01	183	3	81.6	2	3	3	5.80	6.80	57.60	6.86	13.94
196	14	200436305	Control	02-Jan-01	176	3	81.7	2	3	2	5.80	6.48	55.10	7.15	14.03
197	15	200431012	Clone	02-Jan-01	191	3	83.5	2	3	4	5.80	6.16	55.68	6.26	14.26
198	16	200437501	Clone	02-Jan-01	172	3	84.3	2	3	3	6.00	6.79	56.16	6.95	13.70
199	17	200433203	Clone	02-Jan-01	188	3	82.5	2	3	3	5.70	7.34	55.02	9.45	15.16
200	18	200436006	Clone	02-Jan-01	181	3	83.0	2	3	1	5.70	8.02	55.18	7.84	13.99
201	19	200434608	Clone	02-Jan-01	177	3	84.3	2	3	3	5.70	6.66	54.34	8.43	14.71
202	20	200430608	Clone	02-Jan-01	183	3	82.0	2	3	3	5.70	6.20	54.79	7.91	14.41
203	21	200431007	Clone	02-Jan-01	180	3	84.0	2	3	3	5.70	7.78	55.29	7.61	13.76
204	22	200433806	Clone	02-Jan-01	182	3	78.3	2	3	3	5.80	6.44	54.19	7.81	14.73
205	23	200432007	Clone	02-Jan-01	162	3	78.5	2	3	2	5.60	7.08	54.80	7.28	13.88
206	24	200431402	Clone	02-Jan-01	180	3	83.4	2	3	2	5.70	8.06	52.70	8.24	13.56
207	25	200432104	Clone	02-Jan-01	181	3	82.7	2	3	3	5.80	5.99	56.86	7.45	14.61
208	26	200431409	Clone	02-Jan-01	190	3	86.0	2	3	2	5.80	5.93	53.61	8.49	13.90
209	27	200437510	Clone	02-Jan-01	178	3	84.3	2	3	4	5.70	6.09	57.38	7.15	15.41
210	28	200436404	Clone	02-Jan-01	177	3	86.1	2	3	3	5.70	5.93	58.35	7.24	15.66
211	29	200433712	Clone	02-Jan-01	183	3	81.0	2	3	4	6.00	7.25	56.23	8.16	14.15
212	30	200436610	Clone	02-Jan-01	166	3	77.4	2	3	4	5.70	6.23	57.22	7.12	14.62
213	31	200437804	Clone	02-Jan-01	181	3	78.7	2	3	4	5.80	6.94	55.79	7.32	14.26
214	1	200437302	Control	05-Jan-01	181	3	87.7	2	3	3	5.70	6.51	56.51	6.52	13.92
215	2	200433507	Control	05-Jan-01	174	3	86.0	2	3	3	5.70	6.49	56.84	6.54	14.00
216	3	200434306	Control	05-Jan-01	178	3	81.5	2	3	3	5.70	6.72	56.58	7.27	14.15
217	4	200433503	Control	05-Jan-01	173	3	82.5	2	3	3	5.70	6.57	54.30	7.94	13.29
218	5	200430501	Control	05-Jan-01	180	3	82.0	2	3	2	5.90	7.83	56.35	6.41	13.49
219	6	200440603	Control	05-Jan-01	187	3	82.7	2	3	4	5.80	6.42	57.90	6.19	14.09
220	7	200435410	Control	05-Jan-01	176	3	82.2	2	3	3	5.60	6.85	56.00	8.16	13.92
221	8	200434008	Control	05-Jan-01	184	3	82.5	2	3	4	5.80	6.45	55.68	8.01	13.90
222	9	200435411	Control	05-Jan-01	183	3	82.7	2	2	1	5.60	6.07	60.44	8.66	15.65
223	10	200438706	Control	05-Jan-01	177	3	83.0	2	3	4	5.70	6.16	63.35	4.94	14.34
224	11	200439208	Control	05-Jan-01	171	3	81.0	2	3	3	5.70	7.14	53.85	8.28	13.65

Total Number	Day Order	Marc Animal ID	Treatment (Control/Clone)	Slaughter Date	Hot Carcass Weight/lbs	USDA CARCASS MUSCLE (1-3)	CARCASS LENGTH (cm)	FIRMNESS (1-3)	COLOR (1-6)	MARBLING (1-6, 10)	Longissimus pH (24 hr)	LOINEYE AREA (in ²)	hunter (L*)	hunter (a*)	hunter (B*)
225	12	200440911	Control	05-Jan-01	171	3	80.7	2	3	3	5.60	6.83	58.08	6.99	13.30
226	13	200435305	Control	05-Jan-01	185	3	84.4	2	3	3	5.70	6.67	51.62	7.53	12.53
227	14	200433502	Control	05-Jan-01	177	3	84.0	2	3	1	5.60	8.93	54.70	6.48	12.49
228	15	200435301	Control	05-Jan-01	185	3	86.6	2	3	2	5.60	6.91	53.85	7.46	13.97
229	16	200434303	Control	05-Jan-01	154	3	85.0	2	3	2	5.80	6.72	55.49	6.48	13.07
230	17	200431603	Clone	05-Jan-01	181	3	83.5	2	3	1	5.60	8.15	53.46	7.43	12.37
231	18	200433003	Clone	05-Jan-01	173	3	84.0	2	3	3	5.60	6.46	55.42	5.92	13.20
232	19	200432312	Clone	05-Jan-01	176	3	79.5	2	3	4	5.70	6.23	56.54	6.77	14.20
233	20	200432005	Clone	05-Jan-01	180	3	80.0	2	3	3	5.60	5.81	57.67	6.41	14.43
234	21	200436902	Clone	05-Jan-01	172	3	81.0	2	3	4	5.60	8.06	52.95	8.28	12.93
235	22	200436002	Clone	05-Jan-01	177	3	84.2	2	3	3	5.60	7.99	53.16	7.85	13.31
236	23	200431410	Clone	05-Jan-01	174	3	85.8	2	3	2	5.60	7.37	51.64	9.40	13.20
237	24	200432401	Clone	05-Jan-01	176	3	83.0	2	3	2	5.60	6.92	58.10	6.54	13.40
238	25	200432102	Clone	05-Jan-01	180	3	83.4	2	3	3	5.60	7.10	54.77	6.27	13.30
239	26	200436013	Clone	05-Jan-01	176	3	85.5	2	3	3	5.70	6.00	55.61	6.26	12.76
240	27	200432504	Clone	05-Jan-01	170	3	81.7	2	3	3	5.60	7.05	57.77	6.29	14.06
241	28	200438106	Clone	05-Jan-01	179	3	83.5	2	3	4	5.60	7.05	56.52	7.78	14.39
242	29	200432001	Clone	05-Jan-01	182	3	82.0	2	3	2	5.60	7.85	52.92	7.40	12.41
243	30	200438105	Clone	05-Jan-01	170	3	83.0	2	3	4	5.60	6.63	57.48	7.85	15.03
244	31	200431506	Clone	05-Jan-01	175	3	80.2	2	3	3	5.70	6.46	58.12	4.61	13.76
245	32	200437408	Clone	05-Jan-01	179	3	83.5	2	3	5	5.70	6.36	62.55	3.91	14.33
246	1	200433506	Control	09-Jan-01	186	3	85.0	2	3	3	5.60	7.12	56.82	6.83	13.78
247	2	200434302	Control	09-Jan-01	184	3	86.5	2	3	3	5.70	6.84	57.92	7.07	14.60
248	3	200434903	Control	09-Jan-01	183	3	82.5	2	3	4	5.70	7.87	53.86	7.67	12.63
249	4	200435405	Control	09-Jan-01	177	3	84.0	2	3	3	5.70	6.48	57.31	7.55	14.09
250	5	200434905	Control	09-Jan-01	185	3	81.5	2	3	3	5.90	6.67	55.96	6.27	13.49
251	6	200432906	Control	09-Jan-01	183	3	86.7	2	3	3	5.90	7.15	51.41	8.33	12.69
252	7	200435310	Control	09-Jan-01	183	3	86.5	2	3	3	5.90	6.21	51.76	8.48	13.73
253	8	200438402	Control	09-Jan-01	183	3	83.3	2	3	3	5.70	6.68	54.68	7.56	13.23
254	9	200438501	Control	09-Jan-01	187	3	84.0	2	3	3	5.70	6.92	53.47	8.12	13.49
255	10	200434803	Control	09-Jan-01	176	3	85.3	2	3	4	5.80	5.35	51.94	8.14	12.76
256	11	200432904	Control	09-Jan-01	175	3	87.7	2	3	3	5.80	6.58	55.79	6.44	12.95

Total Number	Day Order	Marc Animal ID	Treatment (Control/Clone)	Slaughter Date	Hot Carcass Weight/lbs	USDA CARCASS MUSCLE (1-3)	CARCASS LENGTH (cm)	FIRMNESS (1-3)	COLOR (1-6)	MARBLING (1-6, 10)	Longissimus pH (24 hr)	LOINEYE AREA (in ²)	hunter (L*)	hunter (a*)	hunter (B*)
257	12	200434904	Control	09-Jan-01	180	3	82.2	2	3	3	5.70	7.70	53.09	6.91	12.51
258	13	200430506	Control	09-Jan-01	171	3	83.0	2	3	3	5.60	7.16	53.14	7.64	13.01
259	14	200435403	Control	09-Jan-01	176	3	83.3	2	3	2	5.80	6.36	55.34	7.28	13.34
260	15	200434804	Control	09-Jan-01	179	3	83.7	2	3	2	5.70	6.35	52.73	7.29	12.95
261	16	200434509	Clone	09-Jan-01	178	3	81.8	2	3	3	5.70	7.51	55.10	8.32	13.65
262	17	200434607	Clone	09-Jan-01	178	3	80.3	2	3	3	5.70	6.90	56.35	7.14	13.97
263	18	200437502	Clone	09-Jan-01	186	3	86.5	2	3	3	5.80	8.32	54.29	8.25	13.74
264	19	200434504	Clone	09-Jan-01	180	3	82.0	2	3	4	5.70	7.28	53.75	8.06	13.17
265	20	200436007	Clone	09-Jan-01	190	3	84.0	2	3	3	5.70	7.29	53.51	8.18	14.54
266	21	200432407	Clone	09-Jan-01	178	3	83.8	2	3	2	5.60	6.32	53.88	7.31	13.35
267	22	200434501	Clone	09-Jan-01	180	3	80.6	2	3	2	5.60	7.16	55.10	8.08	14.08
268	23	200430610	Clone	09-Jan-01	173	3	79.5	2	3	2	5.70	6.85	57.84	6.68	14.47
269	24	200436608	Clone	09-Jan-01	184	3	81.0	2	3	4	5.80	6.64	60.77	5.74	14.65
270	25	200433002	Clone	09-Jan-01	181	3	83.2	2	3	2	5.70	8.60	50.40	8.33	11.36
271	26	200431701	Clone	09-Jan-01	180	3	83.0	2	3	3	5.80	7.46	56.10	7.41	13.85
272	27	200433202	Clone	09-Jan-01	170	3	82.5	2	3	3	5.60	7.19	55.88	8.40	14.34
273	28	200434503	Clone	09-Jan-01	180	3	82.6	2	3	3	5.80	6.83	55.15	7.24	13.44
274	29	200430701	Clone	09-Jan-01	178	3	80.3	2	3	3	5.70	8.20	51.59	8.47	12.84
275	30	200435801	Clone	09-Jan-01	188	3	81.0	2	3	5	6.00	6.62	58.85	7.00	14.60
276	1	200430504	Control	11-Jan-01	171	3	79.8	2	3	2	5.60	7.24	54.11	8.42	12.67
277	2	200434006	Control	11-Jan-01	185	3	79.5	2	3	4	5.70	6.32	60.18	7.34	14.28
278	3	200436312	Control	11-Jan-01	175	3	80.3	2	3	5	5.70	5.28	58.63	7.81	15.15
279	4	200432901	Control	11-Jan-01	186	3	83.5	2	3	2	5.90	7.37	50.75	8.67	12.96
280	5	200440903	Control	11-Jan-01	181	3	83.0	2	3	3	5.60	7.43	56.85	6.59	13.62
281	6	200434304	Control	11-Jan-01	182	3	79.2	2	3	4	5.60	5.70	57.48	7.41	15.30
282	7	200436913	Clone	11-Jan-01	177	3	84.0	2	3	3	5.50	6.50	57.24	7.75	14.86
283	8	200431702	Clone	11-Jan-01	184	3	79.0	2	3	3	5.60	6.82	62.03	6.98	16.58
284	9	200434502	Clone	11-Jan-01	179	3	81.2	2	3	3	5.60	7.08	55.37	8.68	14.76
285	10	200436912	Clone	11-Jan-01	177	3	81.2	2	3	4	5.60	6.83	58.55	6.46	14.52
286	11	200431004	Clone	11-Jan-01	171	3	81.5	2	3	2	5.60	8.25	55.46	7.24	12.27
287	12	200436603	Clone	11-Jan-01	180	3	84.7	2	3	4	5.60	7.29	57.02	7.50	14.14
288	13	200432403	Clone	11-Jan-01	174	3	82.5	2	3	3	5.60	7.35	60.01	5.80	14.12

Total Number	Day Order	Marc Animal ID	Treatment (Control/Clone)	Slaughter Date	Hot Carcass Weight/lbs	USDA CARCASS MUSCLE (1-3)	CARCASS LENGTH (cm)	FIRMNESS (1-3)	COLOR (1-6)	MARBLING (1-6, 10)	Longissimus pH (24 hr)	LOINEYE AREA (in ²)	hunter (L*)	hunter (a*)	hunter (B*)
289	14	200431605	Clone	11-Jan-01	185	3	84.0	2	3	3	5.60	6.82	57.47	6.91	14.59
290	15	200431010	Clone	11-Jan-01	175	3	85.0	2	3	2	5.60	7.11	57.91	6.59	13.95
291	16	200430611	Clone	11-Jan-01	188	3	79.8	2	3	4	5.60	6.66	58.52	7.18	14.99
292	17	200431002	Clone	11-Jan-01	179	3	83.2	2	3	2	5.50	7.67	51.86	9.64	12.63
293	18	200436005	Clone	11-Jan-01	180	3	82.5	2	3	1	5.70	8.55	54.21	5.51	12.95
294	19	200436606	Clone	11-Jan-01	178	3	81.5	2	3	2	5.80	6.79	53.29	7.42	13.67
295	20	200436602	Clone	11-Jan-01	177	3	83.5	2	3	2	5.60	7.43	57.21	7.71	14.31
296	21	200431601	Clone	11-Jan-01	180	3	81.5	2	3	1	5.60	7.67	53.45	7.39	13.17
297	22	200438104	Clone	11-Jan-01	178	3	85.2	2	3	2	5.60	6.83	58.20	6.94	14.15
298	23	200437406	Clone	11-Jan-01	182	3	78.3	2	3	4	5.70	5.84	59.67	5.90	13.96
299	24	200435803	Clone	11-Jan-01	174	3	79.5	2	3	5	5.70	6.49	57.74	7.31	14.92
300	25	200434511	Clone	11-Jan-01	185	3	83.3	2	3	2	5.50	6.18	57.77	6.20	14.15
301	1	200438707	Control	17-Jan-01	191	3	86.0	2	3	4	5.70	7.39	58.73	5.89	14.21
302	2	200440909	Control	17-Jan-01	191	3	85.5	2	3	4	5.70	7.00	59.44	5.47	13.76
303	3	200440907	Control	17-Jan-01	195	3	83.5	2	3	4	5.70	6.71	59.52	5.05	13.99
304	4	200433406	Control	17-Jan-01	191	3	83.7	2	3	3	5.60	8.34	56.69	6.81	14.34
305	5	200433303	Control	17-Jan-01	186	3	83.8	2	3	3	5.70	6.91	57.72	7.65	14.45
306	6	200439205	Control	17-Jan-01	180	3	80.0	2	3	4	5.60	8.10	53.66	7.48	12.82
307	7	200433302	Control	17-Jan-01	197	3	84.0	2	3	3	5.60	7.28	54.89	9.16	14.77
308	8	200441202	Control	17-Jan-01	185	3	85.5	2	3	2	5.60	7.64	55.50	7.50	14.13
309	9	200439902	Control	17-Jan-01	199	3	85.7	2	3	4	5.60	6.56	59.39	6.08	15.07
310	10	200439903	Control	17-Jan-01	190	3	81.5	2	3	5	5.70	6.94	59.37	6.48	15.52
311	11	200431708	Clone	17-Jan-01	180	3	79.5	2	3	4	5.60	5.94	55.28	8.29	14.76
312	12	200438607	Clone	17-Jan-01	195	3	81.0	2	3	3	5.80	7.69	56.71	8.34	14.19
313	13	200436604	Clone	17-Jan-01	185	3	83.5	2	3	3	5.70	7.82	57.52	7.20	13.85
314	14	200434101	Clone	17-Jan-01	186	3	81.0	2	3	1	5.60	7.39	50.74	9.06	12.21
315	15	200436607	Clone	17-Jan-01	180	3	84.5	2	3	2	5.70	7.70	60.60	5.39	14.65
316	16	200432507	Clone	17-Jan-01	193	3	83.3	2	3	3	5.60	6.17	55.25	8.47	15.45
317	17	200438610	Clone	17-Jan-01	190	3	82.5	2	3	5	5.90	7.43	61.35	7.08	15.25
318	18	200432008	Clone	17-Jan-01	203	3	79.5	2	3	3	5.70	6.56	55.90	7.21	13.60
319	19	200439505	Clone	17-Jan-01	189	3	80.0	2	3	3	5.70	7.04	57.67	6.31	13.67
320	20	200436901	Clone	17-Jan-01	180	3	82.3	2	3	2	5.60	8.60	57.96	7.30	14.45

Total Number	Day Order	Marc Animal ID	Treatment (Control/Clone)	Slaughter Date	Hot Carcass Weight/lbs	USDA CARCASS MUSCLE (1-3)	CARCASS LENGTH (cm)	FIRMNESS (1-3)	COLOR (1-6)	MARBLING (1-6, 10)	Longissimus pH (24 hr)	LOINEYE AREA (in ²)	hunter (L*)	hunter (a*)	hunter (B*)
321	21	200433007	Clone	17-Jan-01	186	3	81.7	2	3	3	5.70	7.36	54.04	8.28	13.23
322	22	200431503	Clone	17-Jan-01	185	3	83.0	2	3	2	5.60	6.83	59.32	8.01	15.47
323	23	200436001	Clone	17-Jan-01	187	3	85.5	2	3	2	5.70	7.44	56.59	7.09	13.31
324	24	200438101	Clone	17-Jan-01	179	3	85.5	2	3	3	5.60	7.24	54.15	7.88	13.59
325	25	200432304	Clone	17-Jan-01	189	3	83.5	2	3	2	5.60	7.53	57.62	8.52	15.34
326	26	200433709	Clone	17-Jan-01	184	3	82.0	2	3	3	5.80	8.06	54.65	8.18	14.48
327	27	200438602	Clone	17-Jan-01	186	3	83.2	2	3	3	5.60	7.65	55.78	8.36	14.79
328	28	200436905	Clone	17-Jan-01	179	3	81.8	2	3	3	5.60	7.41	59.09	5.98	14.43
329	29	200432003	Clone	17-Jan-01	175	3	82.7	2	3	1	5.60	7.56	53.84	8.34	13.57
330	30	200437803	Clone	17-Jan-01	188	3	81.7	2	3	5	5.90	6.05	54.19	8.15	14.50
331	31	200434606	Clone	17-Jan-01	179	3	81.0	2	3	2	5.60	6.81	52.62	7.89	13.42
332	32	200438609	Clone	17-Jan-01	188	3	81.8	2	3	4	5.60	6.55	55.28	8.98	15.09
333	1	200438705	Control	19-Jan-01	178	3	84.3	2	3	4	5.6	6.86	60.94	4.84	14.79
334	2	200433401	Control	19-Jan-01	189	3	88.0	2	3	2	5.6	8.07	53.95	8.82	14.05
335	3	200431803	Control	19-Jan-01	173	3	83.7	2	3	2	5.6	7.81	55.23	6.28	13.19
336	4	200438709	Control	19-Jan-01	177	3	83.0	2	3	4	5.6	6.22	60.41	5.14	13.91
337	5	200430507	Control	19-Jan-01	177	3	78.5	2	3	3	5.6	6.82	59.31	6.87	14.66
338	6	200434805	Control	19-Jan-01	170	3	85.7	2	3	4	5.6	5.70	55.63	7.82	14.85
339	7	200435304	Control	19-Jan-01	181	3	83.0	2	3	3	5.6	6.65	54.56	5.99	13.17
340	8	200439901	Control	19-Jan-01	182	3	85.5	2	3	5	5.7	5.35	60.87	5.85	14.58
341	9	200435307	Control	19-Jan-01	177	3	85.3	2	3	3	5.6	6.48	53.73	7.48	12.90
342	10	200433208	Clone	19-Jan-01	182	3	80.5	2	3	4	5.6	6.11	54.58	9.84	14.78
343	11	200439502	Clone	19-Jan-01	193	3	84.5	2	3	4	5.7	8.74	60.91	5.28	13.86
344	12	200438608	Clone	19-Jan-01	165	3	80.2	2	3	4	5.8	5.95	62.20	6.09	14.63
345	13	200433801	Clone	19-Jan-01	182	3	84.0	2	3	2	5.6	9.41	59.07	6.38	13.75
346	14	200430801	Clone	19-Jan-01	177	3	85.0	2	3	3	5.6	7.89	55.68	7.11	14.29
347	15	200436601	Clone	19-Jan-01	186	3	84.7	2	3	3	5.6	7.18	54.15	8.32	14.30
348	16	200430602	Clone	19-Jan-01	189	3	80.0	2	3	2	5.5	7.77	58.43	8.28	14.09
349	17	200436004	Clone	19-Jan-01	176	3	82.2	2	3	2	5.6	7.38	52.83	9.66	14.49
350	18	200436903	Clone	19-Jan-01	182	3	83.4	2	3	2	5.6	9.42	56.51	7.47	14.27
351	19	200433001	Clone	19-Jan-01	185	3	82.5	2	3	2	5.6	8.39	58.53	7.19	14.01
352	20	200433802	Clone	19-Jan-01	200	3	82.0	2	3	2	5.5	9.19	55.51	7.17	13.49

Total Number	Day Order	Marc Animal ID	Treatment (Control/Clone)	Slaughter Date	Hot Carcass Weight/lbs	USDA CARCASS MUSCLE (1-3)	CARCASS LENGTH (cm)	FIRMNESS (1-3)	COLOR (1-6)	MARBLING (1-6, 10)	Longissimus pH (24 hr)	LOINEYE AREA (in ²)	hunter (L*)	hunter (a*)	hunter (B*)
353	21	200438611	Clone	19-Jan-01	169	3	83.0	2	3	5	5.6	6.91	58.80	6.88	14.36
354	22	200430702	Clone	19-Jan-01	183	3	84.8	2	3	2	5.6	8.85	60.08	6.31	14.61
355	23	200431602	Clone	19-Jan-01	193	3	80.3	2	3	2	5.6	7.64	53.49	8.57	14.46
356	24	200434602	Clone	19-Jan-01	179	3	84.0	2	3	2	5.6	7.65	56.17	7.29	14.00
357	25	200434109	Clone	19-Jan-01	175	3	84.3	2	3	2	5.7	6.35	52.35	8.48	12.82
358	26	200433010	Clone	19-Jan-01	177	3	82.0	2	3	3	5.7	6.35	54.19	7.30	14.15
359	27	200434104	Clone	19-Jan-01	179	3	82.3	2	3	3	5.7	7.43	56.20	7.39	14.19
360	28	200431005	Clone	19-Jan-01	177	3	84.7	2	3	2	5.6	7.35	55.69	7.49	13.89
361	29	200434107	Clone	19-Jan-01	184	3	81.0	2	3	3	5.8	6.15	56.40	8.36	14.19
362	1	200441205	Control	25-Jan-01	180	3	78.7	2	3	3	5.70	6.50	56.72	7.04	14.16
363	2	200441211	Control	25-Jan-01	174	3	82.0	2	3	4	5.60	6.38	54.33	8.47	14.66
364	3	200441203	Control	25-Jan-01	187	3	83.0	2	3	3	5.70	6.46	55.67	6.76	14.15
365	4	200441201	Control	25-Jan-01	185	3	87.7	2	3	3	5.60	8.03	54.38	6.76	13.23
366	5	200438701	Control	25-Jan-01	180	3	84.7	2	3	2	5.60	7.73	57.38	6.79	13.29
367	6	200441209	Control	25-Jan-01	177	3	86.3	2	3	3	5.60	6.59	54.75	8.19	13.94
368	7	200439904	Control	25-Jan-01	167	3	82.8	2	3	4	5.60	5.45	58.62	6.84	14.19
369	8	200441207	Control	25-Jan-01	178	3	83.6	2	3	4	5.60	6.74	59.79	6.47	14.72
370	9	200436301	Control	25-Jan-01	183	3	85.0	2	3	2	5.60	7.00	60.45	5.96	13.60
371	10	200439204	Control	25-Jan-01	183	3	86.5	2	3	2	5.70	7.99	52.45	7.64	12.73
372	11	200434603	Clone	25-Jan-01	172	3	84.0	2	3	2	5.60	7.60	53.14	8.30	13.97
373	12	200434113	Clone	25-Jan-01	171	3	81.3	2	3	2	5.50	6.93	56.93	7.31	14.20
374	13	200438606	Clone	25-Jan-01	183	3	83.2	2	3	3	5.60	7.89	56.96	7.79	14.02
375	14	200436401	Clone	25-Jan-01	186	3	85.5	2	3	2	5.50	8.75	58.67	7.30	15.15
376	15	200434111	Clone	25-Jan-01	182	3	81.5	2	3	2	5.50	7.61	56.43	7.68	14.38
377	16	200438603	Clone	25-Jan-01	187	3	83.5	2	3	2	5.60	8.58	57.79	6.72	14.25
378	17	200433011	Clone	25-Jan-01	170	3	82.2	2	3	3	5.50	6.74	55.51	7.09	13.22
379	18	200439504	Clone	25-Jan-01	179	3	82.5	2	3	3	5.60	8.33	58.51	5.58	13.91
380	19	200437402	Clone	25-Jan-01	182	3	80.3	2	3	4	5.50	7.58	59.78	7.59	15.79
381	20	200439501	Clone	25-Jan-01	182	3	80.5	2	3	2	5.70	7.54	58.86	5.90	14.50
382	21	200436008	Clone	25-Jan-01	184	3	85.0	2	3	1	5.50	8.56	51.51	9.39	13.37
383	22	200438605	Clone	25-Jan-01	182	3	83.8	2	3	3	5.60	7.57	54.25	8.58	13.82
384	23	200434605	Clone	25-Jan-01	190	3	82.3	2	3	3	5.70	7.59	51.14	9.23	12.87

Total Number	Day Order	Marc Animal ID	Treatment (Control/Clone)	Slaughter Date	Hot Carcass Weight/lbs	USDA CARCASS MUSCLE (1-3)	CARCASS LENGTH (cm)	FIRMNESS (1-3)	COLOR (1-6)	MARBLING (1-6, 10)	Longissimus pH (24 hr)	LOINEYE AREA (in ²)	hunter (L*)	hunter (a*)	hunter (B*)
385	24	200436003	Clone	25-Jan-01	181	3	84.7	2	3	1	5.50	7.45	56.90	7.33	14.15
386	25	200437509	Clone	25-Jan-01	173	3	85.0	2	3	2	5.50	6.49	50.67	9.62	13.17
387	26	200432506	Clone	25-Jan-01	176	3	81.5	2	3	3	5.60	7.61	56.42	7.63	14.06
388	27	200433204	Clone	25-Jan-01	188	3	79.4	2	3	3	5.50	7.34	57.17	8.26	16.23
389	28	200436612	Clone	25-Jan-01	184	3	81.6	2	3	3	5.70	7.34	53.96	7.72	13.76
390	1	200438703	Control	01-Feb-01	176	3	84.5	2	3	3	5.70	6.34	56.71	9.35	15.76
391	2	200439401	Control	01-Feb-01	140	3	81.0	2	3	1	5.70	6.69	50.73	8.07	11.51
392	3	200431804	Control	01-Feb-01	98	3	75.0	2	2	1	5.60	5.12	57.21	6.57	14.39
393	4	200439404	Control	01-Feb-01	174	3	82.6	2	3	3	5.60	7.00	59.82	5.47	14.50
394	5	200438403	Control	01-Feb-01	188	3	81.7	2	3	2	5.60	8.96	55.94	6.55	13.56
395	6	200439503	Clone	01-Feb-01	187	3	79.3	2	3	3	5.80	7.81	57.21	5.47	13.52
396	7	200434102	Clone	01-Feb-01	197	3	84.5	2	3	3	5.60	7.33	51.79	8.13	12.42
397	8	200430603	Clone	01-Feb-01	181	3	80.7	2	3	3	5.70	7.86	54.85	7.13	13.93
398	9	200434604	Clone	01-Feb-01	175	3	83.0	2	3	2	5.50	7.53	52.93	7.76	13.36
399	10	200438604	Clone	01-Feb-01	181	3	80.0	2	3	3	5.60	9.16	58.81	6.85	14.65
400	11	200434105	Clone	01-Feb-01	175	3	82.3	2	3	2	5.60	6.88	50.92	8.49	12.97
401	12	200437401	Clone	01-Feb-01	186	3	80.0	2	3	4	5.70	7.49	58.11	7.28	14.98
402	13	200432306	Clone	01-Feb-01	162	3	84.3	2	3	3	5.60	6.34	54.84	7.58	13.96
403	14	200433708	Clone	01-Feb-01	158	3	78.2	2	3	2	5.60	8.24	57.64	7.67	14.13
404	15	200438102	Clone	01-Feb-01	189	3	81.3	2	3	4	5.70	7.01	55.99	8.36	15.13
405	16	200436403	Clone	01-Feb-01	146	3	78.0	2	2	2	5.60	6.15	55.90	7.09	15.00

Number	Animal	Probe Date	First Rib Backfat/mm	Last Rib Backfat/mm	Lumbar Vertebra/mm
1	200430503	12-Dec-00	20	14	13
2	200430508	12-Dec-00	25	17	17
3	200430604	12-Dec-00	30	21	19
4	200430706	12-Dec-00	27	22	23
5	200430707	12-Dec-00	28	19	19
6	200430709	12-Dec-00	18	15	14
7	200430710	12-Dec-00	28	19	18
8	200430711	12-Dec-00	26	23	21
9	200430804	12-Dec-00	27	18	19
10	200430805	12-Dec-00	21	16	15
11	200430902	12-Dec-00	32	21	22
12	200430905	12-Dec-00	28	22	21
13	200430906	12-Dec-00	34	22	21
14	200431006	12-Dec-00	28	22	21
15	200431009	12-Dec-00	25	18	19
16	200431404	12-Dec-00	20	13	13
17	200431408	12-Dec-00	21	14	15
18	200431411	12-Dec-00	28	21	21
19	200431502	12-Dec-00	24	17	18
20	200431505	12-Dec-00	29	22	24
21	200431608	12-Dec-00	28	14	15
22	200432405	12-Dec-00	28	17	18
23	200433409	12-Dec-00	28	19	21
24	200433509	12-Dec-00	23	14	16
25	200433510	12-Dec-00	25	19	18
26	200433605	12-Dec-00	23	16	17
27	200433807	12-Dec-00	26	19	18
28	200435309	12-Dec-00	28	21	24
29	200436009	12-Dec-00	26	20	22
30	200437301	12-Dec-00	26	19	18
31	200437307	12-Dec-00	26	20	19
32	200430605	14-Dec-00	24	18	18
33	200430607	14-Dec-00	30	21	19
34	200430609	14-Dec-00	20	16	19
35	200430705	14-Dec-00	20	16	15
36	200430708	14-Dec-00	23	16	17
37	200431008	14-Dec-00	26	17	17
38	200431405	14-Dec-00	17	13	13
39	200431407	14-Dec-00	19	13	13
40	200431501	14-Dec-00	18	14	16
41	200431504	14-Dec-00	32	23	21
42	200431606	14-Dec-00	25	17	16
43	200431802	14-Dec-00	20	17	17
44	200432609	14-Dec-00	26	23	20
45	200432903	14-Dec-00	21	14	17
46	200432908	14-Dec-00	30	19	19
47	200432910	14-Dec-00	22	18	22
48	200432911	14-Dec-00	23	15	17
49	200433201	14-Dec-00	26	20	20

Number	Animal	Probe Date	First Rib Backfat/mm	Last Rib Backfat/mm	Lumbar Vertebra/mm
50	200433301	14-Dec-00	22	17	17
51	200433601	14-Dec-00	17	13	15
52	200434001	14-Dec-00	20	15	19
53	200434202	14-Dec-00	17	13	14
54	200434301	14-Dec-00	20	17	19
55	200434507	14-Dec-00	24	19	19
56	200435303	14-Dec-00	17	14	15
57	200435408	14-Dec-00	28	21	21
58	200436011	14-Dec-00	18	13	14
59	200436012	14-Dec-00	28	19	18
60	200436909	14-Dec-00	26	15	16
61	200437303	14-Dec-00	24	17	17
62	200437305	14-Dec-00	24	18	19
63	200437306	14-Dec-00	21	16	17
64	200437308	14-Dec-00	27	20	20
65	200437505	14-Dec-00	24	17	18
66	200437506	14-Dec-00	24	19	21
67	200437507	14-Dec-00	27	19	19
68	200430703	19-Dec-00	17	11	11
69	200430903	19-Dec-00	27	21	19
70	200431003	19-Dec-00	18	13	13
71	200431011	19-Dec-00	20	13	12
72	200431406	19-Dec-00	17	11	13
73	200431604	19-Dec-00	21	15	17
74	200431610	19-Dec-00	19	15	15
75	200432004	19-Dec-00	23	17	17
76	200432103	19-Dec-00	24	17	16
77	200432106	19-Dec-00	25	17	18
78	200432108	19-Dec-00	27	21	19
79	200432308	19-Dec-00	22	20	18
80	200432310	19-Dec-00	24	19	20
81	200432404	19-Dec-00	22	15	16
82	200432406	19-Dec-00	27	18	19
83	200432608	19-Dec-00	24	19	21
84	200432704	19-Dec-00	21	16	15
85	200432705	19-Dec-00	25	20	19
86	200432706	19-Dec-00	23	16	17
87	200432905	19-Dec-00	19	14	15
88	200432912	19-Dec-00	19	16	16
89	200433205	19-Dec-00	22	18	19
90	200433603	19-Dec-00	18	13	13
91	200433702	19-Dec-00	28	20	19
92	200433711	19-Dec-00	24	18	19
93	200434002	19-Dec-00	20	14	14
94	200434207	19-Dec-00	33	24	20
95	200434305	19-Dec-00	20	15	12
96	200434309	19-Dec-00	24	17	16
97	200434401	19-Dec-00	27	17	18
98	200434402	19-Dec-00	25	16	17

Number	Animal	Probe Date	First Rib Backfat/mm	Last Rib Backfat/mm	Lumbar Vertebra/mm
99	200434403	19-Dec-00	36	25	23
100	200434508	19-Dec-00	22	17	16
101	200434512	19-Dec-00	27	17	18
102	200434601	19-Dec-00	19	13	13
103	200436609	19-Dec-00	28	20	18
104	200437304	19-Dec-00	25	22	20
105	200437504	19-Dec-00	23	19	17
106	200438409	19-Dec-00	23	19	18
107	200438506	19-Dec-00	22	17	19
108	200439211	19-Dec-00	19	14	16
109	200439405	19-Dec-00	27	19	18
110	200440604	19-Dec-00	22	14	15
111	200430510	20-Dec-00	22	16	17
112	200431805	20-Dec-00	20	16	18
113	200432601	20-Dec-00	21	17	18
114	200432602	20-Dec-00	28	19	18
115	200432607	20-Dec-00	25	17	17
116	200432610	20-Dec-00	23	16	17
117	200432907	20-Dec-00	28	19	22
118	200432913	20-Dec-00	24	17	18
119	200433306	20-Dec-00	31	22	20
120	200433307	20-Dec-00	31	22	21
121	200433405	20-Dec-00	18	14	15
122	200433408	20-Dec-00	22	15	16
123	200433505	20-Dec-00	19	15	14
124	200433511	20-Dec-00	20	15	15
125	200433512	20-Dec-00	19	14	16
126	200434003	20-Dec-00	23	17	18
127	200434005	20-Dec-00	24	18	19
128	200434203	20-Dec-00	26	19	19
129	200434206	20-Dec-00	25	19	19
130	200434806	20-Dec-00	25	19	19
131	200434807	20-Dec-00	23	18	18
132	200434808	20-Dec-00	24	21	22
133	200434901	20-Dec-00	20	17	16
134	200435302	20-Dec-00	22	15	15
135	200435311	20-Dec-00	32	23	21
136	200435404	20-Dec-00	23	18	17
137	200435407	20-Dec-00	29	21	22
138	200438408	20-Dec-00	20	15	14
139	200438511	20-Dec-00	26	17	16
140	200438513	20-Dec-00	27	20	19
141	200430704	26-Dec-00	20	15	16
142	200430802	26-Dec-00	19	15	15
143	200431401	26-Dec-00	15	10	11
144	200431403	26-Dec-00	21	15	16
145	200431703	26-Dec-00	21	16	17
146	200431704	26-Dec-00	24	20	19
147	200431706	26-Dec-00	33	23	21

Number	Animal	Probe Date	First Rib Backfat/mm	Last Rib Backfat/mm	Lumbar Vertebra/mm
148	200431801	26-Dec-00	20	16	16
149	200432105	26-Dec-00	27	16	15
150	200432402	26-Dec-00	23	17	16
151	200432702	26-Dec-00	20	16	18
152	200433206	26-Dec-00	22	15	18
153	200433602	26-Dec-00	23	18	19
154	200433803	26-Dec-00	26	15	14
155	200433809	26-Dec-00	29	21	21
156	200434007	26-Dec-00	28	20	20
157	200434809	26-Dec-00	25	19	21
158	200434902	26-Dec-00	23	16	20
159	200435308	26-Dec-00	22	18	18
160	200435412	26-Dec-00	25	18	19
161	200436010	26-Dec-00	22	15	15
162	200436303	26-Dec-00	23	17	19
163	200436309	26-Dec-00	24	19	21
164	200436402	26-Dec-00	23	14	15
165	200437309	26-Dec-00	25	19	18
166	200437404	26-Dec-00	25	20	22
167	200437801	26-Dec-00	23	19	18
168	200437802	26-Dec-00	23	20	20
169	200438107	26-Dec-00	32	23	26
170	200438401	26-Dec-00	15	12	12
171	200438404	26-Dec-00	20	17	15
172	200438406	26-Dec-00	22	17	17
173	200438505	26-Dec-00	18	14	15
174	200439210	26-Dec-00	15	13	13
175	200440605	26-Dec-00	25	20	19
176	200430606	28-Dec-00	29	24	22
177	200430803	28-Dec-00	27	18	17
178	200431607	28-Dec-00	31	21	21
179	200432002	28-Dec-00	25	17	17
180	200432006	28-Dec-00	24	18	17
181	200432101	28-Dec-00	19	16	16
182	200432302	28-Dec-00	20	15	16
183	200432501	28-Dec-00	23	19	17
184	200432503	28-Dec-00	26	17	17
185	200432603	28-Dec-00	26	17	18
186	200432604	28-Dec-00	24	18	18
187	200432909	28-Dec-00	20	17	15
188	200433207	28-Dec-00	33	26	27
189	200433404	28-Dec-00	20	15	15
190	200433604	28-Dec-00	22	15	15
191	200433703	28-Dec-00	24	18	19
192	200433704	28-Dec-00	24	17	17
193	200433710	28-Dec-00	30	22	21
194	200433804	28-Dec-00	23	20	21
195	200433808	28-Dec-00	25	21	20
196	200434004	28-Dec-00	24	18	18

Number	Animal	Probe Date	First Rib Backfat/mm	Last Rib Backfat/mm	Lumbar Vertebra/mm
197	200434201	28-Dec-00	22	15	14
198	200434205	28-Dec-00	22	17	16
199	200434308	28-Dec-00	30	19	19
200	200434310	28-Dec-00	29	21	22
201	200434802	28-Dec-00	21	15	16
202	200435306	28-Dec-00	31	21	20
203	200435401	28-Dec-00	25	16	17
204	200435402	28-Dec-00	25	18	19
205	200435413	28-Dec-00	25	17	21
206	200435802	28-Dec-00	32	22	24
207	200436311	28-Dec-00	28	21	22
208	200436405	28-Dec-00	21	16	17
209	200436911	28-Dec-00	26	18	18
210	200438407	28-Dec-00	17	13	13
211	200438410	28-Dec-00	21	16	15
212	200438412	28-Dec-00	24	16	17
213	200438510	28-Dec-00	18	14	15
214	200439202	28-Dec-00	18	14	15
215	200439206	28-Dec-00	23	17	19
216	200439212	28-Dec-00	19	14	15
217	200439406	28-Dec-00	26	19	19
218	200439409	28-Dec-00	23	12	17
219	200440602	28-Dec-00	26	17	20
220	200440908	28-Dec-00	27	18	21
221	200440910	28-Dec-00	31	26	26
222	200430502	2-Jan-01	20	15	15
223	200430509	2-Jan-01	24	16	17
224	200430608	2-Jan-01	23	16	15
225	200431007	2-Jan-01	19	13	12
226	200431012	2-Jan-01	30	21	22
227	200431402	2-Jan-01	22	16	16
228	200431409	2-Jan-01	24	18	19
229	200432007	2-Jan-01	28	21	20
230	200432104	2-Jan-01	25	17	20
231	200432109	2-Jan-01	22	18	18
232	200433005	2-Jan-01	26	18	19
233	200433203	2-Jan-01	32	18	21
234	200433402	2-Jan-01	21	17	17
235	200433403	2-Jan-01	26	19	21
236	200433501	2-Jan-01	17	13	13
237	200433504	2-Jan-01	18	13	14
238	200433701	2-Jan-01	29	22	24
239	200433712	2-Jan-01	29	20	19
240	200433806	2-Jan-01	27	21	21
241	200434009	2-Jan-01	26	17	20
242	200434210	2-Jan-01	32	22	21
243	200434510	2-Jan-01	28	20	19
244	200434608	2-Jan-01	18	14	15
245	200434801	2-Jan-01	34	23	20

Number	Animal	Probe Date	First Rib Backfat/mm	Last Rib Backfat/mm	Lumbar Vertebra/mm
246	200435409	2-Jan-01	30	20	25
247	200436006	2-Jan-01	18	13	12
248	200436305	2-Jan-01	15	12	13
249	200436310	2-Jan-01	26	18	20
250	200436404	2-Jan-01	18	14	13
251	200436610	2-Jan-01	28	22	19
252	200437501	2-Jan-01	23	16	16
253	200437510	2-Jan-01	25	19	20
254	200437804	2-Jan-01	28	22	22
255	200438414	2-Jan-01	30	22	21
256	200438503	2-Jan-01	23	17	19
257	200438504	2-Jan-01	27	19	18
258	200439408	2-Jan-01	28	19	21
259	200440601	2-Jan-01	20	16	16
260	200440606	2-Jan-01	37	23	22
261	200440607	2-Jan-01	32	26	23
262	200440901	2-Jan-01	20	15	17
263	200440906	2-Jan-01	19	14	15
264	200430501	5-Jan-01	19	14	13
265	200431410	5-Jan-01	21	16	13
266	200431506	5-Jan-01	23	16	21
267	200431603	5-Jan-01	15	10	14
268	200432001	5-Jan-01	21	17	15
269	200432005	5-Jan-01	31	25	24
270	200432102	5-Jan-01	21	17	14
271	200432307	5-Jan-01	24	20	20
272	200432311	5-Jan-01	28	20	23
273	200432312	5-Jan-01	24	19	17
274	200432401	5-Jan-01	20	15	13
275	200432502	5-Jan-01	21	15	14
276	200432504	5-Jan-01	22	18	17
277	200433003	5-Jan-01	21	15	13
278	200433502	5-Jan-01	17	12	12
279	200433503	5-Jan-01	20	11	13
280	200433507	5-Jan-01	20	13	8
281	200434008	5-Jan-01	20	16	18
282	200434303	5-Jan-01	14	11	13
283	200434306	5-Jan-01	18	14	15
284	200435301	5-Jan-01	13	12	10
285	200435305	5-Jan-01	19	12	14
286	200435410	5-Jan-01	22	16	18
287	200435411	5-Jan-01	24	18	22
288	200436002	5-Jan-01	21	15	12
289	200436013	5-Jan-01	26	21	18
290	200436902	5-Jan-01	18	15	12
291	200437302	5-Jan-01	23	19	15
292	200437408	5-Jan-01	30	22	21
293	200438105	5-Jan-01	28	23	18
294	200438106	5-Jan-01	29	25	20

Number	Animal	Probe Date	First Rib Backfat/mm	Last Rib Backfat/mm	Lumbar Vertebra/mm
295	200438706	5-Jan-01	24	17	21
296	200439208	5-Jan-01	19	14	17
297	200440603	5-Jan-01	25	18	24
298	200440911	5-Jan-01	18	13	14
299	200430506	9-Jan-01	16	12	12
300	200430610	9-Jan-01	24	19	20
301	200430701	9-Jan-01	18	13	14
302	200431701	9-Jan-01	22	16	17
303	200432407	9-Jan-01	20	16	17
304	200432605	9-Jan-01	25	18	18
305	200432904	9-Jan-01	20	14	16
306	200432906	9-Jan-01	23	18	18
307	200433002	9-Jan-01	17	13	14
308	200433202	9-Jan-01	20	15	16
309	200433506	9-Jan-01	21	16	16
310	200434208	9-Jan-01	34	23	21
311	200434209	9-Jan-01	28	19	20
312	200434302	9-Jan-01	18	16	17
313	200434501	9-Jan-01	22	13	15
314	200434503	9-Jan-01	20	14	13
315	200434504	9-Jan-01	23	17	18
316	200434509	9-Jan-01	22	16	15
317	200434607	9-Jan-01	27	19	20
318	200434803	9-Jan-01	22	19	20
319	200434804	9-Jan-01	25	18	18
320	200434903	9-Jan-01	24	18	19
321	200434904	9-Jan-01	18	14	15
322	200434905	9-Jan-01	24	18	21
323	200435310	9-Jan-01	26	18	22
324	200435403	9-Jan-01	26	18	18
325	200435405	9-Jan-01	23	16	16
326	200435801	9-Jan-01	30	22	22
327	200436007	9-Jan-01	24	19	22
328	200436608	9-Jan-01	26	19	20
329	200436904	9-Jan-01	17	12	12
330	200436910	9-Jan-01	26	19	20
331	200437403	9-Jan-01	38	20	21
332	200437409	9-Jan-01	31	20	19
333	200437410	9-Jan-01	28	19	21
334	200437502	9-Jan-01	17	13	14
335	200438402	9-Jan-01	25	18	18
336	200438501	9-Jan-01	21	17	17
337	200438502	9-Jan-01	24	18	18
338	200439201	9-Jan-01	18	14	15
339	200439209	9-Jan-01	22	15	16
340	200439215	9-Jan-01	21	17	17
341	200440902	9-Jan-01	22	18	18
342	200440912	9-Jan-01	26	18	18
343	200440913	9-Jan-01	28	19	23

Number	Animal	Probe Date	First Rib Backfat/mm	Last Rib Backfat/mm	Lumbar Vertebra/mm
344	200430504	11-Jan-01	21	16	15
345	200430611	11-Jan-01	29	24	21
346	200431002	11-Jan-01	19	14	13
347	200431004	11-Jan-01	17	14	14
348	200431010	11-Jan-01	22	13	15
349	200431601	11-Jan-01	24	16	16
350	200431605	11-Jan-01	26	17	15
351	200431702	11-Jan-01	31	20	22
352	200432301	11-Jan-01	23	19	18
353	200432303	11-Jan-01	22	18	17
354	200432403	11-Jan-01	19	13	13
355	200432901	11-Jan-01	24	17	17
356	200433006	11-Jan-01	28	21	21
357	200433008	11-Jan-01	22	17	16
358	200433706	11-Jan-01	26	16	17
359	200434006	11-Jan-01	24	21	22
360	200434106	11-Jan-01	32	21	22
361	200434108	11-Jan-01	31	20	23
362	200434110	11-Jan-01	31	19	21
363	200434304	11-Jan-01	34	20	21
364	200434502	11-Jan-01	21	16	16
365	200434506	11-Jan-01	24	19	22
366	200434511	11-Jan-01	25	16	17
367	200435803	11-Jan-01	28	18	20
368	200436005	11-Jan-01	18	14	15
369	200436312	11-Jan-01	30	20	21
370	200436602	11-Jan-01	23	17	16
371	200436603	11-Jan-01	20	15	16
372	200436605	11-Jan-01	27	20	19
373	200436606	11-Jan-01	29	18	19
374	200436912	11-Jan-01	23	17	16
375	200436913	11-Jan-01	26	17	21
376	200437406	11-Jan-01	38	24	25
377	200438104	11-Jan-01	24	17	17
378	200440903	11-Jan-01	23	16	15
379	200431503	17-Jan-01	26	22	23
380	200431708	17-Jan-01	31	19	19
381	200432003	17-Jan-01	19	15	16
382	200432008	17-Jan-01	32	25	23
383	200432304	17-Jan-01	23	15	16
384	200432507	17-Jan-01	30	20	17
385	200433007	17-Jan-01	20	17	16
386	200433302	17-Jan-01	25	17	18
387	200433303	17-Jan-01	25	19	23
388	200433406	17-Jan-01	23	16	19
389	200433709	17-Jan-01	25	18	19
390	200434101	17-Jan-01	27	20	21
391	200434606	17-Jan-01	21	16	17
392	200436001	17-Jan-01	21	13	14

Number	Animal	Probe Date	First Rib Backfat/mm	Last Rib Backfat/mm	Lumbar Vertebra/mm
393	200436604	17-Jan-01	21	18	18
394	200436607	17-Jan-01	21	15	16
395	200436901	17-Jan-01	19	15	15
396	200436905	17-Jan-01	23	14	16
397	200437803	17-Jan-01	32	27	26
398	200438101	17-Jan-01	28	21	20
399	200438602	17-Jan-01	20	17	17
400	200438607	17-Jan-01	28	22	23
401	200438609	17-Jan-01	27	21	22
402	200438610	17-Jan-01	26	19	22
403	200438707	17-Jan-01	25	21	21
404	200439205	17-Jan-01	23	16	17
405	200439505	17-Jan-01	32	20	20
406	200439902	17-Jan-01	28	21	24
407	200439903	17-Jan-01	27	21	21
408	200440907	17-Jan-01	30	20	24
409	200440909	17-Jan-01	25	17	18
410	200441202	17-Jan-01	19	14	15
411	200430507	19-Jan-01	26	18	21
412	200430602	19-Jan-01	18	15	15
413	200430702	19-Jan-01	20	14	13
414	200430801	19-Jan-01	18	13	12
415	200431005	19-Jan-01	20	13	13
416	200431602	19-Jan-01	28	19	19
417	200431803	19-Jan-01	21	16	16
418	200432309	19-Jan-01	23	14	17
419	200432703	19-Jan-01	24	15	15
420	200433001	19-Jan-01	18	15	16
421	200433004	19-Jan-01	22	16	16
422	200433010	19-Jan-01	27	23	24
423	200433208	19-Jan-01	29	20	23
424	200433401	19-Jan-01	19	14	17
425	200433707	19-Jan-01	26	23	23
426	200433801	19-Jan-01	21	17	18
427	200433802	19-Jan-01	34	21	22
428	200434104	19-Jan-01	27	18	20
429	200434107	19-Jan-01	25	18	19
430	200434109	19-Jan-01	31	19	19
431	200434112	19-Jan-01	28	19	21
432	200434602	19-Jan-01	19	13	13
433	200434805	19-Jan-01	24	18	19
434	200435304	19-Jan-01	22	18	19
435	200435307	19-Jan-01	23	18	19
436	200436004	19-Jan-01	22	15	16
437	200436302	19-Jan-01	26	17	17
438	200436307	19-Jan-01	25	19	17
439	200436601	19-Jan-01	24	21	19
440	200436611	19-Jan-01	21	15	17
441	200436903	19-Jan-01	17	15	14

Number	Animal	Probe Date	First Rib Backfat/mm	Last Rib Backfat/mm	Lumbar Vertebra/mm
442	200436907	19-Jan-01	25	18	19
443	200437407	19-Jan-01	29	21	19
444	200437411	19-Jan-01	33	19	22
445	200438405	19-Jan-01	20	16	17
446	200438411	19-Jan-01	24	15	17
447	200438413	19-Jan-01	18	14	15
448	200438508	19-Jan-01	27	21	19
449	200438509	19-Jan-01	23	15	16
450	200438608	19-Jan-01	22	16	18
451	200438611	19-Jan-01	28	19	23
452	200438705	19-Jan-01	19	14	17
453	200438709	19-Jan-01	27	17	20
454	200439207	19-Jan-01	22	15	16
455	200439403	19-Jan-01	28	18	19
456	200439407	19-Jan-01	26	20	19
457	200439502	19-Jan-01	20	14	17
458	200439901	19-Jan-01	28	17	21
459	200441204	19-Jan-01	26	19	20
460	200441206	19-Jan-01	33	19	20
461	200441208	19-Jan-01	27	19	19
462	200432506	25-Jan-01	24	17	18
463	200433011	25-Jan-01	25	17	16
464	200433204	25-Jan-01	26	18	16
465	200434111	25-Jan-01	26	17	17
466	200434113	25-Jan-01	29	21	19
467	200434114	25-Jan-01	28	18	20
468	200434603	25-Jan-01	15	11	12
469	200434605	25-Jan-01	19	15	16
470	200436003	25-Jan-01	16	12	13
471	200436008	25-Jan-01	16	12	13
472	200436301	25-Jan-01	23	17	18
473	200436304	25-Jan-01	27	20	18
474	200436306	25-Jan-01	26	21	20
475	200436401	25-Jan-01	16	12	12
476	200436612	25-Jan-01	22	15	15
477	200436906	25-Jan-01	18	12	14
478	200437402	25-Jan-01	30	21	20
479	200437405	25-Jan-01	23	17	18
480	200437509	25-Jan-01	20	15	16
481	200438603	25-Jan-01	23	18	19
482	200438605	25-Jan-01	20	15	16
483	200438606	25-Jan-01	21	18	19
484	200438701	25-Jan-01	23	16	15
485	200438704	25-Jan-01	24	15	16
486	200439203	25-Jan-01	18	14	15
487	200439204	25-Jan-01	19	13	15
488	200439501	25-Jan-01	24	18	18
489	200439504	25-Jan-01	23	16	16
490	200439904	25-Jan-01	25	20	17

Number	Animal	Probe Date	First Rib Backfat/mm	Last Rib Backfat/mm	Lumbar Vertebra/mm
491	200440609	25-Jan-01	32	22	19
492	200440905	25-Jan-01	22	16	16
493	200441201	25-Jan-01	23	15	19
494	200441203	25-Jan-01	28	19	18
495	200441205	25-Jan-01	28	20	19
496	200441207	25-Jan-01	24	19	18
497	200441209	25-Jan-01	22	18	18
498	200441210	25-Jan-01	28	18	20
499	200441211	25-Jan-01	26	18	20
500	200430505	1-Feb-01	19	12	13
501	200430603	1-Feb-01	19	13	14
502	200431804	1-Feb-01	13	10	10
503	200432305	1-Feb-01	25	16	15
504	200432306	1-Feb-01	17	13	13
505	200433304	1-Feb-01	21	14	17
506	200433305	1-Feb-01	24	18	18
507	200433407	1-Feb-01	22	17	17
508	200433705	1-Feb-01	21	16	16
509	200433708	1-Feb-01	19	13	15
510	200434102	1-Feb-01	28	20	19
511	200434103	1-Feb-01	24	16	17
512	200434105	1-Feb-01	27	19	18
513	200434604	1-Feb-01	18	12	12
514	200436403	1-Feb-01	15	10	9
515	200436908	1-Feb-01	19	14	15
516	200437401	1-Feb-01	24	19	20
517	200438102	1-Feb-01	28	19	19
518	200438103	1-Feb-01	25	16	18
519	200438403	1-Feb-01	22	16	18
520	200438601	1-Feb-01	24	19	19
521	200438604	1-Feb-01	20	14	13
522	200438702	1-Feb-01	20	14	17
523	200438703	1-Feb-01	25	16	19
524	200439401	1-Feb-01	12	9	10
525	200439404	1-Feb-01	19	14	14
526	200439503	1-Feb-01	29	23	24

SubstDescription	Units	ME2005-002293 / 200431006	ME2005-000301 / 200432405	ME2005-000951 / 200430605	ME2005-000952 / 200430607	ME2005-000966 / 200434001	ME2005-000969 / 200434202	ME2005-000971 / 200434507	ME2005-002222 / 200433305	ME2005-002223 / 200434206	ME2005-002227 / 200438513	ME2005-003124 / 200431801 Z7-Dec-04	ME2005-003126 / 200432402 Z7-Dec-04	ME2005-003135 / 200435308 Z7-Dec-04	ME2005-003141 / 200437309 Z7-Dec-04	ME2005-003151 / 200439210 Z7-Dec-04	ME2005-003153 / 200430606 Z9-Dec-04	ME2005-003177 / 200435413 Z9-Dec-04	ME2005-003199 / 200430902 Z3-Jan-05	ME2005-003200 / 200430909 Z3-Jan-05	ME2005-003201 / 200430608 Z3-Jan-05	ME2005-003202 / 200431007 Z3-Jan-05	ME2005-003203 / 200431012 Z3-Jan-05	ME2005-003204 / 200431402 Z3-Jan-05	ME2005-003205 / 200431409 Z3-Jan-05
Alanine	%	1.30	1.21	1.35	1.33	1.35	1.36	1.36	1.34	1.49	1.53	1.37	1.61	1.41	1.32	1.38	1.37	1.36	1.45	1.39	1.32	1.38	1.35	1.34	1.42
Arginine	%	1.42	1.34	1.54	1.53	1.63	1.62	1.63	1.42	1.69	1.72	1.48	1.86	1.63	1.55	1.58	1.54	1.51	1.73	1.63	1.54	1.56	1.61	1.61	1.56
Aspartic Acid	%	2.14	1.98	2.21	2.21	2.27	2.22	2.32	2.06	2.51	2.56	2.18	2.74	2.35	2.25	2.26	2.18	2.28	2.48	2.34	2.18	2.36	2.27	2.26	2.39
Cystine	%	0.23	0.23	0.25	0.23	0.25	0.26	0.28	0.25	0.25	0.24	0.25	0.27	0.25	0.25	0.24	0.20	0.24	0.25	0.25	0.26	0.26	0.26	0.26	0.27
Glutamic Acid	%	3.34	3.14	3.60	3.62	3.75	3.58	3.80	3.27	4.00	4.06	3.53	4.41	3.76	3.59	3.67	3.50	3.73	4.08	3.74	3.55	3.86	3.79	3.63	3.80
Glycine	%	0.97	0.92	1.14	1.06	1.16	1.31	1.08	1.27	1.09	1.16	0.95	1.18	1.23	1.05	1.19	1.30	1.05	1.09	1.10	1.05	1.06	1.05	1.14	1.08
Histidine	%	0.96	0.89	0.93	0.94	0.95	0.98	0.94	0.83	1.07	1.03	0.95	1.10	1.02	1.00	0.93	0.87	0.93	1.07	1.03	0.96	1.06	0.95	1.04	0.93
Isoleucine	%	0.98	0.93	1.07	1.07	1.10	1.04	1.10	0.76	1.17	1.15	1.01	1.25	1.11	1.10	0.94	0.88	0.93	1.20	1.13	1.05	1.14	1.10	1.11	0.85
Leucine	%	1.81	1.65	1.85	1.85	1.89	1.81	1.92	1.63	2.05	2.05	1.82	2.22	1.92	1.88	1.85	1.77	1.87	2.08	1.96	1.84	1.98	1.91	1.91	1.85
Lysine, Total	%	1.89	1.79	2.04	2.05	2.11	2.00	2.14	1.77	2.28	2.28	1.99	2.47	2.08	2.07	2.02	1.91	2.01	2.28	2.13	2.01	2.18	2.11	2.11	2.12
Methionine	%	0.57	0.55	0.61	0.59	0.63	0.65	0.71	0.63	0.61	0.58	0.64	0.67	0.59	0.62	0.51	0.59	0.61	0.62	0.64	0.63	0.62	0.65	0.64	0.64
Phenylalanine	%	0.86	0.81	0.91	0.91	0.93	0.91	0.95	0.83	1.09	1.09	0.93	1.14	0.91	0.89	0.91	0.88	0.89	0.97	0.92	0.88	0.97	0.95	0.91	0.96
Proline	%	1.08	0.94	1.02	0.94	1.11	1.19	1.10	1.18	1.19	1.23	0.88	1.18	1.14	1.04	1.05	1.06	0.96	1.04	1.11	1.01	1.04	1.04	1.10	1.11
Serine	%	0.89	0.83	0.91	0.90	0.91	0.91	0.96	0.88	0.98	1.02	0.88	1.07	0.98	0.92	0.98	0.96	0.99	1.01	0.95	0.91	0.93	0.92	0.92	1.00
Threonine	%	1.02	0.94	1.06	1.07	1.11	1.05	1.12	0.98	1.18	1.21	1.09	1.31	1.07	1.04	1.06	1.01	1.08	1.14	1.10	1.01	1.11	1.05	1.06	1.12
Tyrosine	%	0.73	0.70	0.80	0.80	0.81	0.78	0.85	0.71	0.88	0.89	0.79	0.96	0.80	0.79	0.80	0.76	0.80	0.85	0.80	0.78	0.86	0.83	0.78	0.83
Valine	%	1.04	1.00	1.13	1.13	1.16	1.13	1.15	0.84	1.25	1.22	1.06	1.31	1.19	1.17	1.01	0.97	0.98	1.24	1.17	1.12	1.21	1.14	1.19	0.93
Calcium	%	0.0063	0.0061	0.0058	0.0056	0.0054	0.0057	0.0061	0.0059	0.0051	0.0046	0.0045	0.0048	0.0050	0.0050	0.0061	0.0051	0.0050	0.0041	0.0051	0.0067	0.0040	0.0049	0.0048	0.0048
Cholesterol	mg/100 g	47.3	51.7	56.7	54.1	49.0	64.9	91.1	57.7	57.8	61.3	63.3	60.7	59.6	56.4	62.3	62.1	53.8	63.8	63.1	61.6	62.4	65.9	59.7	58.1
C08:0 Octanoic (Caprylic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C10:0 Decanoic (Capric)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C11:0 Undecanoic (Hendecanoic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C12:0 Dodecanoic (Lauroic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C14:0 Tetradecanoic (Myristic)	%	0.06	0.07	0.08	0.11	0.10	0.08	0.07	0.05	0.06	0.10	0.09	0.08	0.07	0.11	0.10	0.10	0.10	0.07	0.07	0.06	0.07	0.09	0.04	0.03
C14:1 Tetradecenoic (Myristoleic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C15:0 Pentadecanoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C15:1 Pentadecenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C16:0 Hexadecanoic (Palmitic)	%	0.95	1.19	1.20	1.98	1.56	1.19	1.10	0.75	1.03	1.61	1.56	1.40	1.10	2.00	1.62	1.74	1.81	1.29	1.14	0.88	1.08	1.42	0.65	0.45
C16:1 Hexadecenoic (Palmitoleic)	%	0.12	0.14	0.16	0.20	0.18	0.16	0.13	0.10	0.13	0.21	0.17	0.17	0.13	0.22	0.17	0.20	0.18	0.12	0.12	0.11	0.13	0.18	0.08	0.06
C17:0 Heptadecanoic (Margaric)	%	<0.01	<0.01	<0.01	0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C17:1 Heptadecenoic (Margaroleic)	%	<0.01	<0.01	<0.01	0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C18:0 Octadecanoic (Stearic)	%	0.44	0.57	0.53	1.03	0.72	0.53	0.55	0.34	0.50	0.74	0.77	0.68	0.50	1.05	0.81	0.85	0.95	0.68	0.54	0.41	0.48	0.64	0.27	0.21
C18:1 Octadecenoic (Oleic)	%	1.57	1.86	1.99	3.36	2.40	2.01	1.83	1.37	1.67	2.47	2.32	2.31	1.76	2.80	2.39	2.74	2.82	1.96	1.88	1.27	1.71	2.23	1.11	0.79
C18:2 Octadecadienoic (Linoleic)	%	0.28	0.36	0.24	0.58	0.41	0.32	0.31	0.22	0.23	0.35	0.46	0.40	0.19	0.24	0.26	0.27	0.31	0.16	0.21	0.13	0.18	0.23	0.13	0.11
C18:3 Octadecatrienoic (Linolenic)	%	0.01	0.02	0.01	0.03	0.01	0.01	0.01	<0.01	0.01	0.02	0.02	0.02	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C18:4 Octadecatetraenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:0 Eicosanoic (Arachidic)	%	<0.01	0.01	0.01	0.02	0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	0.01	<0.01	0.02	0.01	0.02	0.02	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:1 Eicosenoic (Gadoleic)	%	0.04	0.05	0.05	0.08	0.05	0.04	0.04	0.01	0.04	0.03	0.03	0.06	0.05	0.06	0.07	0.07	0.07	0.07	0.06	0.04	0.05	0.06	0.03	0.03
C20:2 Eicosadienoic	%	0.01	0.02	0.01	0.03	0.02	0.02	0.02	0.01	0.01	0.02	0.02	0.02	<0.01	0.01	0.01	0.01	0.02	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:3 Eicosatrienoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:4 Eicosatetraenoic (Arachidonic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:5 Eicosapentaenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C21:5 Heneicosapentaenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C22:0 Docosanoic (Behenic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C22:1 Docosenoic (Erucic)	%	<0.01	<0.01	<0.0																					

SubstestDescription	Units	ME2005-003229 / 200440906 3-Jan-05	ME2005-003227 / 200440901 3-Jan-05	ME2005-003228 / 200440904 3-Jan-05	ME2005-003226 / 200437804 3-Jan-05	ME2005-003225 / 200437803 3-Jan-05	ME2005-003224 / 200437510 3-Jan-05	ME2005-003223 / 200437301 3-Jan-05	ME2005-003222 / 200436610 3-Jan-05	ME2005-003221 / 200436404 3-Jan-05	ME2005-003220 / 200436310 3-Jan-05	ME2005-003219 / 200436305 3-Jan-05	ME2005-003218 / 200436006 3-Jan-05	ME2005-003217 / 200435409 3-Jan-05	ME2005-003216 / 200434608 3-Jan-05	ME2005-003215 / 200434009 3-Jan-05	ME2005-003214 / 200433906 3-Jan-05	ME2005-003213 / 200433712 3-Jan-05	ME2005-003212 / 200433604 3-Jan-05	ME2005-003211 / 200433501 3-Jan-05	ME2005-003210 / 200433403 3-Jan-05	ME2005-003209 / 200433402 3-Jan-05	ME2005-003208 / 200433203 3-Jan-05	ME2005-003207 / 200432104 3-Jan-05	ME2005-003206 / 200432007 3-Jan-05	
Alanine	%	1.37	1.34	1.38	1.40	1.39	1.38	1.42	1.32	1.31	1.37	1.44	1.34	1.39	1.33	1.31	1.40	1.25	1.31	1.40	1.34	1.39	1.38	1.40	1.40	
Arginine	%	1.63	1.65	1.66	1.70	1.69	1.68	1.71	1.60	1.60	1.63	1.69	1.60	1.65	1.57	1.54	1.61	1.53	1.54	1.65	1.58	1.62	1.64	1.64	1.64	1.64
Aspartic Acid	%	2.31	2.24	2.30	2.29	2.28	2.32	2.33	2.22	2.21	2.26	2.31	2.18	2.29	2.19	2.21	2.23	1.90	2.20	2.34	2.20	2.21	2.30	2.30	2.27	
Cystine	%	0.25	0.25	0.23	0.25	0.25	0.26	0.26	0.24	0.24	0.26	0.25	0.24	0.24	0.28	0.24	0.24	0.24	0.26	0.25	0.23	0.24	0.25	0.24	0.24	
Glutamic Acid	%	3.66	3.61	3.63	3.77	3.76	3.77	3.83	3.61	3.57	3.64	3.71	3.49	3.72	3.66	3.67	3.72	3.10	3.59	3.86	3.60	3.65	3.81	3.80	3.56	
Glycine	%	1.11	1.04	1.12	1.13	1.13	1.06	1.15	0.98	1.03	1.18	1.31	1.16	1.18	1.02	1.00	1.24	0.93	1.00	1.07	1.09	1.27	1.08	1.11	1.27	
Histidine	%	1.05	1.02	1.03	1.04	1.01	0.97	1.04	1.05	0.98	0.99	1.03	0.99	0.96	0.98	0.96	0.89	0.99	0.98	1.08	0.98	0.96	1.01	0.99	0.91	
Isoleucine	%	1.12	1.13	1.14	1.12	1.11	1.13	1.20	1.16	1.12	1.13	1.15	1.04	1.06	1.06	1.03	1.02	1.00	1.04	1.11	1.10	1.06	1.12	1.09	1.01	
Leucine	%	1.92	1.89	1.91	1.93	1.92	1.93	1.98	1.90	1.86	1.87	1.91	1.80	1.89	1.90	1.87	1.88	1.81	1.86	1.98	1.88	1.86	1.96	1.95	1.83	
Lysine, Total	%	2.12	2.05	2.05	2.08	2.07	2.09	2.13	2.02	1.98	2.01	2.03	1.92	2.04	2.06	2.04	2.07	1.98	2.01	2.13	2.01	2.02	2.10	2.12	2.03	
Methionine	%	0.59	0.59	0.54	0.60	0.61	0.63	0.64	0.56	0.59	0.67	0.61	0.57	0.67	0.73	0.62	0.60	0.59	0.65	0.63	0.57	0.59	0.60	0.58	0.61	
Phenylalanine	%	0.95	0.91	0.92	0.92	0.92	0.92	0.95	0.91	0.89	0.91	0.93	0.88	0.93	0.90	0.88	0.90	0.88	0.89	0.95	0.91	0.92	0.92	0.93	0.90	
Proline	%	1.01	1.13	1.20	1.25	1.19	1.22	1.27	1.17	1.24	1.38	1.38	1.28	1.34	1.07	1.03	1.33	1.02	0.98	1.07	1.08	1.16	1.04	1.08	1.09	
Serine	%	0.92	0.89	0.93	0.96	0.97	0.99	0.89	0.85	0.88	0.89	0.95	0.95	1.01	0.93	0.93	0.96	0.81	0.92	0.99	0.90	0.94	0.98	0.98	0.98	
Threonine	%	1.06	1.01	1.03	1.04	1.01	1.04	1.00	0.96	0.96	0.98	0.98	0.97	1.01	1.02	1.06	1.06	0.89	1.06	1.12	1.03	1.02	1.08	1.10	1.03	
Tyrosine	%	0.78	0.79	0.80	0.80	0.80	0.81	0.82	0.79	0.78	0.79	0.80	0.76	0.80	0.79	0.77	0.78	0.76	0.79	0.83	0.78	0.77	0.81	0.82	0.77	
Valine	%	1.18	1.18	1.20	1.16	1.15	1.16	1.25	1.20	1.17	1.20	1.20	1.09	1.10	1.09	1.06	1.06	1.02	1.07	1.13	1.14	1.11	1.15	1.13	1.08	
Calcium	%	0.0037	0.0039	0.0070	0.0037	0.0049	0.0045	0.0058	0.0039	0.0038	0.0043	0.0042	0.0052	0.0099	0.0044	0.0049	0.0041	0.0037	0.0038	0.0040	0.0050	0.0039	0.0048	0.0042	0.0039	
Cholesterol	mg/100 g	57.6	57.5	61.2	62.4	65.6	62.1	58.7	63.5	47.3	58.9	63.8	53.9	58.7	56.2	65.9	63.8	64.3	60.9	64.6	66.7	64.4	68.2	68.8	64.9	
C08:0 Octanoic (Caprylic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C10:0 Decanoic (Capric)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C11:0 Undecanoic (Hendecanoic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C12:0 Dodecanoic (Lauric)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C14:0 Tetradecanoic (Myristic)	%	0.04	0.08	0.12	0.07	0.06	0.08	0.05	0.06	0.14	0.10	0.05	0.06	0.06	0.04	0.08	0.09	0.08	0.07	0.10	0.11	0.05	0.07	0.06	0.10	
C14:1 Tetradecenoic (Myristoleic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C15:0 Pentadecanoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C15:1 Pentadecenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C16:0 Hexadecanoic (Palmitic)	%	0.74	1.29	1.95	1.31	0.99	1.33	0.76	1.05	2.26	1.69	0.80	0.99	0.88	0.63	1.39	1.44	1.48	1.11	1.82	2.12	0.90	1.15	1.00	1.62	
C16:1 Hexadecenoic (Palmitoleic)	%	0.08	0.12	0.22	0.13	0.14	0.13	0.09	0.11	0.23	0.21	0.08	0.11	0.10	0.09	0.19	0.17	0.15	0.13	0.23	0.26	0.11	0.16	0.10	0.17	
C17:0 Heptadecanoic (Margaric)	%	<0.01	<0.01	0.01	<0.01	<0.01	0.01	<0.01	<0.01	0.02	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	<0.01	0.01	<0.01	<0.01	<0.01	0.01	0.01	
C17:1 Heptadecenoic (Margaroleic)	%	<0.01	<0.01	0.01	<0.01	<0.01	0.01	<0.01	<0.01	0.02	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	<0.01	<0.01	<0.01	0.01	0.01	
C18:0 Octadecanoic (Stearic)	%	0.37	0.63	0.88	0.67	0.44	0.62	0.33	0.56	1.08	0.77	0.39	0.48	0.37	0.29	0.64	0.72	0.78	0.51	0.79	1.06	0.43	0.52	0.51	0.74	
C18:1 Octadecenoic (Oleic)	%	1.16	1.82	2.97	2.00	1.64	1.91	1.22	1.61	3.34	2.72	1.27	1.41	1.34	1.02	2.23	2.35	2.27	1.74	2.92	3.45	1.41	1.85	1.41	2.25	
C18:2 Octadecadienoic (Linoleic)	%	0.10	0.12	0.21	0.15	0.17	0.14	0.09	0.17	0.31	0.27	0.10	0.11	0.10	0.14	0.22	0.28	0.31	0.18	0.34	0.39	0.19	0.26	0.19	0.23	
C18:3 Octadecatrienoic (Linolenic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C18:4 Octadecatetraenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C20:0 Eicosanoic (Arachidic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	<0.01	0.02	0.02	<0.01	<0.01	<0.01	0.01	
C20:1 Eicosenoic (Gadoleic)	%	0.04	0.08	0.10	0.07	0.05	0.07	0.04	0.05	0.12	0.07	0.05	0.05	0.04	0.01	0.05	0.08	0.05	0.04	0.06	0.09	0.04	0.05	0.03	0.07	
C20:2 Eicosadienoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.02	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	<0.01	0.01	0.02	<0.01	0.01	<0.01	0.01	
C20:3 Eicosatrienoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C20:4 Eicosatetraenoic (Arachidonic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C20:5 Eicosapentaenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C21:5 Heneicosapentaenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C22:0 Docosanoic (Behenic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C22:1 Docosenoic (Erucic)	%	0.01	0.02	0.02	0.02	0.01	0.02	0.01																		

SubstDescription	Units	ME2005-003908 / 051	ME2005-003909 / 047	ME2005-003910 / 051	ME2005-003911 / 059	ME2005-003912 / 049	ME2005-003913 / 054	ME2005-003914 / 048	ME2005-004276 / 200437302 5-Jan-05	ME2005-004279 / 200433307 5-Jan-05	ME2005-004280 / 200434306 5-Jan-05	ME2005-004281 / 200433303 5-Jan-05	ME2005-004282 / 200430501 5-Jan-05	ME2005-004283 / 200440603 5-Jan-05	ME2005-004284 / 200435411 5-Jan-05	ME2005-004285 / 200435410 5-Jan-05	ME2005-004286 / 200434008 5-Jan-05	ME2005-004287 / 200435411 5-Jan-05	ME2005-004289 / 200438706 5-Jan-05	ME2005-004290 / 200439208 5-Jan-05	ME2005-004292 / 200435305 5-Jan-05	ME2005-004293 / 200433302 5-Jan-05	ME2005-004294 / 200435301 5-Jan-05	ME2005-004295 / 200434303 5-Jan-05	ME2005-004296 / 200431603 5-Jan-05
Alanine	%	1.32	1.21	1.33	1.28	1.23	1.33	1.34	1.52	1.43	1.41	1.44	1.46	1.33	1.40	1.33	1.43	1.56	1.38	1.34	1.42	1.35	1.41	1.30	1.37
Arginine	%	1.55	1.44	1.60	1.49	1.44	1.56	1.51	1.80	1.66	1.67	1.66	1.74	1.54	1.65	2.31	1.68	1.84	1.63	1.57	1.71	1.64	1.66	1.52	1.59
Aspartic Acid	%	2.24	2.03	2.22	2.17	2.07	2.14	2.24	2.52	2.34	2.40	2.49	2.50	2.41	2.32	2.34	2.27	2.56	2.38	2.23	2.36	2.29	2.42	2.05	2.29
Cystine	%	0.22	0.22	0.23	0.23	0.21	0.22	0.25	0.28	0.23	0.23	0.25	0.26	0.26	0.26	0.27	0.24	0.27	0.23	0.23	0.27	0.25	0.25	0.25	0.28
Glutamic Acid	%	3.51	3.22	3.54	3.39	3.18	3.32	3.48	4.13	3.67	3.89	3.93	4.12	3.81	3.72	3.72	3.72	4.22	3.89	3.61	3.90	3.72	3.93	3.40	3.82
Glycine	%	1.02	0.94	1.06	0.97	1.04	1.25	1.09	1.36	1.33	1.14	1.10	1.16	0.99	1.27	0.97	1.38	1.44	0.98	1.16	1.07	1.03	1.02	1.27	1.04
Histidine	%	1.07	1.00	1.08	1.00	0.97	1.00	1.02	1.11	0.95	0.99	1.04	1.02	0.99	1.29	0.94	1.07	0.97	0.86	1.01	1.02	1.01	0.86	0.96	0.96
Isoleucine	%	1.02	0.96	1.03	1.01	0.96	0.99	0.99	1.18	0.99	1.05	1.09	1.12	1.08	1.05	1.09	0.98	1.16	1.05	0.96	1.06	1.03	1.10	0.95	1.08
Leucine	%	1.83	1.69	1.82	1.78	1.67	1.73	1.80	2.09	1.85	1.95	2.01	2.06	1.95	1.89	1.93	1.83	2.10	1.94	1.77	1.95	1.87	1.97	1.67	1.88
Lysine, Total	%	2.02	1.88	2.04	1.95	1.87	1.87	1.99	2.32	2.00	2.12	2.21	2.29	2.17	2.12	2.69	1.98	2.36	2.11	1.95	2.15	2.05	2.14	1.81	2.09
Methionine	%	0.55	0.54	0.58	0.61	0.54	0.55	0.63	0.67	0.56	0.57	0.61	0.69	0.65	0.65	0.71	0.59	0.66	0.57	0.59	0.68	0.65	0.65	0.62	0.71
Phenylalanine	%	0.87	0.82	0.88	0.85	0.81	0.83	0.85	1.04	0.87	0.91	0.98	0.94	0.93	0.88	1.06	0.91	1.06	0.87	0.83	0.95	0.92	0.93	0.80	0.90
Proline	%	0.93	0.85	1.04	0.87	0.93	1.07	1.02	1.18	1.03	1.04	1.00	1.09	1.07	1.23	1.03	1.15	1.16	1.18	1.25	1.25	1.19	1.18	1.24	1.20
Serine	%	0.93	0.86	0.92	0.91	0.86	0.85	0.94	1.04	0.99	1.00	1.05	1.05	0.98	0.97	0.95	0.96	1.08	0.98	0.95	0.98	0.95	1.00	0.86	0.93
Threonine	%	1.05	0.92	1.04	1.01	0.95	0.98	1.04	1.21	1.09	1.13	1.15	1.14	1.15	1.07	1.08	1.04	1.22	1.13	1.03	1.09	1.07	1.12	0.92	1.07
Tyrosine	%	0.76	0.71	0.77	0.75	0.70	0.71	0.75	0.90	0.74	0.80	0.84	0.83	0.82	0.75	0.88	0.79	0.91	0.79	0.73	0.85	0.82	0.84	0.69	0.82
Valine	%	1.09	1.01	1.08	1.07	1.03	1.05	1.06	1.27	1.08	1.10	1.14	1.18	1.14	1.12	1.13	1.07	1.26	1.08	1.01	1.09	1.06	1.16	1.04	1.13
Calcium	%	0.0041	0.0042	0.0040	0.0039	0.0050	0.0040	0.0043	0.0045	0.0056	0.0048	0.0048	0.0065	0.0048	0.0046	0.0061	0.0093	0.0054	0.0081	0.0053	0.0068	0.0055	0.0064	0.0047	0.0077
Cholesterol	mg/100 g	59.3	59.0	60.7	60.7	63.5	63.9	61.3	54.5	55.8	58.6	62.6	60.0	69.0	60.9	61.7	62.4	62.4	68.0	60.9	59.4	53.5	53.0	61.2	61.3
C08:0 Octanoic (Caprylic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C10:0 Decanoic (Capric)	%	0.01	0.02	<0.01	0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C11:0 Undecanoic (Hendecanoic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C12:0 Dodecanoic (Lauroic)	%	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C14:0 Tetradecanoic (Myristic)	%	0.17	0.19	0.12	0.12	0.18	0.12	0.07	0.05	0.09	0.07	0.06	0.08	0.09	0.06	0.08	0.13	0.10	0.07	0.09	0.08	0.07	0.05	0.07	0.07
C14:1 Tetradecenoic (Myristoleic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C15:0 Pentadecanoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C15:1 Pentadecenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C16:0 Hexadecanoic (Palmitic)	%	3.17	3.21	2.37	2.26	3.35	2.21	1.24	0.98	1.55	1.31	0.96	1.45	1.49	1.07	1.53	2.26	1.97	1.21	1.47	1.24	1.22	0.75	1.34	1.12
C16:1 Hexadecenoic (Palmitoleic)	%	0.22	0.33	0.20	0.24	0.31	0.24	0.12	0.12	0.19	0.15	0.15	0.17	0.21	0.13	0.15	0.24	0.24	0.15	0.16	0.17	0.14	0.11	0.18	0.14
C17:0 Heptadecanoic (Marganic)	%	0.04	0.02	0.03	0.02	0.03	0.02	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C17:1 Heptadecenoic (Margoleic)	%	0.02	0.02	0.02	0.02	0.02	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C18:0 Octadecanoic (Stearic)	%	1.94	1.79	1.45	1.22	1.91	1.20	0.69	0.48	0.72	0.68	0.40	0.77	0.66	0.57	0.83	1.26	1.02	0.58	0.70	0.54	0.59	0.34	0.66	0.54
C18:1 Octadecenoic (Oleic)	%	3.41	4.52	3.10	3.43	4.80	3.48	1.90	1.77	2.90	2.28	1.66	2.44	2.29	1.86	2.48	3.44	3.23	1.95	2.24	2.18	2.08	1.36	2.35	1.89
C18:2 Octadecadienoic (Linoleic)	%	0.23	0.38	0.34	0.33	0.57	0.37	0.24	0.24	0.37	0.32	0.21	0.35	0.28	0.25	0.38	0.44	0.46	0.25	0.35	0.33	0.41	0.20	0.34	0.26
C18:3 Octadecatrienoic (Linolenic)	%	0.02	0.02	0.01	0.01	0.02	0.01	<0.01	<0.01	0.01	0.01	<0.01	0.01	<0.01	<0.01	0.01	0.02	0.02	0.01	0.01	0.01	0.01	<0.01	0.01	<0.01
C18:4 Octadecatetraenoic	%	0.02	0.02	0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:0 Eicosanoic (Arachidic)	%	0.04	0.03	0.02	0.02	0.03	0.02	0.01	<0.01	0.01	0.01	<0.01	0.01	<0.01	<0.01	0.01	0.01	<0.01	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01
C20:1 Eicosenoic (Gadoleic)	%	0.14	0.14	0.11	0.10	0.14	0.09	0.05	0.05	0.06	0.08	0.05	0.07	0.07	0.05	0.08	0.09	0.09	0.07	0.08	0.07	0.09	0.05	0.07	0.07
C20:2 Eicosadienoic	%	0.02	0.02	0.02	0.02	0.03	0.02	0.01	0.01	0.02	0.02	<0.01	0.02	0.01	0.01	0.02	0.02	0.02	0.01	0.02	0.01	0.02	<0.01	0.02	0.01
C20:3 Eicosatrienoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:4 Eicosatetraenoic (Arachidonic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:5 Eicosapentaenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C21:5 Heneicosapentaenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C22:0 Docosanoic (Behenic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C22:1 Docosenoic (Erucic)	%	0.02	0.02	0.02	0.03	0.01	<0.01	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	<0.01	0.01	0.01	0.01	0.04	<0.01	0.01	0.01
C22:2 Docosadienoic																									

SubstDescription	Units	ME2005-004451 / 20043406 18-Jan-05	ME2005-004450 / 200440907 18-Jan-05	ME2005-004449 / 200440909 18-Jan-05	ME2005-004448 / 200438707 18-Jan-05	ME2005-004447 / 200434511 12-Jan-05	ME2005-004446 / 200435803 12-Jan-05	ME2005-004445 / 200437406 12-Jan-05	ME2005-004444 / 200438104 12-Jan-05	ME2005-004443 / 200431601 12-Jan-05	ME2005-004442 / 200436602 12-Jan-05	ME2005-004441 / 200436606 12-Jan-05	ME2005-004440 / 200436005 12-Jan-05	ME2005-004439 / 200431010 12-Jan-05	ME2005-004438 / 200431605 12-Jan-05	ME2005-004437 / 200431010 12-Jan-05	ME2005-004436 / 200431605 12-Jan-05	ME2005-004435 / 200432903 12-Jan-05	ME2005-004434 / 200436603 12-Jan-05	ME2005-004433 / 200431004 12-Jan-05	ME2005-004432 / 200436912 12-Jan-05	ME2005-004431 / 200434502 12-Jan-05	ME2005-004430 / 200431702 12-Jan-05	ME2005-004429 / 200436913 12-Jan-05	ME2005-004428 / 200434304 12-Jan-05	
Alanine	%	1.37	1.40	1.37	1.42	1.38	1.43	1.38	1.50	1.54	1.55	1.53	1.52	1.54	1.47	1.68	1.50	1.49	1.41	1.49	1.49	1.43	1.53	1.43	1.43	
Arginine	%	1.57	1.63	1.62	1.70	1.65	1.70	1.54	1.53	1.58	1.64	1.60	1.59	1.62	1.51	1.68	1.66	1.63	1.55	1.63	1.62	1.53	1.66	1.55	1.58	
Aspartic Acid	%	2.30	2.32	2.37	2.45	2.34	2.44	2.25	2.48	2.53	2.61	2.43	2.55	2.63	2.53	2.54	2.55	2.48	2.36	2.43	2.40	2.30	2.60	2.42	2.42	
Cystine	%	0.24	0.25	0.23	0.24	0.22	0.24	0.24	0.25	0.25	0.24	0.24	0.24	0.25	0.23	0.23	0.23	0.27	0.24	0.27	0.26	0.23	0.23	0.26	0.22	0.25
Glutamic Acid	%	3.68	3.75	3.84	3.94	3.79	3.93	3.52	3.80	3.89	4.13	3.81	3.97	4.13	3.96	3.98	4.05	3.96	3.72	3.91	3.86	3.65	4.18	3.86	3.81	
Glycine	%	1.18	1.26	1.09	1.16	1.14	1.18	1.21	1.06	1.14	1.14	1.36	1.16	1.12	1.02	1.68	1.18	1.12	1.09	1.21	1.28	1.19	1.08	1.01	1.05	
Histidine	%	0.99	0.91	1.04	1.07	1.00	1.04	1.02	0.88	0.92	0.94	0.93	0.99	1.04	1.00	1.00	1.03	1.00	0.96	1.01	0.91	0.94	0.97	0.99	1.03	
Isoleucine	%	0.86	0.95	1.03	1.11	1.08	1.10	1.12	0.81	0.90	0.97	0.89	0.96	0.92	0.81	0.86	0.98	1.03	1.00	1.03	1.00	0.91	0.95	0.97	1.03	
Leucine	%	1.83	1.88	1.94	2.02	1.92	2.00	1.88	1.82	1.89	2.01	1.83	1.96	1.99	1.90	1.88	1.98	1.98	1.89	1.93	1.90	1.82	2.03	1.93	1.95	
Lysine, Total	%	2.06	2.11	2.19	2.27	2.17	2.27	2.08	2.00	2.07	2.18	1.96	2.08	2.15	2.05	2.03	2.15	2.17	2.06	2.09	2.07	1.97	2.20	2.08	2.10	
Methionine	%	0.59	0.60	0.58	0.56	0.56	0.59	0.58	0.58	0.60	0.58	0.55	0.56	0.58	0.51	0.53	0.71	0.65	0.68	0.64	0.65	0.60	0.64	0.62	0.64	
Phenylalanine	%	0.89	0.92	0.95	0.98	0.95	0.98	0.96	1.03	1.07	1.11	1.03	1.09	1.10	1.03	1.07	1.05	1.08	1.03	1.05	1.04	0.98	1.08	1.05	1.05	
Proline	%	0.96	0.98	0.93	0.96	1.04	0.97	1.07	0.97	1.06	1.25	1.23	1.16	1.19	1.19	1.54	1.18	1.17	1.19	1.30	1.31	1.27	1.24	1.04	1.12	
Serine	%	0.97	0.98	0.99	1.00	0.95	0.99	0.87	1.02	1.04	1.04	0.98	1.02	1.04	1.03	1.06	1.06	1.03	0.97	1.01	1.00	0.97	1.08	1.00	0.98	
Threonine	%	1.11	1.12	1.14	1.19	1.13	1.20	1.05	1.08	1.11	1.19	1.08	1.14	1.18	1.11	1.10	1.15	1.15	1.09	1.12	1.10	1.06	1.22	1.14	1.23	
Tyrosine	%	0.79	0.81	0.83	0.86	0.83	0.86	0.76	0.82	0.84	0.91	0.81	0.87	0.89	0.86	0.83	0.87	0.87	0.83	0.85	0.83	0.80	0.91	0.86	0.85	
Valine	%	0.94	1.03	1.10	1.17	1.14	1.18	1.22	0.89	0.96	1.02	0.97	1.02	0.99	0.86	0.97	1.06	1.10	1.05	1.07	0.98	1.01	1.04	1.09	1.09	
Calcium	%	0.0051	0.0047	0.0046	0.0044	0.0055	0.0043	0.0048	0.0071	0.0050	0.0060	0.0051	0.0055	0.0039	0.0046	0.0051	0.0050	0.0061	0.0053	0.0045	0.0070	0.0047	0.0069	0.0054	0.0058	
Cholesterol	mg/100 g	48.6	57.9	54.8	53.4	58.2	61.6	55.9	55.9	54.7	56.7	57.4	58.4	51.4	60.9	57.4	54.2	54.6	57.6	53.8	58.7	56.7	59.2	58.3	56.6	
C08:0 Octanoic (Caprylic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C10:0 Decanoic (Capric)	%	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C11:0 Undecanoic (Hendecanoic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C12:0 Dodecanoic (Lauric)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C14:0 Tetradecanoic (Myristic)	%	0.13	0.12	0.07	0.08	0.13	0.09	0.11	0.11	0.08	0.07	0.18	0.08	0.06	0.08	0.14	<0.01	0.10	0.13	0.13	0.09	0.08	0.05	0.06	0.07	
C14:1 Tetradecenoic (Myristoleic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C15:0 Pentadecanoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C15:1 Pentadecenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C16:0 Hexadecanoic (Palmitic)	%	2.56	1.77	1.22	1.31	2.04	1.60	2.11	1.84	1.36	1.06	2.96	1.33	0.95	1.41	2.52	0.86	1.68	2.35	2.20	1.44	1.49	0.80	1.01	1.26	
C16:1 Hexadecenoic (Palmitoleic)	%	0.28	0.21	0.15	0.14	0.22	0.19	0.24	0.17	0.17	0.12	0.32	0.16	0.11	0.12	0.27	0.09	0.16	0.32	0.29	0.19	0.19	0.12	0.13	0.15	
C17:0 Heptadecanoic (Margaric)	%	0.01	0.01	<0.01	<0.01	<0.01	0.01	0.02	<0.01	<0.01	<0.01	0.01	<0.01	<0.01	0.01	0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C17:1 Heptadecenoic (Margaroleic)	%	0.01	0.01	<0.01	<0.01	0.01	0.01	0.02	<0.01	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C18:0 Octadecanoic (Stearic)	%	1.28	0.76	0.60	0.65	0.98	0.71	1.11	0.93	0.62	0.47	1.38	0.60	0.45	0.83	1.27	0.42	0.85	1.05	1.06	0.61	0.72	0.36	0.48	0.57	
C18:1 Octadecenoic (Oleic)	%	3.92	2.68	1.97	2.04	3.08	2.63	3.41	2.95	2.12	1.61	4.42	2.11	1.53	2.10	3.91	1.32	2.51	3.75	3.65	2.35	2.39	1.31	1.55	2.09	
C18:2 Octadecadienoic (Linoleic)	%	0.38	0.30	0.18	0.23	0.34	0.29	0.37	0.35	0.25	0.18	0.50	0.27	0.25	0.31	0.40	0.17	0.24	0.44	0.45	0.24	0.29	0.12	0.15	0.23	
C18:3 Octadecatrienoic (Linolenic)	%	0.01	<0.01	<0.01	<0.01	0.01	<0.01	0.01	0.01	<0.01	<0.01	0.02	<0.01	<0.01	0.01	0.01	<0.01	<0.01	0.02	0.02	<0.01	0.01	<0.01	<0.01	<0.01	
C18:4 Octadecatetraenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C20:0 Eicosanoic (Arachidic)	%	0.02	0.02	0.02	0.01	0.01	0.02	0.02	0.02	0.02	0.01	0.03	0.02	<0.01	0.01	0.02	<0.01	0.02	0.02	0.01	<0.01	0.01	<0.01	<0.01	0.01	
C20:1 Eicosanoic (Gadoleic)	%	0.15	0.14	0.09	0.09	0.15	0.19	0.20	0.19	0.11	0.10	0.20	0.11	0.07	0.10	0.20	0.07	0.17	0.11	0.11	0.09	0.08	0.05	0.05	0.10	
C20:2 Eicosadienoic	%	0.02	0.01	<0.01	0.01	0.02	0.01	0.02	0.02	<0.01	<0.01	0.02	0.01	0.01	0.01	0.02	0.01	0.02	0.02	0.02	0.01	0.01	<0.01	<0.01	<0.01	
C20:3 Eicosatrienoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C20:4 Eicosatetraenoic (Arachidonic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C20:5 Eicosapentaenoic	%	0.01	<0.01	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C21:5 Heneicosapentaenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C22:0 Docosanoic (Behenic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	
C22:1 Docosenoic (Erucic)	%	0.02	0.01	<0.01	<0.01	0.01	0.02	0.03	0.02	0.02																

SubstDescription	Units	ME2005-005560 / 200438703 2-Feb-05	ME2005-005559 / 200434107 20-Jan-05	ME2005-005558 / 200431005 20-Jan-05	ME2005-005557 / 20043104 20-Jan-05	ME2005-005556 / 200430710 20-Jan-05	ME2005-005555 / 200434109 20-Jan-05	ME2005-005554 / 200436402 20-Jan-05	ME2005-005553 / 200431602 20-Jan-05	ME2005-005552 / 200430702 20-Jan-05	ME2005-005551 / 200438611 20-Jan-05	ME2005-005550 / 200433802 20-Jan-05	ME2005-005549 / 200433901 20-Jan-05	ME2005-005548 / 200438903 20-Jan-05	ME2005-005547 / 200438604 20-Jan-05	ME2005-005546 / 200436602 20-Jan-05	ME2005-005545 / 200438601 20-Jan-05	ME2005-005544 / 200438901 20-Jan-05	ME2005-005543 / 200433801 20-Jan-05	ME2005-005542 / 200438608 20-Jan-05	ME2005-005541 / 200439902 20-Jan-05	ME2005-005540 / 200433208 20-Jan-05	ME2005-005539 / 200435307 20-Jan-05	ME2005-005538 / 200439901 20-Jan-05	ME2005-005537 / 200435304 20-Jan-05
Alanine	%	1.28	1.31	1.43	1.43	1.49	1.43	1.55	1.50	1.47	1.49	1.53	1.54	1.45	1.27	1.27	1.32	1.35	1.38	1.37	1.38	1.27	1.30	1.30	1.33
Arginine	%	1.52	1.37	1.45	1.44	1.44	1.43	1.52	1.48	1.44	1.44	1.50	1.47	1.42	1.50	1.48	1.53	1.60	1.66	1.61	1.66	1.48	1.47	1.52	1.56
Aspartic Acid	%	2.09	1.62	2.11	2.06	1.94	2.03	2.20	2.24	2.15	2.19	2.27	2.19	2.16	2.22	2.10	2.30	2.32	2.44	2.30	2.34	2.18	2.27	2.29	2.24
Cystine	%	0.26	0.24	0.27	0.25	0.27	0.26	0.27	0.28	0.27	0.28	0.26	0.27	0.26	0.27	0.24	0.25	0.27	0.28	0.24	0.25	0.27	0.26	0.27	0.25
Glutamic Acid	%	3.51	3.94	4.56	4.44	4.22	4.45	4.77	4.84	4.62	4.70	4.89	4.68	4.69	3.63	3.38	3.77	3.84	4.10	3.74	3.90	3.58	3.69	3.81	3.70
Glycine	%	0.96	1.21	1.21	1.32	1.84	1.42	1.57	1.25	1.31	1.29	1.25	1.51	1.20	0.97	1.18	1.01	1.09	1.02	1.19	1.16	1.07	1.01	1.04	1.11
Histidine	%	0.97	0.82	0.92	0.96	0.83	0.84	0.88	0.95	0.92	0.91	0.93	0.87	0.91	0.91	0.86	0.94	0.96	0.92	0.96	0.94	0.89	0.95	0.91	0.91
Isoleucine	%	1.01	0.95	1.04	1.02	0.93	1.01	0.91	0.92	0.89	0.91	0.91	0.88	0.88	0.83	0.78	0.87	0.88	0.92	0.85	0.86	0.73	0.77	0.81	0.81
Leucine	%	1.80	1.70	1.87	1.82	1.66	1.79	1.86	1.91	1.84	1.87	1.93	1.85	1.85	1.80	1.67	1.85	1.87	1.97	1.84	1.87	1.72	1.80	1.83	1.80
Lysine, Total	%	1.99	1.73	1.90	1.86	1.69	1.81	1.90	1.94	1.86	1.91	1.96	1.88	1.89	1.95	1.80	2.03	2.02	2.16	1.98	2.05	1.88	1.95	1.99	1.96
Methionine	%	0.65	0.58	0.67	0.64	0.68	0.63	0.67	0.69	0.67	0.68	0.60	0.64	0.64	0.69	0.57	0.62	0.64	0.66	0.55	0.56	0.64	0.64	0.64	0.56
Phenylalanine	%	0.91	0.82	0.89	0.87	0.81	0.85	0.90	0.91	0.86	0.89	0.92	0.88	0.87	0.87	0.82	0.89	0.90	0.94	0.96	0.97	0.86	0.88	0.91	0.89
Proline	%	1.10	0.90	0.90	0.96	1.18	1.00	1.09	0.96	0.99	1.01	1.02	1.08	0.97	0.87	0.96	0.90	0.97	0.95	1.06	0.99	0.97	1.00	1.06	1.07
Serine	%	0.89	0.87	1.10	1.07	1.01	1.04	1.20	1.20	1.16	1.19	1.21	1.18	1.16	0.94	0.91	0.98	0.99	1.04	0.97	0.99	0.95	0.97	0.97	0.96
Threonine	%	1.02	0.95	1.19	1.16	1.07	1.14	1.23	1.26	1.20	1.24	1.24	1.25	1.04	0.97	1.08	1.09	1.15	1.06	1.10	0.98	1.05	1.05	1.04	1.04
Tyrosine	%	0.81	0.72	0.78	0.75	0.68	0.73	0.78	0.81	0.77	0.80	0.84	0.78	0.79	0.78	0.72	0.80	0.81	0.85	0.82	0.83	0.76	0.79	0.80	0.79
Valine	%	1.09	1.02	1.13	1.12	1.06	1.11	0.99	1.00	0.97	0.99	1.03	0.98	0.96	0.88	0.84	0.92	0.93	0.97	0.93	0.94	0.77	0.83	0.87	0.86
Calcium	%	0.0049	0.0045	0.0042	0.0043	0.0060	0.0048	0.0051	0.0046	0.0067	0.010	0.0046	0.0047	0.0046	0.0050	0.0044	0.0057	0.0047	0.0044	0.0049	0.0069	0.0048	0.0083	0.0049	0.0061
Cholesterol	mg/100 g	58.2	61.0	55.7	64.2	62.1	65.1	59.7	61.0	66.9	70.5	63.0	64.0	62.4	60.2	68.2	59.9	62.8	65.6	64.8	60.2	64.7	67.8	65.9	68.7
C08:0 Octanoic (Caprylic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C10:0 Decanoic (Capric)	%	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01
C11:0 Undecanoic (Hendecanoic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C12:0 Dodecanoic (Lauric)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C14:0 Tetradecanoic (Myristic)	%	0.12	0.13	0.08	0.11	0.06	0.09	0.06	0.06	0.08	0.08	0.09	0.06	0.06	0.07	0.14	0.04	0.12	0.04	0.09	0.10	0.04	0.08	0.11	0.16
C14:1 Tetradecenoic (Myristoleic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C15:0 Pentadecanoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C15:1 Pentadecenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C16:0 Hexadecanoic (Palmitic)	%	2.05	2.13	1.28	1.82	1.12	1.62	1.04	0.89	1.45	1.23	1.51	0.79	0.97	1.12	2.59	0.62	1.99	0.65	1.54	1.64	0.67	1.31	1.89	2.66
C16:1 Hexadecenoic (Palmitoleic)	%	0.19	0.23	0.17	0.22	0.13	0.25	0.12	0.13	0.17	0.16	0.17	0.11	0.12	0.15	0.27	0.08	0.17	0.08	0.20	0.18	0.08	0.18	0.17	0.31
C17:0 Heptadecanoic (Margaric)	%	0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C17:1 Heptadecenoic (Margaroleic)	%	0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C18:0 Octadecanoic (Stearic)	%	1.01	1.08	0.56	0.79	0.60	0.77	0.56	0.40	0.73	0.55	0.68	0.34	0.46	0.53	1.25	0.29	1.02	0.33	0.69	0.80	0.34	0.55	0.95	1.33
C18:1 Octadecenoic (Oleic)	%	2.83	3.49	2.22	2.84	1.87	2.80	1.67	1.55	2.42	2.13	2.47	1.45	1.61	1.85	3.91	1.03	2.64	1.27	2.49	2.74	1.13	2.39	2.72	3.94
C18:2 Octadecadienoic (Linoleic)	%	0.21	0.44	0.31	0.29	0.20	0.27	0.25	0.24	0.28	0.23	0.34	0.20	0.19	0.26	0.32	0.16	0.25	0.21	0.31	0.38	0.16	0.37	0.28	0.45
C18:3 Octadecatrienoic (Linolenic)	%	<0.01	0.02	0.01	<0.01	<0.01	0.01	0.01	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01	<0.01	<0.01	<0.01	0.01	0.02	<0.01	0.01	<0.01	0.01
C18:4 Octadecatetraenoic	%	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:0 Eicosanoic (Arachidic)	%	0.03	0.01	0.02	0.02	<0.01	<0.01	0.01	<0.01	0.01	0.01	0.02	<0.01	0.01	<0.01	0.02	<0.01	0.03	<0.01	0.01	<0.01	0.01	0.02	0.02	0.02
C20:1 Eicosenoic (Gadoleic)	%	0.23	0.11	0.08	0.15	0.07	0.06	0.08	0.06	0.09	0.10	0.13	0.06	0.10	0.06	0.22	0.04	0.22	0.03	0.09	0.09	0.03	0.10	0.19	0.19
C20:2 Eicosadienoic	%	0.01	0.02	0.01	0.01	<0.01	0.01	0.01	<0.01	0.01	<0.01	0.02	<0.01	<0.01	0.01	0.02	<0.01	0.01	0.01	0.01	0.02	<0.01	0.02	0.01	0.02
C20:3 Eicosatrienoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:4 Eicosatetraenoic (Arachidonic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:5 Eicosapentaenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C21:5 Heneicosapentaenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C22:0 Docosanoic (Behenic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C22:1 Docosenoic (Erucic)	%	0.02	0.01	0.01	0.02	<0.01	<0.01	0.01	0.01	0.01	0.01														

SubstestDescription	Units	ME2005-005561 / 200439401 2-Feb-05	ME2005-005562 / 200431804 2-Feb-05	ME2005-005563 / 200439404 2-Feb-05	ME2005-005564 / 200438403 2-Feb-05	ME2005-005565 / 200439303 2-Feb-05	ME2005-005566 / 200434102 2-Feb-05	ME2005-005567 / 200430803 2-Feb-05
Alanine	%	1.41	1.33	1.43	1.47	1.39	1.49	1.38
Arginine	%	1.69	1.55	1.61	1.66	1.66	1.71	1.54
Aspartic Acid	%	2.39	2.30	2.43	2.54	2.50	2.36	2.35
Cystine	%	0.26	0.24	0.27	0.28	0.27	0.28	0.27
Glutamic Acid	%	3.97	3.80	3.88	4.06	3.82	3.71	3.71
Glycine	%	1.22	1.08	1.12	1.09	1.12	1.58	1.05
Histidine	%	1.02	0.95	0.99	1.02	1.16	1.04	1.01
Isoleucine	%	0.87	0.82	1.05	1.13	1.14	1.07	1.13
Leucine	%	1.92	1.84	1.94	2.04	2.04	1.91	1.88
Lysine, Total	%	2.10	2.05	2.14	2.24	2.18	2.06	2.09
Methionine	%	0.60	0.56	0.63	0.65	0.58	0.74	0.65
Phenylalanine	%	0.98	0.95	1.04	1.07	1.01	0.98	1.02
Proline	%	1.19	1.10	1.15	1.22	0.97	1.19	1.14
Serine	%	1.02	0.99	1.00	1.01	1.05	0.99	0.85
Threonine	%	1.11	1.08	1.10	1.16	1.08	1.02	1.04
Tyrosine	%	0.85	0.82	0.84	0.88	0.89	0.81	0.80
Valine	%	0.92	0.90	1.12	1.20	1.24	1.19	1.20
Calcium	%	0.0092	0.0064	0.0049	0.0049	0.0045	0.0048	0.0044
Cholesterol	mg/100 g	68.9	66.3	65.8	64.8	61.3	63.9	63.9
C08:0 Octanoic (Caprylic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C10:0 Decanoic (Capric)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C11:0 Undecanoic (Hendecanoic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C12:0 Dodecanoic (Lauric)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C14:0 Tetradecanoic (Myristic)	%	0.05	0.12	0.08	0.03	0.05	0.09	0.09
C14:1 Tetradecenoic (Myristoleic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C15:0 Pentadecanoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C15:1 Pentadecenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C16:0 Hexadecanoic (Palmitic)	%	0.86	1.96	1.31	0.56	0.78	1.52	1.47
C16:1 Hexadecenoic (Palmitoleic)	%	0.10	0.19	0.15	0.08	0.11	0.20	0.19
C17:0 Heptadecanoic (Margaric)	%	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C17:1 Heptadecenoic Margaroleic	%	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C18:0 Octadecanoic (Stearic)	%	0.41	1.02	0.62	0.27	0.36	0.67	0.67
C18:1 Octadecenoic (Oleic)	%	1.39	2.82	1.97	1.03	1.37	2.63	2.47
C18:2 Octadecadienoic (Linoleic)	%	0.24	0.37	0.28	0.14	0.22	0.32	0.34
C18:3 Octadecatrienoic (Linolenic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.01
C18:4 Octadecatetraenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:0 Eicosanoic (Arachidic)	%	<0.01	0.02	0.01	<0.01	<0.01	0.01	0.01
C20:1 Eicosenoic (Gadoleic)	%	0.06	0.18	0.09	0.03	0.04	0.11	0.10
C20:2 Eicosadienoic	%	0.01	0.02	0.01	<0.01	0.01	0.01	0.02
C20:3 Eicosatrienoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:4 Eicosatetraenoic (Arachidonic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C20:5 Eicosapentaenoic	%	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C21:5 Heneicosapentaenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C22:0 Docosanoic (Behenic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C22:1 Docosenoic (Erucic)	%	<0.01	0.03	0.01	<0.01	<0.01	0.01	0.02
C22:2 Docosadienoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C22:3 Docosatrienoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C22:4 Docosatetraenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C22:5 Docosapentaenoic	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C22:6 Docosahexanoic	%	<0.01	0.04	0.02	<0.01	<0.01	0.02	0.02
C24:0 Tetracosanoic (Lignoceric)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
C24:1 Tetracosenoic (Nervonic)	%	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
Iron	%	0.00072	0.00059	0.00066	0.00062	0.00061	0.00067	0.00055
Niacin	mg/100 g	9.76	12.0	11.7	11.5	9.48	11.5	11.0
Phosphorus	%	0.21	0.22	0.22	0.22	0.22	0.22	0.22
Vitamin B12	mcg/100 g	1.04	1.11	0.821	0.913	0.419	.987	1.03
Vitamin B6	mg/100 g	0.370	0.344	0.290	0.301	0.429	0.424	0.413
Zinc	%	0.0015	0.0016	0.0015	0.0015	0.0015	0.0013	0.0014

Animal ID	Estradiol (pg/mL)	IGF-1 (ng/mL)
200430503	15.10	234.30
200430504	10.48	249.03
200430507	11.87	236.61
200430508	10.50	220.57
200430510	9.49	181.81
200430602	10.29	423.34
200430603	9.70	256.34
200430604	13.14	286.52
200430605	13.85	215.09
200430606	11.03	261.56
200430607	17.73	234.76
200430609	21.21	323.89
200430611	6.84	131.17
200430702	8.38	401.41
200430703	12.89	456.17
200430704	23.75	279.30
200430705	18.16	300.40
200430706	12.66	120.47
200430707	13.73	243.51
200430708	19.48	336.94
200430709	9.81	381.04
200430710	11.29	327.05
200430711	11.68	170.62
200430801	8.87	119.52
200430802	7.90	315.10
200430803	7.82	284.30
200430804	15.60	202.10
200430805	14.39	255.20
200430902	9.43	128.25
200430903	8.90	142.42
200430905	14.54	61.60
200430906	17.20	75.47
200431002	7.78	325.82
200431003	10.93	326.45
200431004	9.16	397.17
200431005	6.68	305.27
200431006	17.99	211.34
200431008	14.09	213.63
200431009	17.62	226.24
200431010	10.13	231.94
200431011	6.86	130.86
200431401	10.77	388.79
200431403	10.08	152.27
200431404	15.23	136.04
200431405	16.19	244.83
200431406	16.54	318.47
200431407	17.04	256.94
200431408	17.44	275.05
200431411	11.78	196.85
200431501	19.62	104.00
200431502	21.65	132.91
200431503	12.07	210.96
200431504	15.24	66.33
200431505	23.65	109.25
200431601	8.67	455.49
200431602	6.75	440.83

Animal ID	Estradiol (pg/mL)	IGF-1 (ng/mL)
200431604	10.44	366.43
200431605	6.81	347.54
200431606	15.64	227.88
200431607	6.63	324.73
200431608	29.27	99.00
200431610	10.57	346.65
200431702	8.90	356.96
200431703	3.28	311.53
200431704	11.67	170.25
200431706	7.45	327.22
200431708	7.61	244.62
200431801	11.99	315.19
200431802	18.97	344.92
200431803	9.03	339.05
200431804	6.17	12.12
200431805	4.60	265.45
200432002	8.32	276.80
200432003	10.58	265.68
200432004	9.21	394.13
200432006	10.96	251.21
200432008	18.16	175.18
200432101	10.43	482.62
200432103	13.33	440.47
200432105	11.10	171.34
200432108	11.97	346.03
200432302	9.91	417.78
200432304	14.01	349.05
200432306	5.20	284.69
200432308	9.43	364.04
200432310	6.96	306.87
200432402	11.15	407.32
200432403	11.99	452.08
200432404	4.98	417.86
200432405	10.53	269.47
200432406	6.43	309.52
200432501	12.45	347.05
200432503	13.83	451.86
200432506	6.93	292.88
200432507	14.35	348.97
200432601	9.07	216.61
200432602	13.62	251.19
200432604	12.88	267.96
200432607	13.40	141.39
200432609	19.59	139.04
200432610	6.43	150.00
200432702	7.43	310.70
200432704	9.38	102.12
200432705	8.60	103.61
200432706	9.18	218.59
200432901	8.97	158.22
200432907	9.27	102.10
200432909	9.34	116.61
200432911	16.59	207.26
200432913	14.63	126.01
200433001	10.61	303.70
200433007	10.34	375.13

Animal ID	Estradiol (pg/mL)	IGF-1 (ng/mL)
200433010	11.76	311.37
200433011	7.56	335.40
200433201	13.05	346.05
200433204	9.54	171.85
200433205	6.53	291.92
200433206	9.67	115.72
200433207	8.22	134.85
200433208	10.16	213.54
200433301	5.58	202.51
200433302	6.93	215.36
200433303	9.78	239.21
200433306	14.67	144.77
200433307	8.58	190.21
200433401	6.13	537.12
200433404	7.38	457.47
200433405	10.17	431.29
200433406	12.55	342.35
200433408	8.84	312.89
200433409	17.78	185.61
200433505	13.34	367.82
200433509	14.93	329.70
200433510	16.86	358.39
200433511	11.27	271.46
200433512	13.95	277.41
200433601	7.06	160.77
200433602	12.99	181.41
200433603	8.70	420.01
200433604	11.33	427.39
200433605	14.18	221.22
200433708	3.57	49.89
200433709	8.34	229.19
200433710	8.08	251.76
200433711	10.67	265.23
200433801	7.61	214.18
200433802	8.62	431.25
200433803	12.83	286.91
200433804	7.96	343.11
200433807	14.94	172.12
200433808	10.05	166.98
200433809	14.99	200.28
200434001	14.66	190.34
200434003	7.51	181.35
200434004	9.13	220.89
200434005	9.12	123.89
200434006	12.25	115.39
200434007	11.12	36.73
200434101	10.25	283.97
200434102	9.16	240.47
200434104	8.86	265.40
200434105	4.38	267.79
200434107	7.03	181.22
200434109	8.98	178.95
200434111	7.73	113.32
200434113	6.15	162.30
200434201	14.38	303.94
200434202	9.22	213.24

Animal ID	Estradiol (pg/mL)	IGF-1 (ng/mL)
200434203	10.40	111.37
200434206	22.00	303.95
200434301	8.56	400.00
200434304	8.72	351.16
200434310	7.40	431.49
200434401	11.24	454.82
200434402	9.93	312.04
200434403	10.42	303.87
200434502	9.28	393.08
200434504	11.41	311.43
200434507	5.72	287.78
200434509	15.60	493.41
200434511	7.03	345.13
200434512	9.59	398.78
200434601	11.04	327.52
200434602	9.88	507.74
200434603	6.75	506.35
200434604	7.34	270.51
200434605	3.81	332.80
200434606	17.64	468.80
200434607	15.79	479.65
200434802	9.43	317.27
200434803	12.94	362.02
200434804	15.17	283.12
200434805	9.42	346.44
200434806	12.71	101.60
200434807	17.11	74.27
200434808	10.74	160.83
200434809	45.08	123.88
200434901	15.14	302.59
200434902	16.16	341.39
200434903	12.25	317.83
200434904	14.87	404.95
200434905	11.79	190.47
200435302	19.64	590.88
200435303	9.82	431.25
200435304	11.13	427.98
200435306	3.73	302.48
200435307	8.05	295.52
200435308	20.55	239.60
200435309	16.09	244.26
200435310	11.30	222.48
200435311	15.79	253.48
200435401	4.95	254.50
200435402	9.10	290.73
200435403	12.46	251.02
200435404	13.30	270.41
200435405	11.95	372.24
200435407	8.50	213.00
200435412	11.69	264.17
200435413	7.92	145.05
200435801	12.02	227.27
200435802	11.37	454.36
200435803	3.84	116.09
200436001	11.03	418.70
200436003	6.20	364.63

Animal ID	Estradiol (pg/mL)	IGF-1 (ng/mL)
200436004	7.29	340.85
200436005	11.53	325.21
200436007	13.72	261.76
200436008	13.59	562.30
200436009	26.56	214.14
200436010	13.56	339.88
200436011	9.00	424.65
200436012	16.10	332.88
200436301	19.71	424.87
200436303	20.11	156.52
200436309	7.09	145.65
200436312	11.75	219.12
200436401	10.15	343.00
200436402	7.85	134.23
200436403	6.27	145.81
200436405	8.17	173.98
200436601	6.30	258.60
200436602	10.53	240.08
200436603	9.74	389.69
200436604	13.00	311.43
200436606	14.99	120.57
200436607	119.40	512.17
200436608	12.46	96.27
200436612	7.59	265.56
200436901	7.92	348.45
200436903	7.67	268.40
200436905	7.33	399.80
200436909	13.87	371.03
200436911	8.42	705.36
200436912	7.75	424.84
200436913	9.98	290.65
200437301	19.56	324.59
200437303	10.81	530.28
200437305	9.47	348.74
200437307	14.43	183.12
200437309	12.20	239.36
200437401	3.27	477.57
200437402	8.99	360.37
200437404	6.39	225.13
200437406	8.97	168.68
200437502	6.68	397.32
200437504	8.90	408.13
200437506	9.40	317.28
200437509	8.16	258.84
200437801	4.76	247.13
200437802	6.51	120.25
200437803	9.83	378.40
200438101	12.68	428.18
200438102	5.50	316.16
200438104	9.37	357.01
200438107	10.21	171.16
200438401	10.04	362.21
200438402	12.11	290.48
200438403	9.97	238.56
200438404	10.60	223.73
200438406	10.73	233.31

Animal ID	Estradiol (pg/mL)	IGF-1 (ng/mL)
200438408	12.80	336.29
200438501	14.40	394.85
200438505	13.25	606.67
200438511	14.56	361.86
200438513	12.24	238.07
200438602	13.76	392.03
200438603	9.38	196.28
200438604	7.20	295.92
200438605	12.86	223.85
200438606	11.18	124.03
200438607	8.26	196.31
200438608	9.33	82.24
200438609	12.17	216.17
200438610	9.85	253.93
200438611	4.93	158.64
200438701	7.57	277.13
200438703	8.81	177.07
200438705	7.90	487.31
200438707	13.45	274.60
200438709	5.64	390.63
200439202	10.50	231.46
200439204	13.87	399.85
200439205	12.43	397.10
200439206	6.72	616.30
200439210	30.67	284.42
200439401	9.04	3.04
200439404	8.15	280.34
200439406	3.16	280.25
200439501	7.38	307.22
200439502	9.18	360.53
200439503	8.46	377.64
200439504	8.76	282.87
200439505	8.18	355.53
200439901	8.34	268.01
200439902	11.48	177.21
200439903	10.21	225.60
200439904	4.90	117.95
200440602	7.87	248.26
200440605	10.34	470.35
200440903	11.85	234.02
200440907	8.82	247.11
200440909	10.78	282.74
200441201	10.35	347.53
200441202	8.85	296.02
200441203	13.90	88.16
200441205	10.49	170.93
200441207	8.09	200.26
200441209	9.80	252.21
200441211	11.46	189.10

Total Number	Slaughter Order/Day	Marc Animal ID	Treatment	Slaughter Date	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Urobilinogen	Nitrite	Leukocytes
1	1	200433509	Control	13-Dec-2004	0	0	0	1.020	80	6.5	0.1	0	0	0
2	2	200437307	Control	13-Dec-2004	0	0	0	1.010	0	8.0	0.3	0	0	0
3	3	200430508	Control	13-Dec-2004	0	0	0	1.030	0	6.0	0.3	0	0	0
4	4	200437301	Control	13-Dec-2004	0	0	0	1.020	0	7.5	0.3	0	0	0
5	5	200433409	Control	13-Dec-2004	0	0	0	1.020	0	7.5	0.3	0	0	0
6	6	200435309	Control	13-Dec-2004	0	0	0	1.020	0	7.0	0.3	0	0	0
7	7	200430503	Control	13-Dec-2004	0	0	0	1.030	0	7.0	0.1	0	0	0
8	8	200433510	Control	13-Dec-2004	0	0	0	1.020	0	7.0	0.1	0	0	0
9	9	200430709	Clone	13-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
10	10	200431505	Clone	13-Dec-2004	0	0	0	1.025	0	6.5	0.1	0	0	0
11	11	200430706	Clone	13-Dec-2004	0	0	0	1.030	10	7.0	0.1	0	0	0
12	12	200431408	Clone	13-Dec-2004	0	0	0	1.025	0	6.5	0.1	0	0	0
13	13	200430906	Clone	13-Dec-2004	0	0	0	1.030	0	7.0	0.3	0	0	0
14	14	200430707	Clone	13-Dec-2004	0	0	0	1.005	0	8.0	0.1	0	0	0
15	15	200430604	Clone	13-Dec-2004	0	0	0	1.010	10	8.0	0.1	0	0	0
16	16	200432405	Clone	13-Dec-2004	0	0	0	1.020	0	7.5	0.1	0	0	0
17	17	200431411	Clone	13-Dec-2004	0	0	0	1.015	0	7.5	0.1	0	0	0
18	18	200431006	Clone	13-Dec-2004	0	0	0	1.025	0	6.5	0.1	0	0	0
19	19	200436009	Clone	13-Dec-2004	0	0	0	1.010	0	7.5	0.1	0	0	0
20	20	200431502	Clone	13-Dec-2004	0	0	0	1.010	0	8.0	0.0	0	0	0
21	21	200430711	Clone	13-Dec-2004	0	0	0	1.010	0	8.0	0.3	0	0	0
22	22	200430710	Clone	13-Dec-2004	0	0	0	1.010	10	7.5	0.1	0	0	0
23	23	200430805	Clone	13-Dec-2004	0	0	0	1.015	0	7.5	0.1	0	0	0
24	24	200430902	Clone	13-Dec-2004	0	0	0	1.020	0	6.5	0.0	0	0	0
25	25	200431608	Clone	13-Dec-2004	0	0	0	1.010	0	7.5	0.0	0	0	0
26	26	200433605	Clone	13-Dec-2004	0	0	0	1.005	0	8.0	0.1	0	0	0
27	27	200430905	Clone	13-Dec-2004	0	0	0	1.015	0	7.5	0.1	0	0	0
28	28	200433807	Clone	13-Dec-2004	0	0	0	1.010	0	7.5	0.1	0	0	0
29	29	200430804	Clone	13-Dec-2004	0	0	0	1.010	10	7.5	0.0	0	0	0
30	30	200431404	Clone	13-Dec-2004	0	0	0	1.010	0	7.5	0.1	0	0	0
31	31	200431009	Clone	13-Dec-2004	0	0	0	1.010	10	7.5	0.1	0	0	0
32	1	200432911	Control	15-Dec-2004	0	0	0	1.005	0	8.0	0.1	0	0	0

Total Number	Slaughter Order/Day	Marc Animal ID	Treatment	Slaughter Date	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Urobilinogen	Nitrite	Leukocytes
33	2	200434001	Control	15-Dec-2004	0	0	0	1.020	0	8.0	0.1	0	0	0
34	3	200431802	Control	15-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
35	4	200434301	Control	15-Dec-2004	0	0	0	1.010	0	7.5	0.1	0	0	0
36	5	200435303	Control	15-Dec-2004	0	0	0	1.005	0	8.0	0.1	0	0	0
37	6	200437305	Control	15-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
38	7	200432609	Control	15-Dec-2004	0	0	0	1.015	0	8.0	0.1	0	0	0
39	8	200437303	Control	15-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
40	9	200434202	Control	15-Dec-2004	0	0	0	1.010	80	8.0	0.1	0	0	0
41	10	200433301	Control	15-Dec-2004	0	0	0	1.005	0	8.0	0.1	0	0	0
42	11	200437506	Clone	15-Dec-2004	0	0	0	1.010	10	7.5	0.1	0	0	0
43	12	200436011	Clone	15-Dec-2004	0	0	0	1.005	0	8.0	0.1	0	0	0
44	13	200430708	Clone	15-Dec-2004	0	0	0	1.005	0	8.0	0.1	0	0	0
45	14	200430705	Clone	15-Dec-2004	0	0	0	1.005	0	8.0	0.1	0	0	0
46	15	200431606	Clone	15-Dec-2004	0	0	0	1.005	0	8.0	0.1	0	0	0
47	16	200434507	Clone	15-Dec-2004	0	0	0	1.030	80	6.0	0.1	0	0	0
48	17	200431504	Clone	15-Dec-2004	0	0	0	1.005	0	8.0	0.1	0	0	0
49	18	200436012	Clone	15-Dec-2004	0	0	0	1.020	0	7.5	0.1	0	0	0
50	19	200433601	Clone	15-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
51	20	200431008	Clone	15-Dec-2004	0	0	0	1.010	0	7.5	0.1	0	0	0
52	21	200430609	Clone	15-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
53	22	200431501	Clone	15-Dec-2004	0	0	0	1.010	0	7.5	0.1	0	0	0
54	23	200436909	Clone	15-Dec-2004	0	0	0	1.005	0	8.0	0.1	0	0	0
55	24	200431407	Clone	15-Dec-2004	0	0	0	1.005	0	8.0	0.1	0	0	0
56	25	200431405	Clone	15-Dec-2004	0	0	0	1.010	0	7.5	0.1	0	0	0
57	26	200430605	Clone	15-Dec-2004	0	0	0	1.010	0	7.5	0.1	0	0	0
58	27	200433201	Clone	15-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
59	28	200430607	Clone	15-Dec-2004	0	0	0	1.015	200	8.0	0.1	0	0	0
60	1	200431604	Clone	20-Dec-2004	0	0	0	1.010	0	8.0	0.3	0	0	0
61	2	200433603	Clone	20-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
62	3	200432103	Clone	20-Dec-2004	0	0	0	1.020	0	7.5	0.3	0	0	0
63	4	200430703	Clone	20-Dec-2004	0	0	0	1.005	0	8.0	1.0	0	0	0
64	5	200437504	Clone	20-Dec-2004	0	0	0	1.010	0	8.0	1.0	0	0	0

Total Number	Slaughter Order/Day	Marc Animal ID	Treatment	Slaughter Date	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Urobilinogen	Nitrite	Leukocytes
65	6	200434512	Clone	20-Dec-2004	0	0	0	1.005	0	8.0	1.0	0	0	0
66	7	200434403	Clone	20-Dec-2004	0	0	0	1.010	0	8.0	0.3	0	0	0
67	8	200432704	Clone	20-Dec-2004	0	0	0	1.010	10	8.0	0.3	0	0	0
68	9	200433711	Clone	20-Dec-2004	0	0	0	1.010	80	8.0	0.3	0	0	0
69	10	200430903	Clone	20-Dec-2004	0	0	0	1.030	0	7.0	0.3	0	0	0
70	11	200433205	Clone	20-Dec-2004	0	0	0	1.010	10	8.0	1.0	0	0	0
71	12	200432004	Clone	20-Dec-2004	0	0	0	1.010	0	8.0	0.3	0	0	0
72	13	200432404	Clone	20-Dec-2004	0	0	0	1.020	0	7.5	0.3	0	0	0
73	14	200431003	Clone	20-Dec-2004	0	0	0	1.005	0	8.0	0.1	0	0	0
74	15	200431406	Clone	20-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
75	16	200432108	Clone	20-Dec-2004	0	0	0	1.020	0	8.0	3.0	0	0	0
76	17	200431610	Clone	20-Dec-2004	0	0	0	1.005	10	8.0	0.3	0	0	0
77	18	200431011	Clone	20-Dec-2004	0	0	0	1.005	0	8.0	3.0	0	0	0
78	19	200434402	Clone	20-Dec-2004	0	0	0	1.005	0	8.0	1.0	0	0	0
79	20	200432310	Clone	20-Dec-2004	0	0	0	1.005	0	8.0	1.0	0	0	0
80	21	200432308	Clone	20-Dec-2004	0	0	0	1.010	200	8.0	1.0	0	0	0
81	22	200432406	Clone	20-Dec-2004	0	0	0	1.005	0	8.0	0.3	0	0	0
82	23	200432706	Clone	20-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
83	24	200434601	Clone	20-Dec-2004	0	0	0	1.010	0	8.0	0.3	0	0	0
84	25	200432705	Clone	20-Dec-2004	0	0	0	1.005	10	8.0	0.3	0	0	0
85	26	200434401	Clone	20-Dec-2004	0	0	0	1.020	0	8.0	1.0	0	0	0
86	1	200433505	Control	21-Dec-2004	0	0	0	1.005	0	8.0	0.3	0	0	0
87	2	200434206	Control	21-Dec-2004	0	0	0	1.025	10	7.5	3.0	0	0	0
88	3	200432602	Control	21-Dec-2004	0	0	0	1.005	0	8.0	0.3	0	0	0
89	4	200438511	Control	21-Dec-2004	0	0	0	1.010	0	8.0	1.0	0	0	0
90	5	200438408	Control	21-Dec-2004	0	0	0	1.020	10	8.0	3.0	0	0	0
91	6	200438513	Control	21-Dec-2004	0	0	0	1.020	0	8.0	3.0	0	0	0
92	7	200433405	Control	21-Dec-2004	0	0	0	1.020	0	8.0	1.0	0	0	0
93	8	200435302	Control	21-Dec-2004	0	0	0	1.020	0	8.0	0.1	0	0	0
94	9	200435311	Control	21-Dec-2004	0	0	0	1.025	200	7.5	0.3	0	0	0
95	10	200435407	Control	21-Dec-2004	0	0	0	1.010	0	8.0	0.3	0	0	0
96	11	200433512	Control	21-Dec-2004	0	0	0	1.010	10	8.0	0.3	0	0	0

Total Number	Slaughter Order/Day	Marc Animal ID	Treatment	Slaughter Date	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Urobilinogen	Nitrite	Leukocytes
97	12	200434808	Control	21-Dec-2004	0	0	0	1.010	0	8.0	1.0	0	0	0
98	13	200432607	Control	21-Dec-2004	0	0	0	1.010	0	8.0	3.0	0	0	0
99	14	200430510	Control	21-Dec-2004	0	0	0	1.015	0	8.0	3.0	0	0	0
100	15	200431805	Control	21-Dec-2004	0	0	0	1.020	0	8.0	0.1	0	0	0
101	16	200434807	Control	21-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
102	17	200433306	Control	21-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
103	18	200434005	Control	21-Dec-2004	0	0	0	1.010	0	8.0	0.0	0	0	0
104	19	200432610	Control	21-Dec-2004	0	0	0	1.010	10	8.0	0.1	0	0	0
105	20	200435404	Control	21-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
106	21	200433511	Control	21-Dec-2004	0	0	0	1.020	0	8.0	0.1	0	0	0
107	22	200433307	Control	21-Dec-2004	0	0	0	1.020	0	7.5	0.1	0	0	0
108	23	200434003	Control	21-Dec-2004	0	0	0	1.020	0	8.0	3.0	0	0	0
109	24	200433408	Control	21-Dec-2004	0	0	0	1.010	10	8.0	0.1	0	0	0
110	25	200432907	Control	21-Dec-2004	0	0	0	1.010	0	8.0	1.0	0	0	0
111	26	200432913	Control	21-Dec-2004	0	0	0	1.030	25	6.0	1.0	0	0	0
112	27	200432601	Control	21-Dec-2004	0	0	0	1.030	0	6.5	1.0	0	0	0
113	28	200434203	Control	21-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
114	29	200434901	Control	21-Dec-2004	0	0	0	1.020	10	8.0	0.3	0	0	0
115	30	200434806	Control	21-Dec-2004	0	0	0	1.015	0	8.0	0.1	0	0	0
116	1	200431801	Control	27-Dec-2004	0	0	0	1.010	0	8.0	3.0	0	0	0
117	2	200435308	Control	27-Dec-2004	0	0	0	1.020	10	7.5	0.1	0	0	0
118	3	200434902	Control	27-Dec-2004	0	0	0	1.015	0	8.0	3.0	0	0	0
119	4	200436303	Control	27-Dec-2004	0	0	0	1.020	0	8.0	0.3	0	0	0
120	5	200438401	Control	27-Dec-2004	0	0	0	1.005	0	8.0	1.0	0	0	0
121	6	200435412	Control	27-Dec-2004	0	0	0	1.010	10	8.0	3.0	0	0	0
122	7	200438404	Control	27-Dec-2004	0	0	0	1.015	10	8.0	0.3	0	0	0
123	8	200436309	Control	27-Dec-2004	0	0	0	1.010	0	8.0	3.0	0	0	0
124	9	200438406	Control	27-Dec-2004	0	0	0	1.010	10	8.0	1.0	0	0	0
125	10	200437309	Control	27-Dec-2004	0	0	0	1.015	0	8.0	0.3	0	0	0
126	11	200434007	Control	27-Dec-2004	0	0	0	1.005	0	8.0	1.0	0	0	0
127	12	200440605	Control	27-Dec-2004	0	0	0	1.005	0	8.0	0.3	0	0	0
128	13	200438505	Control	27-Dec-2004	0	0	0	1.010	0	8.0	3.0	0	0	0

Total Number	Slaughter Order/Day	Marc Animal ID	Treatment	Slaughter Date	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Urobilinogen	Nitrite	Leukocytes
129	14	200434809	Control	27-Dec-2004	0	0	0	1.015	0	8.0	1.0	0	0	0
130	15	200439210	Control	27-Dec-2004	0	0	0	1.005	0	8.0	0.3	0	0	0
131	16	200431704	Clone	27-Dec-2004	0	0	0	1.010	80	8.0	0.1	0	0	0
132	17	200436010	Clone	27-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
133	18	200431703	Clone	27-Dec-2004	0	0	0	1.015	80	8.0	0.3	0	0	0
134	19	200431706	Clone	27-Dec-2004	0	0	0	1.010	0	8.0	3.0	0	0	0
135	20	200431403	Clone	27-Dec-2004	0	0	0	1.010	0	8.0	1.0	0	0	0
136	21	200431401	Clone	27-Dec-2004	0	0	0	1.010	0	8.0	0.1	0	0	0
137	22	200430802	Clone	27-Dec-2004	0	0	0	1.005	0	8.0	1.0	0	0	0
138	23	200432402	Clone	27-Dec-2004	0	0	0	1.010	0	8.0	0.3	0	0	0
139	24	200433803	Clone	27-Dec-2004	0	0	0	1.005	0	8.0	3.0	0	0	0
140	25	200437404	Clone	27-Dec-2004	0	0	0	1.005	0	8.0	1.0	0	0	0
141	26	200433602	Clone	27-Dec-2004	0	0	0	1.010	0	8.0	1.0	0	0	0
142	27	200430704	Clone	27-Dec-2004	0	0	0	1.010	0	8.0	3.0	0	0	0
143	28	200436402	Clone	27-Dec-2004	0	0	0	1.010	0	7.5	0.3	0	0	0
144	29	200437802	Clone	27-Dec-2004	0	0	0	1.030	0	6.5	0.1	0	0	0
145	30	200433206	Clone	27-Dec-2004	0	0	0	1.010	0	7.5	0.1	0	0	0
146	31	200432105	Clone	27-Dec-2004	0	0	0	1.020	0	8.0	0.3	0	0	0
147	32	200432702	Clone	27-Dec-2004	0	0	0	1.005	0	8.0	0.3	0	0	0
148	33	200437801	Clone	27-Dec-2004	0	0	0	1.005	0	8.0	3.0	0	0	0
149	34	200438107	Clone	27-Dec-2004	0	0	0	1.015	0	7.5	0.1	0	0	0
150	35	200433809	Clone	27-Dec-2004	0	0	0	1.010	0	8.0	1.0	0	0	0
151	1	200435401	Control	29-Dec-04	0	0	0	1.010	0	8.0	1.0	0	0	0
152	2	200433404	Control	29-Dec-04	0	0	0	1.020	0	8.0	0.3	0	0	0
153	3	200439206	Control	29-Dec-04	0	0	0	1.020	10	7.5	3.0	0	0	0
154	4	200432604	Control	29-Dec-04	0	0	0	1.025	10	6.5	0.3	0	0	0
155	5	200434310	Control	29-Dec-04	0	0	0	1.010	10	8.0	3.0	0	0	0
156	6	200434004	Control	29-Dec-04	0	0	0	1.010	200	8.0	1.0	0	0	0
157	7	200435413	Control	29-Dec-04	0	0	0	1.015	10	8.0	3.0	0	0	0
158	8	200440602	Control	29-Dec-04	0	0	0	1.030	10	6.0	1.0	0	0	0
159	9	200432909	Control	29-Dec-04	0	0	0	1.015	0	8.0	3.0	0	0	0
160	10	200439202	Control	29-Dec-04	0	0	0	1.010	0	8.0	0.3	0	0	0

Total Number	Slaughter Order/Day	Marc Animal ID	Treatment	Slaughter Date	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Urobilinogen	Nitrite	Leukocytes
161	11	200434802	Control	29-Dec-04	0	0	0	1.010	0	8.0	1.0	0	0	0
162	12	200434201	Control	29-Dec-04	0	0	0	1.005	0	8.5	3.0	0	0	0
163	13	200439406	Control	29-Dec-04	0	0	0	1.010	0	8.0	3.0	0	0	0
164	14	200435402	Control	29-Dec-04	0	0	0	1.010	0	8.0	1.0	0	0	0
165	15	200435306	Control	29-Dec-04	0	0	0	1.010	0	8.0	1.0	0	0	0
166	16	200432002	Clone	29-Dec-04	0	0	0	1.010	0	8.0	3.0	0	0	0
167	17	200432101	Clone	29-Dec-04	0	0	0	1.025	0	6.0	0.0	0	0	0
168	18	200432503	Clone	29-Dec-04	0	0	0	1.010	0	7.5	0.1	0	0	0
169	19	200431607	Clone	29-Dec-04	0	0	0	1.015	0	8.0	0.3	0	0	0
170	20	200433710	Clone	29-Dec-04	0	0	0	1.020	10	7.5	0.1	0	0	0
171	21	200436405	Clone	29-Dec-04	0	0	0	1.010	0	8.0	0.3	0	0	0
172	22	200432006	Clone	29-Dec-04	0	0	0	1.015	10	8.0	0.1	0	0	0
173	23	200430803	Clone	29-Dec-04	0	0	0	1.025	10	7.5	0.1	0	0	0
174	24	200436911	Clone	29-Dec-04	0	0	0	1.010	0	8.0	0.1	0	0	0
175	25	200433604	Clone	29-Dec-04	0	0	0	1.005	0	8.0	0.1	0	0	0
176	26	200432501	Clone	29-Dec-04	0	0	0	1.010	0	7.5	0.1	0	0	0
177	27	200433207	Clone	29-Dec-04	0	0	0	1.010	10	8.0	0.1	0	0	0
178	28	200432302	Clone	29-Dec-04	0	0	0	1.020	10	8.0	0.1	0	0	0
179	29	200433804	Clone	29-Dec-04	0	0	0	1.005	0	8.5	0.3	0	0	0
180	30	200435802	Clone	29-Dec-04	0	0	0	1.010	10	8.5	0.1	0	0	0
181	31	200433808	Clone	29-Dec-04	0	0	0	1.020	10	8.0	0.1	0	0	0
182	32	200430606	Clone	29-Dec-04	0	0	0	1.020	10	7.5	0.3	0	0	0
183	1	200433402	Control	3-Jan-05	0	0	0	1.020	0	7.5	1.0	0	0	0
184	2	200433504	Control	3-Jan-05	0	0	0	1.020	0	7.5	1.0	0	0	0
185	3	200430502	Control	3-Jan-05	0	0	0	1.005	0	8.0	1.0	0	0	0
186	4	200433403	Control	3-Jan-05	0	0	0	1.015	0	8.0	0.3	0	0	0
187	5	200436310	Control	3-Jan-05	0	0	0	1.030	0	6.0	1.0	0	0	0
188	6	200438503	Control	3-Jan-05	0	0	0	1.025	0	7.0	3.0	0	0	0
189	7	200440601	Control	3-Jan-05	0	0	0	1.030	0	6.5	1.0	0	0	0
190	8	200435409	Control	3-Jan-05	0	0	0	1.010	200	8.0	1.0	0	0	0
191	9	200434009	Control	3-Jan-05	0	0	0	1.010	25	8.0	0.1	0	0	0
192	10	200440901	Control	3-Jan-05	0	0	0	1.015	0	8.0	0.3	0	0	0

Total Number	Slaughter Order/Day	Marc Animal ID	Treatment	Slaughter Date	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Urobilinogen	Nitrite	Leukocytes
193	11	200440906	Control	3-Jan-05	0	0	0	1.020	0	7.5	0.3	0	0	0
194	12	200433501	Control	3-Jan-05	0	0	0	1.020	25	7.5	0.1	0	0	0
195	13	200430509	Control	3-Jan-05	0	0	0	1.015	80	8.0	1.0	0	0	0
196	14	200436305	Control	3-Jan-05	0	0	0	1.010	0	7.5	0.1	0	0	0
197	15	200431012	Clone	3-Jan-05	0	0	0	1.020	25	7.5	0.3	0	0	0
198	16	200437501	Clone	3-Jan-05	0	0	0	1.010	80	8.0	1.0	0	0	0
199	17	200433203	Clone	3-Jan-05	0	0	0	1.030	0	6.0	0.1	0	0	0
200	18	200436006	Clone	3-Jan-05	0	0	0	1.020	25	8.0	3.0	0	0	0
201	19	200434608	Clone	3-Jan-05	0	0	0	1.010	10	8.0	1.0	0	0	0
202	20	200430608	Clone	3-Jan-05	0	0	0	1.005	10	8.0	0.3	0	0	0
203	21	200431007	Clone	3-Jan-05	0	0	0	1.005	80	8.0	0.0	0	0	0
204	22	200433806	Clone	3-Jan-05	0	0	0	1.030	10	6.5	0.3	0	0	0
205	23	200432007	Clone	3-Jan-05	0	0	0	1.005	0	8.0	1.0	0	0	0
206	24	200431402	Clone	3-Jan-05	0	0	0	1.005	0	8.0	0.3	0	0	0
207	25	200432104	Clone	3-Jan-05	0	0	0	1.015	10	8.0	0.3	0	0	0
208	26	200431409	Clone	3-Jan-05	0	0	0	1.010	0	8.0	0.1	0	0	0
209	27	200437510	Clone	3-Jan-05	0	0	0	1.010	25	7.5	0.3	0	0	0
210	28	200436404	Clone	3-Jan-05	0	0	0	1.010	0	8.0	0.1	0	0	0
211	29	200433712	Clone	3-Jan-05	0	0	0	1.010	0	8.0	0.1	0	0	0
212	30	200436610	Clone	3-Jan-05	0	0	0	1.020	0	7.0	0.1	0	0	0
213	31	200437804	Clone	3-Jan-05	0	0	0	1.010	10	8.0	0.3	0	0	0
214	1	200437302	Control	6-Jan-05	0	0	0	1.030	80	6.5	1.0	0	0	0
215	2	200433507	Control	6-Jan-05	0	0	0	1.010	200	7.5	1.0	0	0	0
216	3	200434306	Control	6-Jan-05	0	0	0	1.010	0	7.5	0.3	0	0	0
217	4	200433503	Control	6-Jan-05	0	0	0	1.010	0	7.5	0.3	0	0	0
218	5	200430501	Control	6-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
219	6	200440603	Control	6-Jan-05	0	0	0	1.020	0	7.0	1.0	0	0	0
220	7	200435410	Control	6-Jan-05	0	0	0	1.030	0	6.0	0.1	0	0	0
221	8	200434008	Control	6-Jan-05	0	0	0	1.020	0	7.5	3.0	0	0	0
222	9	200435411	Control	6-Jan-05	0	0	0	1.020	10	7.0	0.3	0	0	0
223	10	200438706	Control	6-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
224	11	200439208	Control	6-Jan-05	0	0	0	1.020	200	7.5	1.0	0	0	0

Total Number	Slaughter Order/Day	Marc Animal ID	Treatment	Slaughter Date	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Urobilinogen	Nitrite	Leukocytes
225	12	200440911	Control	6-Jan-05	0	0	0	1.030	10	6.0	1.0	0	0	0
226	13	200435305	Control	6-Jan-05	0	0	0	1.030	80	6.0	0.3	0	0	0
227	14	200433502	Control	6-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
228	15	200435301	Control	6-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
229	16	200434303	Control	6-Jan-05	0	0	0	1.010	0	8.0	1.0	0	0	0
230	17	200431603	Clone	6-Jan-05	0	0	0	1.005	0	8.0	1.0	0	0	0
231	18	200433003	Clone	6-Jan-05	0	0	0	1.015	0	7.5	0.3	0	0	0
232	19	200432312	Clone	6-Jan-05	0	0	0	1.030	200	6.0	3.0	0	0	0
233	20	200432005	Clone	6-Jan-05	0	0	0	1.015	80	8.0	0.1	0	0	0
234	21	200436902	Clone	6-Jan-05	0	0	0	1.015	0	7.5	0.1	0	0	0
235	22	200436002	Clone	6-Jan-05	0	0	0	1.020	0	7.5	0.3	0	0	0
236	23	200431410	Clone	6-Jan-05	0	0	0	1.010	10	7.5	0.1	0	0	0
237	24	200432401	Clone	6-Jan-05	0	0	0	1.010	10	8.0	3.0	0	0	0
238	25	200432102	Clone	6-Jan-05	0	0	0	1.005	0	8.0	1.0	0	0	0
239	26	200436013	Clone	6-Jan-05	0	0	0	1.030	0	6.5	0.3	0	0	0
240	27	200432504	Clone	6-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
241	28	200438106	Clone	6-Jan-05	0	0	0	1.010	0	8.0	0.1	0	0	0
242	29	200432001	Clone	6-Jan-05	0	0	0	1.020	0	8.0	1.0	0	0	0
243	30	200438105	Clone	6-Jan-05	0	0	0	1.010	0	8.0	1.0	0	0	0
244	31	200431506	Clone	6-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
245	32	200437408	Clone	6-Jan-05	0	0	0	1.020	0	7.0	1.0	0	0	0
246	1	200433506	Control	10-Jan-05	0	0	0	1.010	0	7.5	0.3	0	0	0
247	2	200434302	Control	10-Jan-05	0	0	0	1.020	0	7.5	0.3	0	0	0
248	3	200434903	Control	10-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
249	4	200435405	Control	10-Jan-05	0	0	0	1.030	10	6.5	1.0	0	0	0
250	5	200434905	Control	10-Jan-05	0	0	0	1.020	25	7.0	3.0	0	0	0
251	6	200432906	Control	10-Jan-05	0	0	0	1.020	200	7.0	3.0	0	0	0
252	7	200435310	Control	10-Jan-05	0	0	0	1.020	10	7.5	0.3	0	0	0
253	8	200438402	Control	10-Jan-05	0	0	0	1.020	10	7.5	1.0	0	0	0
254	9	200438501	Control	10-Jan-05	0	0	0	1.020	0	7.5	0.3	0	0	0
255	10	200434803	Control	10-Jan-05	0	0	0	1.010	0	7.5	0.3	0	0	0
256	11	200432904	Control	10-Jan-05	0	0	0	1.005	0	8.0	3.0	0	0	0

Total Number	Slaughter Order/Day	Marc Animal ID	Treatment	Slaughter Date	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Urobilinogen	Nitrite	Leukocytes
257	12	200434904	Control	10-Jan-05	0	0	0	1.005	0	8.0	3.0	0	0	0
258	13	200430506	Control	10-Jan-05	0	0	0	1.010	0	8.0	1.0	0	0	0
259	14	200435403	Control	10-Jan-05	0	0	0	1.005	0	8.0	1.0	0	0	0
260	15	200434804	Control	10-Jan-05	0	0	0	1.005	0	8.0	1.0	0	0	0
261	16	200434509	Clone	10-Jan-05	0	0	0	1.020	25	7.0	0.3	0	0	0
262	17	200434607	Clone	10-Jan-05	0	0	0	1.010	0	7.0	0.3	0	0	0
263	18	200437502	Clone	10-Jan-05	0	0	0	1.005	0	8.0	1.0	0	0	0
264	19	200434504	Clone	10-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
265	20	200436007	Clone	10-Jan-05	0	0	0	1.005	0	8.0	0.3	0	0	0
266	21	200432407	Clone	10-Jan-05	0	0	0	1.005	0	8.0	1.0	0	0	0
267	22	200434501	Clone	10-Jan-05	0	0	0	1.005	0	8.0	1.0	0	0	0
268	23	200430610	Clone	10-Jan-05	0	0	0	1.020	80	7.5	0.3	0	0	0
269	24	200436608	Clone	10-Jan-05	0	0	0	1.020	0	6.5	0.1	0	0	0
270	25	200433002	Clone	10-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
271	26	200431701	Clone	10-Jan-05	0	0	0	1.005	0	8.0	0.3	0	0	0
272	27	200433202	Clone	10-Jan-05	0	0	0	1.005	0	8.0	0.1	0	0	0
273	28	200434503	Clone	10-Jan-05	0	0	0	1.025	0	6.0	0.1	0	0	0
274	29	200430701	Clone	10-Jan-05	0	0	0	1.020	0	7.0	1.0	0	0	0
275	30	200435801	Clone	10-Jan-05	0	0	0	1.010	0	7.5	0.1	0	0	0
276	1	200430504	Control	12-Jan-05	0	0	0	1.010	0	7.5	0.1	0	0	0
277	2	200434006	Control	12-Jan-05	0	0	0	1.025	200	6.5	1.0	0	0	0
278	3	200436312	Control	12-Jan-05	0	0	0	1.010	0	7.5	3.0	0	0	0
279	4	200432901	Control	12-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
280	5	200440903	Control	12-Jan-05	0	0	0	1.010	80	8.0	3.0	0	0	0
281	6	200434304	Control	12-Jan-05	0	0	0	1.020	0	6.5	0.3	0	0	0
282	7	200436913	Clone	12-Jan-05	0	0	0	1.005	10	8.0	1.0	0	0	0
283	8	200431702	Clone	12-Jan-05	0	0	0	1.005	0	8.0	0.1	0	0	0
284	9	200434502	Clone	12-Jan-05	0	0	0	1.005	0	8.0	0.1	0	0	0
285	10	200436912	Clone	12-Jan-05	0	0	0	1.020	0	6.0	0.1	0	0	0
286	11	200431004	Clone	12-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
287	12	200436603	Clone	12-Jan-05	0	0	0	1.010	0	7.5	0.1	0	0	0
288	13	200432403	Clone	12-Jan-05	0	0	0	1.005	0	8.0	1.0	0	0	0

Total Number	Slaughter Order/Day	Marc Animal ID	Treatment	Slaughter Date	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Urobilinogen	Nitrite	Leukocytes
289	14	200431605	Clone	12-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
290	15	200431010	Clone	12-Jan-05	0	0	0	1.005	0	8.0	1.0	0	0	0
291	16	200430611	Clone	12-Jan-05	0	0	0	1.030	10	6.0	0.1	0	0	0
292	17	200431002	Clone	12-Jan-05	0	0	0	1.015	0	7.5	0.3	0	0	0
293	18	200436005	Clone	12-Jan-05	0	0	0	1.005	80	8.0	0.3	0	0	0
294	19	200436606	Clone	12-Jan-05	0	0	0	1.010	0	7.5	0.3	0	0	0
295	20	200436602	Clone	12-Jan-05	0	0	0	1.005	0	8.0	3.0	0	0	0
296	21	200431601	Clone	12-Jan-05	0	0	0	1.000	0	8.0	1.0	0	0	0
297	22	200438104	Clone	12-Jan-05	0	0	0	1.005	0	8.0	1.0	0	0	0
298	23	200437406	Clone	12-Jan-05	0	0	0	1.005	0	8.0	0.3	0	0	0
299	24	200435803	Clone	12-Jan-05	0	0	0	1.005	200	8.0	1.0	0	0	0
300	25	200434511	Clone	12-Jan-05	0	0	0	1.020	0	7.0	0.1	0	0	0
301	1	200438707	Control	18-Jan-05	0	0	0	1.005	0	8.0	0.1	0	0	0
302	2	200440909	Control	18-Jan-05	0	0	0	1.030	0	6.0	1.0	0	0	0
303	3	200440907	Control	18-Jan-05	0	0	0	1.030	80	6.5	3.0	0	0	0
304	4	200433406	Control	18-Jan-05	0	0	0	1.010	0	7.5	1.0	0	0	0
305	5	200433303	Control	18-Jan-05	0	0	0	1.005	0	8.0	0.3	0	0	0
306	6	200439205	Control	18-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
307	7	200433302	Control	18-Jan-05	0	0	0	1.010	0	7.5	0.1	0	0	0
308	8	200441202	Control	18-Jan-05	0	0	0	1.010	0	7.5	1.0	0	0	0
309	9	200439902	Control	18-Jan-05	0	0	0	1.010	0	7.0	0.1	0	0	0
310	10	200439903	Control	18-Jan-05	0	0	0	1.010	200	7.5	0.1	0	0	0
311	11	200431708	Clone	18-Jan-05	0	0	0	1.010	200	8.0	3.0	0	0	0
312	12	200438607	Clone	18-Jan-05	0	0	0	1.010	0	8.0	0.1	0	0	0
313	13	200436604	Clone	18-Jan-05	0	0	0	1.010	0	7.5	3.0	0	0	0
314	14	200434101	Clone	18-Jan-05	0	0	0	1.010	0	7.5	0.1	0	0	0
315	15	200436607	Clone	18-Jan-05	0	0	0	1.015	200	7.5	1.0	0	0	0
316	16	200432507	Clone	18-Jan-05	0	0	0	1.020	0	7.5	0.3	0	0	0
317	17	200438610	Clone	18-Jan-05	0	0	0	1.025	0	6.5	0.1	0	0	0
318	18	200432008	Clone	18-Jan-05	0	0	0	1.010	0	7.5	0.3	0	0	0
319	19	200439505	Clone	18-Jan-05	0	0	0	1.005	0	7.5	0.3	0	0	0
320	20	200436901	Clone	18-Jan-05	0	0	0	1.010	0	8.0	0.1	0	0	0

Total Number	Slaughter Order/Day	Marc Animal ID	Treatment	Slaughter Date	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Urobilinogen	Nitrite	Leukocytes
321	21	200433007	Clone	18-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
322	22	200431503	Clone	18-Jan-05	0	0	0	1.020	0	7.0	0.1	0	0	0
323	23	200436001	Clone	18-Jan-05	0	0	0	1.005	0	8.0	0.3	0	0	0
324	24	200438101	Clone	18-Jan-05	0	0	0	1.005	0	8.0	0.3	0	0	0
325	25	200432304	Clone	18-Jan-05	0	0	0	1.025	0	6.5	0.1	0	0	0
326	26	200433709	Clone	18-Jan-05	0	0	0	1.030	0	6.0	0.1	0	0	0
327	27	200438602	Clone	18-Jan-05	0	0	0	1.010	0	7.5	0.3	0	0	0
328	28	200436905	Clone	18-Jan-05	0	0	0	1.005	0	8.0	0.3	0	0	0
329	29	200432003	Clone	18-Jan-05	0	0	0	1.020	0	7.0	0.3	0	0	0
330	30	200437803	Clone	18-Jan-05	0	0	0	1.020	0	7.0	0.3	0	0	0
331	31	200434606	Clone	18-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
332	32	200438609	Clone	18-Jan-05	0	0	0	1.020	0	6.5	0.1	0	0	0
333	1	200438705	Control	20-Jan-05	0	0	0	1.005	0	8.0	3.0	0	0	0
334	2	200433401	Control	20-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
335	3	200431803	Control	20-Jan-05	0	0	0	1.005	0	8.0	0.3	0	0	0
336	4	200438709	Control	20-Jan-05	0	0	0	1.010	0	7.5	0.1	0	0	0
337	5	200430507	Control	20-Jan-05	0	0	0	1.020	0	7.0	1.0	0	0	0
338	6	200434805	Control	20-Jan-05	0	0	0	1.005	0	7.5	1.0	0	0	0
339	7	200435304	Control	20-Jan-05	0	0	0	1.010	0	8.0	3.0	0	0	0
340	8	200439901	Control	20-Jan-05	0	0	0	1.010	10	8.0	0.1	0	0	0
341	9	200435307	Control	20-Jan-05	0	0	0	1.005	10	8.0	0.3	0	0	0
342	10	200433208	Clone	20-Jan-05	0	0	0	1.010	10	8.0	1.0	0	0	0
343	11	200439502	Clone	20-Jan-05	0	0	0	1.030	10	6.0	1.0	0	0	0
344	12	200438608	Clone	20-Jan-05	0	0	0	1.015	0	7.5	1.0	0	0	0
345	13	200433801	Clone	20-Jan-05	0	0	0	1.020	0	7.0	0.3	0	0	0
346	14	200430801	Clone	20-Jan-05	0	0	0	1.020	0	7.0	3.0	0	0	0
347	15	200436601	Clone	20-Jan-05	0	0	0	1.005	0	8.0	0.3	0	0	0
348	16	200430602	Clone	20-Jan-05	0	0	0	1.005	0	8.0	1.0	0	0	0
349	17	200436004	Clone	20-Jan-05	0	0	0	1.010	0	7.5	0.1	0	0	0
350	18	200436903	Clone	20-Jan-05	0	0	0	1.010	0	7.5	3.0	0	0	0
351	19	200433001	Clone	20-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
352	20	200433802	Clone	20-Jan-05	0	0	0	1.025	10	7.0	3.0	0	0	0

Total Number	Slaughter Order/Day	Marc Animal ID	Treatment	Slaughter Date	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Urobilinogen	Nitrite	Leukocytes
353	21	200438611	Clone	20-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
354	22	200430702	Clone	20-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
355	23	200431602	Clone	20-Jan-05	0	0	0	1.010	0	8.0	0.3	0	0	0
356	24	200434602	Clone	20-Jan-05	0	0	0	1.010	0	8.0	1.0	0	0	0
357	25	200434109	Clone	20-Jan-05	0	0	0	1.005	0	8.0	0.3	0	0	0
358	26	200433010	Clone	20-Jan-05	0	0	0	1.015	10	8.0	0.1	0	0	0
359	27	200434104	Clone	20-Jan-05	0	0	0	1.015	0	8.0	0.3	0	0	0
360	28	200431005	Clone	20-Jan-05	0	0	0	1.010	0	8.0	0.1	0	0	0
361	29	200434107	Clone	20-Jan-05	0	0	0	1.005	0	8.0	0.1	0	0	0
362	1	200441205	Control	26-Jan-2005	0	0	0	1.030	10	6.0	0.3	0	0	0
363	2	200441211	Control	26-Jan-2005	0	0	0	1.015	0	7.5	0.3	0	0	0
364	3	200441203	Control	26-Jan-2005	0	0	0	1.030	0	6.0	0.3	0	0	0
365	4	200441201	Control	26-Jan-2005	0	0	0	1.025	0	6.5	1.0	0	0	0
366	5	200438701	Control	26-Jan-2005	0	0	0	1.010	0	7.5	0.3	0	0	0
367	6	200441209	Control	26-Jan-2005	0	0	0	1.030	0	6.0	0.3	0	0	0
368	7	200439904	Control	26-Jan-2005	0	0	0	1.010	0	7.5	1.0	0	0	0
369	8	200441207	Control	26-Jan-2005	0	0	0	1.030	0	6.0	0.3	0	0	0
370	9	200436301	Control	26-Jan-2005	0	0	0	1.010	0	7.0	0.1	0	0	0
371	10	200439204	Control	26-Jan-2005	0	0	0	1.005	0	7.5	3.0	0	0	0
372	11	200434603	Clone	26-Jan-2005	0	0	0	1.005	0	8.0	3.0	0	0	0
373	12	200434113	Clone	26-Jan-2005	0	0	0	1.015	0	8.0	20.0	0	0	0
374	13	200438606	Clone	26-Jan-2005	0	0	0	1.010	0	7.5	0.1	0	0	0
375	14	200436401	Clone	26-Jan-2005	0	0	0	1.005	0	8.0	3.0	0	0	0
376	15	200434111	Clone	26-Jan-2005	0	0	0	1.015	0	7.5	0.3	0	0	0
377	16	200438603	Clone	26-Jan-2005	0	0	0	1.200	0	7.0	0.1	0	0	0
378	17	200433011	Clone	26-Jan-2005	0	0	0	1.030	10	6.0	0.3	0	0	0
379	18	200439504	Clone	26-Jan-2005	0	0	0	1.010	0	7.5	0.3	0	0	0
380	19	200437402	Clone	26-Jan-2005	0	0	0	1.010	0	8.0	1.0	0	0	0
381	20	200439501	Clone	26-Jan-2005	0	0	0	1.010	0	7.5	0.3	0	0	0
382	21	200436008	Clone	26-Jan-2005	0	0	0	1.010	0	8.0	0.3	0	0	0
383	22	200438605	Clone	26-Jan-2005	0	0	0	1.005	0	8.0	1.0	0	0	0
384	23	200434605	Clone	26-Jan-2005	0	0	0	1.010	0	8.0	1.0	0	0	0

Total Number	Slaughter Order/Day	Marc Animal ID	Treatment	Slaughter Date	Glucose	Bilirubin	Ketone	Specific Gravity	Blood	pH	Protein	Urobilinogen	Nitrite	Leukocytes
385	24	200436003	Clone	26-Jan-2005	0	0	0	1.005	0	8.0	3.0	0	0	0
386	25	200437509	Clone	26-Jan-2005	0	0	0	1.010	200	8.0	1.0	0	0	0
387	26	200432506	Clone	26-Jan-2005	0	0	0	1.015	0	8.0	0.3	0	0	0
388	27	200433204	Clone	26-Jan-2005	0	0	0	1.020	0	8.0	1.0	0	0	0
389	28	200436612	Clone	26-Jan-2005	0	0	0	1.010	0	8.0	1.0	0	0	0
390	1	200438703	Control	2-Feb-2005	0	0	0	1.020	0	7.0	0.1	0	0	0
391	2	200439401	Control	2-Feb-2005	0	0	0	1.030	0	6.0	1.0	0	0	0
392	3	200431804	Control	2-Feb-2005	0	0	0	1.025	0	7.0	3.0	0	0	0
393	4	200439404	Control	2-Feb-2005	0	0	0	1.030	0	6.5	3.0	0	0	0
394	5	200438403	Control	2-Feb-2005	0	0	0	1.010	0	8.0	0.1	0	0	0
395	6	200439503	Clone	2-Feb-2005	0	0	0	1.010	0	8.0	0.1	0	0	0
396	7	200434102	Clone	2-Feb-2005	0	0	0	1.010	0	8.0	0.1	0	0	0
397	8	200430603	Clone	2-Feb-2005	0	0	0	1.005	0	8.5	1.0	0	0	0
398	9	200434604	Clone	2-Feb-2005	0	0	0	1.025	0	6.5	0.3	0	0	0
399	10	200438604	Clone	2-Feb-2005	0	0	0	1.005	0	8.0	1.0	0	0	0
400	11	200434105	Clone	2-Feb-2005	0	0	0	1.010	0	8.0	0.1	0	0	0
401	12	200437401	Clone	2-Feb-2005	0	0	0	1.015	0	7.5	0.1	0	0	0
402	13	200432306	Clone	2-Feb-2005	0	0	0	1.015	0	7.5	0.3	0	0	0
403	14	200433708	Clone	2-Feb-2005	0	0	0	1.005	0	8.5	0.3	0	0	0
404	15	200438102	Clone	2-Feb-2005	0	0	0	1.010	0	8.0	0.3	0	0	0
405	16	200436403	Clone	2-Feb-2005	0	0	0	1.010	0	8.0	1.0	0	0	0

Appendix G:
Unpublished Data

Appendix G: Unpublished Data

In July 2001, the Center issued a CVM Update requesting that clone producers not introduce meat or milk from clones or their progeny into food or feed until the Agency had time to complete a review of the safety of those food products. At that time, CVM also requested that clone producers submit data to the Center on the health of clones and their progeny, and on meat and milk composition of clones and progeny. Two large datasets were submitted by two companies: Cyagra, Inc. and ViaGen, Inc. Reviews of these data are provided in Appendices E and F, respectively. In addition, several small datasets were submitted to the Center, and included tables and reports of veterinarians and technicians for neonatal cattle and swine, as well as data on juvenile development, reproductive function and maturity in cattle. These data were originally included with Chapter V in the release of the Draft Risk Assessment on December 28, 2006, and are now presented in their entirety in this Appendix and reviewed below. One additional dataset has also been submitted by J.L. Edwards from the University of Tennessee, who also provided additional information in a teleconference on September 12, 2007.

A. Perinatal Development in Cattle Clones (Node 2)

Body temperature, pulse and respiration rate data were submitted covering the first 72 hours of life for 19 clone calves of unknown breed(s) from a commercial cloning company (Table G-1).

Mean \pm SD for body temperature, pulse and respiration at birth for the 19 calves were 103.0 ± 1.2 ° F, 95.2 ± 30.34 beats/min, and 53.9 ± 19.4 breaths/min, respectively. At 24 hours, means were 101.8 ± 0.7 ° F, 133.1 ± 19.2 beats/min, and 57.4 ± 21.1 breaths/min. At 48 hours, means were 102.0 ± 0.5 ° F, 135.8 ± 17.9 beats/min, and 56.1 ± 24.8 breaths/min. Values for one calf were not available for the 72 hour measurements, such that means and standard deviations represent 18 calves. Those values were 102.7 ± 0.7 ° F, 138.6 ± 19.3 beats/min, and 53.1 ± 20.9 breaths/min. Heart and respiration rates vary with age. Respiration rates in growing steers (age not specified) were noted to be 79 ± 3.2 breaths/min in one study (Nihsen et al. 2004).

Breukelman et al. (2004) noted basal fetal heart rates in late gestation AI pregnancies to be 111.6 ± 1.4 beats/min. By comparison, basal heart rate of three week old heifer calves averaged 88.1 ± 4.04 beats/min (Van Reenen et al. 2005).

Calf ID	Calving Date	Temperature (F)				Pulse				Respiration			
		Birth	24h	48h	72h	Birth	24h	48h	72h	Birth	24h	48h	72h
1	2/12/2001	104.8	103	102.2	102.6	54	132	144	116	48	44	48	84
2	3/28/2001	101.2	101.7	101.7	101.8	120	114	138	138	36	42	42	36
3	4/10/2001	103	101.6	101.4	103.6	100	120	126	140	46	44	44	48
4	4/12/2001	104.3	101.2	101.6	102.6	64	120	132	140	30	48	44	36
5	4/13/2001	102.6	101.1	102	103.6	100	120	120	140	44	56	48	48
6	4/10/2001	102.9	101.5	102.3	102.5	116	144	128	126	68	54	56	39
7	4/11/2001	100.5	101.1	101.7	102.9	112	120	160	152	54	48	40	40
8	1/15/2002	102.4	102	101.8	102	60	144	140	128	60	48	28	20
9	1/30/2002	103.2	101.3	102	101.1	128	136	140	132	98	56	52	44
10	1/31/2002	105.2	101.5	101.6	102.2	150	140	140	115	36	32	24	40
11	1/29/2002	102.4	102	102.5	102.6	66	132	126	138	66	108	102	78
12	3/27/2002	102.5	102.5	102	102.4	60	192	104	156	60	54	40	56
13	3/21/2002	103.4	100.8	101.3	103	108	108	132	156	72	36	30	24
14	4/9/2002	103.9	101.3	102.1	102.9	40	114	120	120	24	80	84	72
15	4/4/2002	104	102.2	102.2		90	132	180		24	56	72	
16	5/1/2002	103.4	101.3	102.4	103	90	120	120	102	50	48	68	68
17	4/30/2002	101.3	103.2	101.5	102	120	140	120	180	78	88	64	66
18	11/11/2002	103.3	103.1	103.2	103.4	100	150	150	160	70	100	120	60
19	11/12/2002	102	101.6	101.8	103.8	130	150	160	156	60	48	60	96

The birth records of two Holstein heifer clones were submitted by a private cloning firm. Both heifers were delivered by C-section. One calf was a breech position (posterior presentation with hind limbs under body); the other calf was in a normal posterior position, with hind limbs extended. The placentae of these calves were described as normal, with some large and some small placentomes described for one placenta. Calves were described as normal, weighing 45 and 47.7 kg each at time of delivery. Both calves had normal umbilici. Some fluid was noted in the lungs of both calves, but they were described as breathing normally, and although some meconium staining was noted, there was no indication that meconium had been inhaled. Body temperatures were 100 and 102.6°F, slightly below and above the average temperature for cattle. Blood glucose, packed cell volume (PCV), blood total protein, and IgG concentrations were monitored for the first 23 to 27 hours after birth (Table G-2). Blood glucose was low for both calves prior to first feeding, then increased to normal levels by the second feeding. Total protein also increased steadily following feeding, and IgG levels were listed as “> 10” after the first colostrum feeding. The units for IgG measurements were not provided, and PCV values were the

only hematology data provided, so these data are difficult to interpret. Total protein and serum glucose values are comparable to age-matched non-clone cattle in the Cyagra dataset by the second post-feeding blood sample.

	Calf 1			Calf 2		
	Pre-feeding	1 st feeding	2 nd feeding	Pre-feeding	1 st feeding	2 nd feeding
Glucose	20	35	93	21	29	83
PCV	29	28	25	30	33	29
Total Protein	5	5.3	7.5	4.8	5.0	6.0
IgG	N/A	>10	>10	N/A	>10	>10

B. Juvenile Development in Cattle Clones (Node 3)

A private veterinary clinic submitted hematology and clinical chemistry data on three bull clones, ranging in age from 5 to 7 months at the time of the first sampling. Animals were sampled a total of three times within a six week period. Most variables measured were within the reference range used by the diagnostic lab conducting the tests (Marshfield Laboratories, Marshfield, WI). Values outside the reference range for the testing laboratory are listed in Tables G-3 a, b, and c.

CVM contacted Marshfield Laboratories on September 21, 2005, regarding the source of their reference range. According to the laboratory, the reference range for hematology and clinical chemistry was established on blood samples taken from female dairy animals between 1 and 8 years of age. As discussed in Appendix E, it is important that the selected reference range is appropriate to the animals being tested. In this case, the use of a reference range established using post-pubertal, near-adult and adult females may not provide an appropriate comparison for pre-pubertal, rapidly growing males. As no contemporary comparator animals were sampled, other published reference ranges (Meyer and Harvey 2004; Green 1998; Duncan and Prasse 2003) were used for cross-comparison. Results indicated that one clone (Clone #3) on the second sampling date had one analyte that was outside any of the reference ranges used. For Clone #3, cholesterol was low on October 13. However, all of this animal's hematology and clinical chemistry values were considered within published ranges on the third sampling date, approximately two weeks later. Because serum cholesterol can be affected by diet and time since the last meal, the single low value for this animal was judged not to be biologically relevant.

Table G-3a: Hematology and Clinical Chemistry for Three Holstein Bulls (Bull #1)									
							RMI N	RMAX	Units
Date Section 1 Collected		9/22/03		10/13/03		10/27/03			
Hemogram-Vet (VCLT)									
Red Blood Cell Count	.	7.03					5	10	x 10 ⁶ /uL
Hemoglobin	.	9.10					8	15	g/dL
Hematocrit	.	29.00					24	46	%
Mean Corpuscular Volume	.	41.20					40	60	fL
Mean Corpuscular Hemoglobin	.	12.90					11	17	pg
Mean Corpuscular Hgb Conc.	.	31.40					30	36	g/dL
Red Cell Distribution Width	L	21.60					26	30	%
Platelet Count	H	720.00					230	690	x 10 ³ /uL
White Blood Cell Count	.	5.30					4	12	x 10 ³ /uL
Seg. Neutrophil Absolute #	.	1.64					0.6	4	x 10 ³ /uL
Banded Neutrophil Absolute #	.	0.00					0	0.12	x 10 ³ /uL
Lymphocyte Absolute #	.	3.39					2.5	7.5	x 10 ³ /uL
Act Lymphocyte Absolute #	.	0.00							x 10 ³ /uL
Monocyte Absolute #	.	0.16					0.03	0.84	x 10 ³ /uL
Eosinophil Absolute #	.	0.11					0	2.4	x 10 ³ /uL
Basophil Absolute #	.	0.00					0	0.2	x 10 ³ /uL
Other Absolute #	.	0.00					0	0	x 10 ³ /uL
Blast Absolute #		0.00							x 10 ³ /uL
Promyelocyte Absolute #		0.00							x 10 ³ /uL
Myelocyte Absolute #		0.00							x 10 ³ /uL
Metamyelocyte Absolute #		0.00							x 10 ³ /uL
Differential, Vet. (VDIF)									
Segmented Neutrophils		31							%
Lymphocytes		64							%
Monocytes		3							%
Eosinophils		2							%
Basophils									%

Table G-3a: Hematology and Clinical Chemistry for Three Holstein Bulls (Bull #1)										
							RMI N	RMAX	Units	
Date Section 1 Collected			9/22/03		10/13/03		10/27/03			
	Poikilocytosis / polychromasia?		no							
	Glucose	L	47.0	.	78.0	H	87.0	55	79	mg/dL
	AST (GOT)	.	79.0	L	56.0	.	69.0	57	108	U/L
	SDH	.	12.8	.	13.3	.	15.8	12.2	46	U/L
	Total Bilirubin	.	0.1	.	0.1	.	0.1	0.1	0.4	mg/dL
	Cholesterol	L	95.0	L	90.0	L	95.0	112	331	mg/dL
	Total Protein	.	6.8	.	7.6	.	7.2	6.3	8.5	g/dL
	Albumin	.	3.4	.	3.5	.	3.5	3.2	4.3	g/dL
	Urea N	.	12.0	L	7.0	.	8.0	8	22	mg/dL
	Creatinine	L	0.5	.	0.6	L	0.5	0.6	1.4	mg/dL
	Phosphorous	.	9.0	.	8.3	.	8.6	4.4	9.2	mg/dL
	Calcium	.	10.5	.	10.1	.	10.5	7.9	10.5	mg/dL
	Sodium	.	143.0	.	150.0	.	145.0	140	151	mmol/L
	Potassium	.	5.2	.	4.8	.	5.1	3.7	5.6	mmol/L
	Chloride	.	100.0	H	110.0	.	102.0	100	109	mmol/L
	Bicarbonate	.	25.0	.	25.0	.	29.0	22	29	mmol/L
	CK	.	221.0	.	190.0	.	157.0	50	271	U/L
	GGT	.	19.0	.	14.0	.	13.0	12	30	U/L
	Anion Gap	H	23.0	.	20.0	.	19.0	13.6	21.6	mmol/L
	Hemolysis/lipemia?		no		no		no			

Table G-3b: Hematology and Clinical Chemistry for Three Holstein Bulls (Bull #1)										
							RMI N	RMAX	Units	
Date Section 1 Collected			9/22/03		10/13/03		10/27/03			
Hemogram-Vet (VCLT)										
	Red Blood Cell Count	.	7.83				5	10		x 10 ⁶ /uL
	Hemoglobin	.	9.90				8	15		g/dL
	Hematocrit	.	31.20				24	46		%
	Mean Corpuscular Volume	L	39.80				40	60		fL
	Mean Corpuscular Hemoglobin	.	12.60				11	17		pg
	Mean Corpuscular Hgb Conc.	.	31.70				30	36		g/dL

Table G-3b: Hematology and Clinical Chemistry for Three Holstein Bulls (Bull #1)									
							RMI N	RMAX	Units
Date Section 1 Collected		9/22/03		10/13/03		10/27/03			
Red Cell Distribution Width	L	21.60					26	30	%
Platelet Count	H	769.00					230	690	x 10 ³ /uL
White Blood Cell Count	.	10.10					4	12	x 10 ³ /uL
Seg. Neutrophil Absolute #	.	2.63					0.6	4	x 10 ³ /uL
Banded Neutrophil Absolute #	.	0.00					0	0.12	x 10 ³ /uL
Lymphocyte Absolute #	.	6.87					2.5	7.5	x 10 ³ /uL
Act Lymphocyte Absolute #	.	0.00							x 10 ³ /uL
Monocyte Absolute #	.	0.20					0.03	0.84	x 10 ³ /uL
Eosinophil Absolute #	.	0.10					0	2.4	x 10 ³ /uL
Basophil Absolute #	H	0.30					0	0.2	x 10 ³ /uL
Other Absolute #	.	0.00					0	0	x 10 ³ /uL
Blast Absolute #		0.00							x 10 ³ /uL
Promyelocyte Abs. #		0.00							x 10 ³ /uL
Myelocyte Absolute #		0.00							x 10 ³ /uL
Metamyelocyte Abs. #		0.00							x 10 ³ /uL
Differential, Vet. (VDIF)									
Segmented Neutrophils		26							%
Lymphocytes		68							%
Monocytes		2							%
Eosinophils		1							%
Basophils		3							%
Poikilocytosis / polychromasia?		no							
Glucose	L	4.0	.	76.0	H	88.0	55	79	mg/dL
AST (GOT)	.	71.0	L	46.0	.	69.0	57	108	U/L
SDH	L	11.2	L	7.3	.	16.7	12.2	46	U/L
Total Bilirubin	.	0.2	.	0.1	.	0.1	0.1	0.4	mg/dL
Cholesterol	L	88.0	L	83.0	L	86.0	112	331	mg/dL
Total Protein	.	7.5	.	7.8	.	7.6	6.3	8.5	g/dL
Albumin	.	3.7	.	3.4	.	3.6	3.2	4.3	g/dL
Urea N	.	10.0	.	8.0	.	13.0	8	22	mg/dL
Creatinine	L	0.5	L	0.5	.	0.6	0.6	1.4	mg/dL
Phosphorous	.	7.8	.	7.3	.	8.4	4.4	9.2	mg/dL

Table G-3b: Hematology and Clinical Chemistry for Three Holstein Bulls (Bull #1)									
							RMI N	RMAX	Units
Date Section 1 Collected		9/22/03		10/13/03		10/27/03			
Calcium	H	10.6	.	9.7	.	9.5	7.9	10.5	mg/dL
Sodium	.	142.0	.	144.0	.	145.0	140	151	mmol/L
Potassium	.	5.3	.	4.6	.	4.4	3.7	5.6	mmol/L
Chloride	.	101.0	.	104.0	.	103.0	100	109	mmol/L
Bicarbonate	.	24.0	.	27.0	.	28.0	22	29	mmol/L
CK	.	234.0	.	172.0	.	179.0	50	271	U/L
GGT	.	14.0	.	13.0	.	15.0	12	30	U/L
Anion Gap	H	22.0	.	18.0	.	18.0	13.6	21.6	mmol/L
Hemolysis / lipemia?		no		no		no			

Table G-3c: Hematology and Clinical Chemistry for Three Holstein Bulls (Bull #1)									
							RMI N	RMAX	Units
Date Section 1 Collected		9/22/03		10/13/03		10/27/03			
Hemogram-Vet (VCLT)									
Red Blood Cell Count	.	7.41					5	10	$\times 10^6/\mu\text{L}$
Hemoglobin	.	10.10					8	15	g/dL
Hematocrit	.	31.10					24	46	%
Mean Corpuscular Volume	.	41.90					40	60	fL
Mean Corpuscular Hemoglobin	.	13.70					11	17	pg
Mean Corpuscular Hgb Conc.	.	32.60					30	36	g/dL
Red Cell Distribution Width	L	21.90					26	30	%
Platelet Count	.	461.00					230	690	$\times 10^3/\mu\text{L}$
White Blood Cell Count	.	4.80					4	12	$\times 10^3/\mu\text{L}$
Seg. Neutrophil Absolute #	.	1.87					0.6	4	$\times 10^3/\mu\text{L}$
Banded Neutrophil Absolute #	.	0.00					0	0.12	$\times 10^3/\mu\text{L}$
Lymphocyte Absolute #	.	2.59					2.5	7.5	$\times 10^3/\mu\text{L}$
Act Lymphocyte Abs. #	.	0.00							$\times 10^3/\mu\text{L}$
Monocyte Absolute #	.	0.19					0.03	0.84	$\times 10^3/\mu\text{L}$
Eosinophil Absolute #	.	0.10					0	2.4	$\times 10^3/\mu\text{L}$
Basophil Absolute #	.	0.05					0	0.2	$\times 10^3/\mu\text{L}$

Table G-3c: Hematology and Clinical Chemistry for Three Holstein Bulls (Bull #1)									
							RMI N	RMAX	Units
Date Section 1 Collected		9/22/03		10/13/03		10/27/03			
Other Absolute #	.	0.00					0	0	x 10 ³ /uL
Blast Absolute #		0.00							x 10 ³ /uL
Promyelocyte Abs. #		0.00							x 10 ³ /uL
Myelocyte Absolute #		0.00							x 10 ³ /uL
Metamyelocyte Abs. #		0.00							x 10 ³ /uL
Differential, Vet. (VDIF)									
Segmented Neutrophils		39							%
Lymphocytes		54							%
Monocytes		4							%
Eosinophils		2							%
Basophils		1							%
Poikilocytosis / polychromasia?		no							
Glucose	L	52.0	.	76.0	H	84.0	55	79	mg/dL
AST (GOT)	.	72.0	L	53.0	.	81.0	57	108	U/L
SDH	L	9.7	L	10.1	.	18.6	12.2	46	U/L
Total Bilirubin	.	0.2	.	0.1	.	0.2	0.1	0.4	mg/dL
Cholesterol	L	85.0	L	80.0	L	87.0	112	331	mg/dL
Total Protein	.	6.6	.	7.4	.	7.4	6.3	8.5	g/dL
Albumin	.	3.6	.	3.4	.	3.3	3.2	4.3	g/dL
Urea N	.	10.0	.	8.0	.	8.0	8	22	mg/dL
Creatinine	L	0.5	.	0.6	L	0.5	0.6	1.4	mg/dL
Phosphorous	.	8.8	.	8.1	.	9.0	4.4	9.2	mg/dL
Calcium	H	10.7	.	9.7	.	10.1	7.9	10.5	mg/dL
Sodium	.	143.0	.	145.0	.	144.0	140	151	mmol/L
Potassium	.	4.9	.	4.9	.	5.1	3.7	5.6	mmol/L
Chloride	L	99.0	.	105.0	.	101.0	100	109	mmol/L
Bicarbonate	.	26.0	.	26.0	.	29.0	22	29	mmol/L
CK	.	179.0	.	134.0	.	172.0	50	271	U/L
GGT	.	15.0	.	16.0	.	17.0	12	30	U/L
Anion Gap	H	23.0	.	19.0	.	19.0	13.6	21.6	mmol/L
Hemolysis / lipemia?		no		no		no			

C. Reproductive Development and Function in Cattle Clones (Node 4)

In response to CVM's request for additional data on reproductive maturity of clones, results were submitted on semen evaluations of four post-pubertal bull clones. Semen was collected by a commercial reproduction service, from May 15, 2003 through June 19, 2003. Age of the bulls at time of sampling was not recorded. Bulls were collected three times daily, approximately every three days during the observation period. Data consists of hand-written notes provided by the technician, and includes information on semen volume, concentration, and percentage of normal sperm in samples. Sperm motility was not presented in these reports. Percent normal sperm was not assessed in all samples. A summary of the results (means, minimum and maximums) is in Table G-4. The complete table (Table G-10) is presented at the end of this Appendix. Because the original data was sent as a fax and difficult to read, some of the hand written notes may not be accurately transcribed. CVM attempted but was unable to establish contact with the service to clarify these notes.

Clone #	Volume (ml)			Concentration (x10 ⁶)			Normal Sperm (%)		
	mean	max	min	mean	max	min	mean	max	min
1	4.1	4.9	3.0	169.5	100	276	5.0	8.0	2.0
2	3.8	6.5	3.0	686.7	1870	307	51.0	71.0	25.0
3	5.0	7.9	3.5	712.1	1581	396	69.5	76.0	62.0
4	4.6	6.9	3.0	730	1649	73	63.9	80.0	54.0

Reference ranges differ somewhat, but in general normal ranges for ejaculate volume are from 4 to 15 mL, sperm concentrations from 800 to 1200 x 10⁶ sperm/mL, and percent normal sperm range from 65 to 95 percent for bulls (Sorenson 1979; Beardon and Fuquay 1980; Hafez and Hafez 2000). Based on these data, unless Clone #1 was very young, he likely would have failed a breeding soundness exam, due to the very low concentration and percentage of normal sperm in the samples. Clone #2 might be considered marginal, and depending on other, unrecorded variables such as motility, and the perceived value of his genetics might have been judged acceptable. The other two clones appear to have acceptable semen, based on the limited data presented.

In 2003, Galli et al. (unpublished) presented data to CVM on three clones of a Holstein bull as a follow-up to their 2002 study on cloning (Table G-5). Scrotal circumferences of two of the clones (Clones 1 and 2) were similar to the expected range for bulls 18-24 months old (31 and 33 cm, respectively, at 22 months old vs. 32-33 cm for 18 to 24 month old bulls). Semen quality measurements on two of the clones (Clones 2 and 3) were also considered within the normal

range for young bulls, although only data on volume (5.27 and 3.35 ml) and sperm concentration (691 and 736 million/ml) were presented.

	Scrotal Circumference (cm)	Collections (#) ¹	Ejaculates (#)	Average volume (ml)	Concentration (x10 ⁶ /ml)
Clone 1	31				
Clone 2	33	17	32	5.27	691
Clone 3		11	19	3.35	736

¹ Semen was collected at irregular intervals

Semen with ≥ 50 percent motility were frozen and thawed. Post thaw motility averaged > 40 percent. Semen from Clone # 2 resulted in a 75 percent *in vitro* fertilization rate. Semen from this clone was also used to test AI pregnancy rates on four farms. The total number of cows bred (n = 63) was small, and no contemporaneous comparator was used, so the value of this data is limited, and effects of individual farm management cannot be assessed. With these caveats in mind, the results of these tests by farm are presented in Table G-6.

Farm	Cows bred	Cows pregnant	Pregnancies lost	Pregnancy rate %	Loss rate %
1	30*	22	2	73	9
2	20	10	0	50	0
3	3	1	0	33	0
4	10	8	0	80	0
total	63	41	2	65	5

*Results for two rounds of insemination were presented. It is not clear whether any of the individual cows were bred twice.

Pregnancy rates to AI in cattle vary considerably, and are affected by multiple factors, such as the ability of farm personnel to detect cows in heat, appropriate timing of insemination relative to the onset of heat, and environmental, production and nutritional factors. Studies in U.S. dairy cattle indicate that overall pregnancy rates to first AI are 40 percent or less (Lucy 2001; El-Zarkouny et al. 2004). Given the small number of cows in this study, and the lack of a contemporaneous comparator to assess the influence of farm, definitive conclusions are not possible. However, overall pregnancy rates to this bull clone do not appear worse than the U.S. average.

D. Post-pubertal Maturation in Cattle Clones (Node 5)

In response to CVM's request for data concerning this developmental node, data on two heifer clones, approximately 14 months old, was submitted to CVM (see Table G-7).

These data consist of Certificates of Veterinary Inspection, results of serological testing for Bovine Leucosis Virus (BLV), Bovine Viral Diarrhea (BVD), and hematology from the state of Wisconsin. Both heifers tested negative for BLV and BVD. According to the hematology report, both heifers had red cell distribution widths (RDW) slightly below the reference range used by the testing laboratory (22.4 and 24.0 vs. range of 26.0-30.0 percent). As discussed in Appendix E for Cyagra clones, RDW is only indicative of a health problem (anemia) when coupled with primary indicators such as low red blood cell count (RBC), hemoglobin and/or hematocrit. As all other hematology values were within the reference range, there is no evidence to indicate an underlying health problem in these animals.

Table G-7: Hematology and Clinical Chemistry for Two Holstein Heifer Clones								
			Heifer #1		Heifer #2	RMIN	RMAX	Units
Date Collected			12/10/2000		12/11/2001			
Hemogram-Vet								
Red Blood Cell Count	.		7.43	.	7.81	5	10	x 10 ⁶ /uL
Hemoglobin	.		11	.	11.7	8	15	g/dL
Hematocrit	.		30.6	.	32.7	24	46	%
Mean Corpuscular Volume	.		41.2	.	41.8	40	60	fL
Mean Corpuscular Hemoglobin	.		14.8	.	15	11	17	pg
Mean Corpuscular Hgb Conc.	.		35.9	.	35.9	30	36	g/dL
Red Cell Distribution Width	L		22.4	L	24	26	30	%
Platelet Count	.		449	.	322	230	690	x 10 ³ /uL
White Blood Cell Count	.		7.1	.	7.2	4	12	x 10 ³ /uL
Seg. Neutrophil Absolute #	.		1.99	.	1.66	0.6	4	x 10 ³ /uL
Banded Neutrophil Absolute #	.		0	.	0	0	0.12	x 10 ³ /uL
Lymphocyte Absolute #	.		3.91	.	4.39	2.5	7.5	x 10 ³ /uL
Act Lymphocyte Absolute #	.		0	.	0			x 10 ³ /uL
Monocyte Absolute #	.		0.78	.	0.72	0.03	0.84	x 10 ³ /uL
Eosinophil Absolute #	.		0.43	.	0.36	0	2.4	x 10 ³ /uL
Basophil Absolute #	.		0	.	0.07	0	0.2	x 10 ³ /uL

Table G-7: Hematology and Clinical Chemistry for Two Holstein Heifer Clones							
		Heifer #1		Heifer #2	RMIN	RMAX	Units
Date Collected		12/10/2000		12/11/2001			
Other Absolute #	.	0	.	0	0	0	x 10 ³ /uL
Blast Absolute #		0		0			x 10 ³ /uL
Promyelocyte Absolute #		0		0			x 10 ³ /uL
Myelocyte Absolute #		0		0			x 10 ³ /uL
Metamyelocyte Absolute #s		0		0			x 10 ³ /uL
Differential, Vet.							
Segmented Neutrophils		28		23			%
Lymphocytes		55		61			%
Monocytes		11		10			%
Eosinophils		6		5			%
Basophils				1			%
Morphology		*		*			%
Poikilocytosis / polychromasia?		no		no			

E. Summary of Cloning Studies at University of Tennessee

The original table was submitted to CVM and is presented at the end of this Appendix. The table presents data for all developmental nodes for Jersey cattle clones, including data on pregnancy losses and outcomes for surrogates. Data for progeny of clones was not included. The table represents outcomes of 46 third-trimester pregnancies of female Jersey cattle clones identified by cell line.

1. Developmental Node 1: Pregnancy and Parturition

The summary tables from the UT study indicate a total of 46 third-trimester pregnancies. Seventeen pregnancies terminated between 174 and 268 days gestation resulting from spontaneous abortion, premature delivery, or euthanasia of the surrogate dam due to advancing hydrops. In all, 27 surrogate dams experienced hydrops (27/46 or 59 percent of dams) in this cohort. This is a higher incidence of this condition than reported in previous studies or other data directly submitted to the Agency. For 10 of the 27 hydropic surrogate dams (37 percent) the condition was severe enough to require euthanasia. Twenty-five surrogate dams delivered by C-section, and an additional five cows required assistance with vaginal delivery. According to Dr. Edwards many of the C-sections were planned, due to the high incidence of hydrops.

2. Developmental Node 2: Perinatal Period

Twenty-seven calves were born alive in the UT study, of which 19 survived the critical first seven days post-partum (70 percent). Causes of perinatal death are listed in the table provided by UT, but include placental, skeletal, respiratory, renal, hepatic, and cardiac defects and lesions; aspiration of amniotic fluid; and hydrocephalus. One calf was euthanized at seven days of age. The only finding listed for this animal was blindness.

Average birth weight of these Jersey heifer clones was 31.62 kg, and ranged from a very small and non-viable calf (6.36 kg) to 45 kg. Average birth weight for conventionally bred Jersey heifers is 23-24 kg (Bonczek et al. 1992).

3. Developmental Node 3: Juvenile Period

Of the 19 calves that survived the perinatal period, 12 lived to one year of age (63 percent). One of the seven animals that died was discussed in Lawrence et al. (2005), and reviewed in Chapter V. This calf died at nine months of age from enterotoxemia due to infection with Type A *Clostridium perfringens*. The other six calves died between the ages of 41 days to eight months. Four of these calves experienced some type of gastro-intestinal problem (scouring, rumen stasis, acidosis, anorexia), with other complications (pneumonia, umbilical abscess, hepatic defects) and severe weight loss. Common pathogens such as *Mycobacterium paratuberculosis avium* and Bovine Viral Diarrhea were ruled out as potential causes of this “wasting” (Edwards teleconference). The remaining two calves were both diagnosed on necropsy with hepatic defects.

4. Developmental Node 4: Reproductive Development and Function

Information on reproductive function from the UT study is limited, and only for five females which were later euthanized for the purpose of herd reduction and one female euthanized following post partum complications (retained fetal membranes, metritis). Of these six animals, two delivered “normal” calves, but no other information on these calves (including gender) was included. The other four clones delivered stillborn calves following apparently normal pregnancies.

5. Developmental Node 5: Post Pubertal Maturation and Aging

Of the 12 calves in the UT study surviving the juvenile period, one died at 1 year of age. The heifer was noted to experience periodic scours that were unresponsive to treatment. Another heifer was euthanized at 2.5 years of age following delivery of a stillborn calf. This clone suffered complications due to retained fetal membranes (metritis) and was also noted to have had periodic scours unresponsive to treatment. According to Dr. Edwards, this clone recovered from the postpartum complications, but was euthanized due to scouring and severe weight loss. As mentioned above, five clones were euthanized at four years of age for the purposes of herd reduction. These five euthanized cows were identified as healthy at time of euthanasia. All five were noted to suckle other animals, as was the clone euthanized at 2.5 years of age. This behavior is sometimes noted in adult cattle, and may reflect how the clones were raised and weaned. The seven remaining clones are four years old, and the table indicates that their health is “within normal limits”. According to Dr. Edwards, the seven remaining clones do not exhibit the aberrant suckling behavior. She attributed this to differences in how the calves were raised. The earlier group, which developed the aberrant behavior were only offered a bottle to test suckle reflex, and thereafter were fed by bucket. The latter group, from which the seven living clones derive, were fed by bottle until weaning. One of these adult clones apparently had patent urachus at one time, but is currently listed as “within normal limits”. According to Dr. Edwards, the attending veterinarian noted unusually large vessels in the urachus. Another of these living clones was noted to have had an enlarged umbilicus that was surgically corrected, and is otherwise considered “within normal limits”. The seven remaining clones were not bred, due to the voluntary moratorium on introducing meat or milk from clones into the food chain.

F. Perinatal Development in Swine Clones (Node 2)

Additional data was submitted by a commercial cloning company on birth weight and average daily gain (ADG) during the first three months of life as well as body temperature and heart rate of pigs during the first 2 days after birth. Breed of pigs was not identified. Data on body temperature and heart rate was available for five pigs, while birth weight and ADG were available for three of the five animals. According to the information submitted, two of the five piglets, both from the same litter and weighing 1.0 kg at birth, died within 48 hours of birth. The cause of death was not provided. The five pigs were born in two litters. The data are presented in Tables G-8 and G-9.

Litter #	Piglet ID #	Birth date	BW (kg)	Age @ weighing	Period	Weight (Kg)	ADG2 (KG/day)
1	1	5/24/2002	1.2	18	17	2.7	0.06
1	1	5/24/2002	1.2	40	22	10	0.33
1	1	5/24/2002	1.2	60	20	23.3	0.67
1	1	5/24/2002	1.2	74	14	31.6	0.59
1	1	5/24/2002	1.2	94	20	50	0.92
1	2	5/24/2002	1.4	18	17	2.8	0.08
1	2	5/24/2002	1.4	40	22	8.6	0.26
1	2	5/24/2002	1.4	60	20	18.9	0.52
1	2	5/24/2002	1.4	74	14	25.8	0.49
1	2	5/24/2002	1.4	94	20	44.2	0.92
2	4	5/27/2002	1.1	15	17	3.2	0.15
2	4	5/27/2002	1.1	37	22	10.9	0.35
2	4	5/27/2002	1.1	57	20	24.6	0.69
2	4	5/27/2002	1.1	71	14	33.8	0.66
2	4	5/27/2002	1.1	91	20	51.5	0.89

Litter #	Pig ID #	Respiration Rate					Heart Rate					Blood Temperature				
		D1	D2	D3	D4	D5	D1	D2	D3	D4	D5	D1	D2	D3	D4	D5
1	1	48.3 +/- 3.8	36 +/- 2.3	37.9 +/- 2.1	38.9 +/- 1.7	41.3 +/- 2.8	188.6 +/- 14.4	159.8 +/- 13.8	167.4 +/- 8.1	160 +/- 6.1	176.0 +/- 5.6	99.3 +/- 1.2	101.5 +/- 0.2	101. 9 +/- 0.3	101.2 +/- 0.4	101.3 +/- 0.3
1	2	51.8 +/- 5.7	39.1 +/- 2.2	37.3 +/- 1.7	43.7 +/- 3.7	39.5 +/- 1.8	202.3 +/- 7.3	148.6 +/- 14.6	162.3 +/- 5.9	176.7 +/- 6.7	157.4 +/- 4.8	99.2 +/- 1.0	101.8 +/- 0.4	102. 4 +/- 0.2	100.8 +/- 0.3	101.3 +/- 0.3
2	3	64.3 +/- 9.8	36.9 +/- 8.3	NA	NA	NA	162.3 +/- 10.6	173.5 +/- 2.3	NA	NA	NA	100.6 +/- 0.5	101.1 +/- 0.2	NA	NA	NA
2	4	43 +/- 3.0	34.9 +/- 1.2	NA	NA	NA	206.4 +/- 13.8	169.0 +/- 5.8	NA	NA	NA	101.0 +/- 0.5	101.1 +/- 0.3	NA	NA	NA
2	5	59.8 +/- 5.9	42.2 +/- 3.2	NA	NA	NA	172.6 +/- 13.9	180.4 +/- 6.1	NA	NA	NA	98.8 +/- 0.5	101.5 +/- 0.3	NA	NA	NA

Note: Pigs 3, 4, and 5 were from the same litter. Pigs 1 and 2 were from another litter. Pigs 3 and 5 died shortly after birth. The absence of data for pig 4 was not explained.

Birth weight and ADG vary depending on breed of swine. The breed of swine in this dataset was not reported, making interpretation of these data difficult. Likewise, interpreting respiration and heart rates in animals not typically handled is problematic, since the stress of handling tends to increase respiration and heart rates. Body temperatures of the five clones during the first two days are somewhat low; however, as noted earlier in this chapter, neonatal swine generally need supplemental heat because they lack the ability to thermoregulate. For growth, the available reference values for non-clone comparator swine and their progeny presented in the ViaGen dataset (Appendix F) is instructive. Growth rates in this dataset and the ViaGen dataset for non-clone comparators are similar. Average heart rate of day old conventional pigs was reported as 190.75 ± 36.45 bpm in one study (Foster et al. 2001).

G. Conclusions for unpublished data

Although limited in content and scope, these small datasets are consistent with the Cyagra and ViaGen datasets and with the published literature on cattle and swine clones. When considered in context with other data sources and compared to data from reference texts for conventionally bred animals, these data support the overall conclusions that cattle and swine clones generally fall within established ranges for clinical measures of health (heart rate, respiration, body

temperature, and hematology), and do not present any unique health issues. The poor semen quality of two bull clones in one small cohort cannot be explained given the limited information available on these animals. It is not unusual for bulls to fail reproductive soundness exams, and such failure may result from injury, infection, or environmental stress, among other reasons.

Sperm Characteristics of Four Clones of a Crossbred Beef Bull

Nuclear Donor (crossbred beef bull) Collection	Date	Vol (ml)	Conc X108	No. Ejac.	Dose	Exp. Yield	A Dilute	Filled	ACL	Inc	Norm	Abnormalities %				Results	
												Head	Droplet	Tail	Ph		
Clone #5	05/16/03	5.6	1253	1	35	200	112.2	198		30/	MDR's N 30%						
Clone #5	05/16/03	6.0	779	2	35	133	74.7				MDR's						
Clone #5	05/16/03	5.0	73	3	35	10	10	143		55	54	1	3	26	16		
Clone #5	05/14/03	2.9	889	1	35	72	40.3	65		50/50	MDR's N 40% ???						Disc.
Clone #5	05/14/03	3.3	1022	2	35	96	53.9	94		55	60	6	4	21	9		Disc.
Clone #5	05/14/03	5.4	697	3	35	107	60.2	100		45/45	MDR's Stressed						Disc.
Clone #5	05/22/03	6.1	576	1	35	100	56.2	99	97	55	?						Cane
Clone #5	05/22/03	5.6	415	2 & 3	35	66	371	60	55	55	68	8	4	10	10		Cane
Clone #5	05/27/03	5.5	1649	1	35	259	145.1	253		40/40	MDR's N 35%						Disc.
Clone #5	05/27/03	4.1	707	2	35	82	46.3	82		40/40	MDR's N 30% ???						Disc.
Clone #5	05/27/03	5.6	692	3	35	110	62	104	102	55	80	1	0	10	9		Cane
Clone #5	05/29/03	4.3	1198	1	35	147	82.4	153		50/50	MDR's N 30%						Disc.
Clone #5	05/29/03	3.2	342	2	35	31	17.5	27	25	50							Cane
Clone #5	05/29/03	3.6	617	3	35	63	35.5	60	56	55							Cane
Clone #5	06/02/03	5.0	1288	1	35	184	103	183	181	55							Cane
Clone #5	06/02/03	5.0	1649	2	35	235	131.9	229	227	55							Cane
Clone #5	06/02/03	5.1	502	3	25	102	57.3	96	94	60	78	2	2	10	8		Cane
Clone #5	06/05/03	3.0	697	1	25	83	46.5	80		20/25							Disc.
Clone #5	06/05/03	5.0	595	2	25	119	66.6	116	114	50	68	2	2	13	15		Cane
Clone #5	06/05/03	6.9	595	3	25	159	89	159	156	50							Cane
Clone #5	06/09/03	7.0	118	1	35	223	125	228		30/30	46	1	3	25	25		Disc.
Clone #5	06/09/03	3.5	881	2	25	183	68.9	124		40/40							Disc.
Clone #5	06/09/03	4.0	740	3	25	118	66.1	119		45/45	52	2	4	12	30		Disc.
Clone #5	06/12/03	6.2	1166	2	25	289	161.9	288		30/35							Disc.
Clone #5	06/12/03	5.6	291	3	25	65	36.5	62		30/30	73	2	0	8	17		Disc.
Clone #5	06/12/03	4.5	672	4	25	120	67.7	118	116	55	73	4	0	12	11		Cane
Clone #5	06/16/03	3.4	779	1	25	105	59.3	105		15/	MDR's						Disc.
Clone #5	06/16/03	14.5	683	2,3,4	25	397	222.4	397		50/	51	3	3	30	13		Disc.
Clone #5	06/19/03			1	Disc												
Clone #5	06/19/03	3.9	936	2		146	81.7										
Clone #5	06/19/03	4.5	817	3	25	147	82.3										
Clone #5	06/19/03	4.0	534	4	25	85	47.8										

Note: The data was transcribed from handwritten notes of the technician performing collection/evaluation of sperm

Appendix H:

The Comprehensive Veterinary Examination

Appendix H: The Comprehensive Veterinary Examination

CVM recognizes that readers of this Risk Assessment will have diverse training and expertise, and not all will be familiar with the practice of veterinary medicine. The purpose of this Appendix is to (1) acquaint readers with a process used by veterinarians (and specifically veterinarians working with livestock) as part of the evaluation of the health of an animal, and (2) how we interpreted veterinary examination reports on animal clones as part of the evaluation of the overall risk assessment process. It is not intended to be a guidance for veterinarians working with animal clones or any other livestock.

A. Introduction to the Comprehensive Veterinary Examination

The standard veterinary procedure that forms the basis of the CVE is referred to as the “Problem Oriented Medical Approach (POM).” Traditionally, this approach involves a Chief Complaint followed by a Physical Exam, which then generates a Problem List. The veterinarian then develops a list of Differential Diagnoses (probable causes), from which a Diagnostic Plan is formulated, including the conduct and interpretation of laboratory work. This leads to a revised Problem List based on laboratory data that implicate particular diseases or conditions. The new information results in a revised Differential Diagnosis and revised Diagnostic Plan (if additional work is still needed). All of the data finally results in a Therapeutic Plan. The owner of the animal is then informed by a Client Education Plan (Weed 1970).

Comprehensive veterinary examinations (CVEs) are systematic approaches for examining domestic livestock animals, and making informed judgments as to their health. The CVE contains both objective and subjective information, and requires experienced veterinarians familiar with the animal breeds and species to be evaluated. For each breed and species, the veterinarian is able to judge whether any observation is “normal” within the range of biologic variability. The POM is the standard medical examination and evaluation technique. This approach compares the non-healthy patient to the clinically normal animal. Each abnormal observation is recorded in a “problem list.” The veterinarian ultimately refines and compiles the problem list to develop a list of differential diagnoses that would account for the problems found on the exam. The notion of “normal” is difficult to define and articulate, and, of necessity, requires subjective assessment and expert judgment. In contrast, measurements such as temperature, pulse, respiratory rate, weight, height, *etc.*, are objective and, in principle, easier to define as “within normal range.” Experienced examiners will agree that published “normal ranges”

of these values can be slightly exceeded without judging the animal to be unhealthy. The CVE, by an experienced practitioner, employs both objective and subjective information to maximize the information gathered in examinations.

B. The Importance of Species-Specific Standards

As the purpose of this Appendix is to provide an overview of the CVE, it will not explicitly address every difference among the four species under consideration in this Risk Assessment. This Appendix will discuss the general approaches that are employed in performing such examinations. Species differ in, among other things, size, shape, demeanor, physiology, nutritional needs, and husbandry. Because of this, cows are examined differently from swine, goats, or sheep. For example, an experienced veterinarian or animal handler would not approach a dairy cow by moving toward her head first. Dairy cows are accustomed to being approached from the rear because that is how they are approached when they are milked. Other species-specific differences may be physiologically determined (*e.g.*, birth weight ranges, reproductive behavior, gestation length) or as the result of species- or breed-specific husbandry (*e.g.*, age of weaning, handling, and feeding practices). Thus, comparing the birth weight of Holstein and Longhorn calves would be inappropriate, as is comparing piglet weaning weights in facilities that wean at different ages.

C. How a Veterinarian Performs a Comprehensive Veterinary Examination

The physical examination portion of a CVE for any age of animal usually begins with an observation from a distance, so as not to influence the animal's response. An animal approached by a human who is not normally involved in the animal's care can affect its demeanor and respiratory rate. First, the consulting veterinarian observes the animal's behavior and general appearance from a distance far enough not to provoke anxiety. The veterinarian notes the animal's posture, resting respiratory rate, vocalizations, eating/cud chewing, excretory functions (defecation/feces and urination), gait, body condition, conformation, and general skin and coat appearance.

The subsequent close examination occurs in a systematic fashion. A CVE is generally carried out in anatomical order, usually from head to tail (except for dairy cows). If any abnormalities are discovered during the course of the examination, they are listed by functional systems, which include

- Special senses (eyes, ears, nose, mouth)
- Skin and Coat

- Musculo/Skeletal (including a body condition score)
- Respiratory
- Cardio/Vascular
- Neurological
- Gastro/Intestinal
- Genito/Urinary
- Peripheral Lymph Nodes.

Often, the veterinarian begins with inserting a thermometer in the rectum to measure body temperature, and, if appropriate for the species, takes a peripheral pulse in the tail (sheep with docked tails are the exception). The veterinarian then begins the examination at the head. The eyes, ears, mouth, and nose are examined. Hydration, anemia status, and other physiological measures can be assessed by examining mucous membrane color, feeling the gums for moistness, and pinching the skin to look for tenting, a sign of dehydration. The veterinarian then looks at the eyes to determine the degree to which they may be sunken from possible dehydration, whether there is any yellow coloration (jaundice from liver dysfunction), abnormal odors or discharges, whether the pupils are of similar size and symmetry (neurological function), and may perform a retinal fundic exam. While working on the head, the veterinarian can look at, smell, and touch the mouth, teeth, tongue, gums, lips, ear position, eyelid position, nostril flaring, feel for muscle atrophy, even breath from the nostrils, tongue tone, and look for drooling (ptyalism). The veterinarian then proceeds to the neck, where s/he looks, listens (auscults) and feels (palpates) for lumps, enlarged lymph nodes, salivary gland abnormalities, larynx, changes in the size or symmetry of the thyroid gland or trachea, the strength and regularity of the jugular pulse, and abnormal tracheal and pharyngeal sounds.

The next area examined is the thorax. The veterinarian observes respiratory excursions more carefully than from the initial distance overview, and notes abnormalities such as abdominal breathing or forced expirations. The veterinarian auscults (listens with a stethoscope) the heart and lungs and listens for signs of thoracic disease. Some veterinarians choose to look at the front legs and feet at this time and note any skin, musculoskeletal, hoof, or other lesions. Another approach is to look at all four legs and gait separately as its own system.

The abdomen of the animal is examined by auscultation and palpation. In ruminants, for example, the veterinarian watches the left side of the animal's abdomen for rumen movements; these are related to cud chewing. S/he then listens to the left side of the abdomen to hear the rumen moving, and palpates the prefemoral and prescapular lymph

nodes. Peritonitis (in cows) is investigated by pinching the withers to see if the cow buckles down or lordoses (arches downward) her spine. If lordosis is not observed, the animal may have peritonitis caused by hardware disease (traumatic reticulo-pericarditis, usually caused by accidental ingestion of nails or wire). In addition, while on the left side, the veterinarian pushes in on the rumen to determine fill and consistency, uses a stethoscope to listen for “normal” borborygmi (intestinal sounds), and thumps on the cow’s side to determine whether a displaced abomasum is present by listening for the characteristic pinging sound of the displacement. While examining the abdomen, s/he also has access to the umbilicus, the udder (females), prepuce (males), tail, perineum, escutcheon, and back legs. The right side of the abdomen is examined similar to the left. Pings on the right have different meanings than those on the left in all species. Lymph nodes are palpated on the right as well. The abdomen may be balloted (punched to feel for a wave of fluid). In smaller ruminants and pigs, the veterinarian palpates the abdomen by placing her/his hands behind the rib cage and pushing in and up (toward the spine) to feel for any abnormal viscera and for a gravid uterus.

In mature female animals, the veterinarian checks the mammary glands for symmetry, temperature fluctuations, swelling, or pain. The veterinarian then examines the teats at their distal end for streak canal abnormalities. Next s/he examines the milk for abnormalities such as watery or clumpy consistency, clotting, an off-color or odor, or the presence of blood. The mammary lymph nodes are examined.

External genitalia are also examined as part of the CVE (additional examinations are required for Breeding Soundness Examinations), including the vulva, prepuce, scrotum and testicles. Internal female and male genitalia are examined during rectal palpation.

If the limbs had not been examined during the thoracic and abdominal exam, they would be examined next. Skin, hoof, conformation, swollen or hot joints, sore ligaments, and lameness (gait abnormalities) are evaluated.

As ancillary tests, the veterinarian may elect to perform rectal palpation for gastrointestinal, genitourinary, cardiovascular, renal, lymph nodes, and musculoskeletal abnormalities.

1. Specific Considerations for Neonatal CVEs

Practitioners generally recommend examining calves, piglets, kids, and lambs within a few hours of birth for general demeanor including alertness, ability to stand, ability to suckle, respiratory excursions, mobility, etc. The neonate is then examined for cleft

palate, nasal abnormalities, and joint and limb abnormalities, and their chests are auscultated for cardiac abnormalities. Body temperature is also measured. Non-patent anus, or *atresia ani* may be discovered. Pigs, specifically, are examined for umbilical or inguinal hernias.

The newborn animals are weighed, and those measurements entered into a birth weight database used to calculate Expected Progeny Differences (EPD), especially in beef cattle. (Cattle producers use these real data to improve their herds.) Birth weights also determine “calving ease” for the dams. Passive transfer of immunity from colostrum is measured by taking blood samples and measuring IgG and total protein at approximately 24 hours post partum after the calf has had an opportunity to consume colostrum.

2. Specific Considerations for Juvenile CVEs

In addition to the general physical exam information on weaning weight, age at weaning, and weight gain is often collected. The producer often keeps health records, which may include sick days, diagnoses, therapeutic records, laboratory data, etc. In particular, animals may be monitored for infections at the navel, in the joints, lungs (pneumonia), and gastrointestinal system (diarrhea).

3. Specific Considerations for Mature Animal CVEs

Most cattle, swine, sheep, and goats do not live out their natural lifespans in commercial production facilities. Thus, for utility in assessing health of the animal prior to use, producers may perform formal CVEs as close to the animal’s final use (*e.g.*, slaughter) as possible, although health records may be maintained by producers for as long as the animal is alive. Health records include illnesses, therapies, growth, weight gain, and might also include feed efficiency (for meat animals), milk production, reproductive records, and dry matter intake in dairy cows, among others.

4. Specific Consideration for Reproductive Stage Examinations

a. Males

Breeding Soundness Examinations (BSEs) have been developed and standardized by many professional societies and graduate veterinary schools. Among others, the Society of Theriogenology has a standardized protocol for bulls (www.therio.org). Its utility in comparison with others has been reviewed by Higdon et al. (2000). Shipley (1999) and Levis at the Ohio Pork Industry Center of the Ohio State University Extension (<http://porkinfo.osu.edu/levis.html#Top>) have published guides for evaluating

reproductive performance in boars (intact male pigs). Breeding soundness exams have been developed for the small ruminants (*e.g.*, rams and bucks) and can be found at web sites of the Utah State University Extension and the University of Minnesota, among others.

Briefly, a BSE includes evaluation of reproductive behavior, anatomical status, and evaluation of semen quality. Behavioral observations include libido, recognition of receptive females, and appropriate mounting behavior. Anatomic evaluations include body condition, with particular attention to the eyes, feet, and legs. Physical examinations are generally performed externally and rectally. The testes, scrotum, and epididymis are palpated, and scrotal circumference measured. The penis and prepuce are examined for appropriate anatomic development (*e.g.*, penile deviations) as well as inflammation, abscesses, or adhesions. The prostate and seminal vesicles are evaluated by rectal palpation. Semen quality is evaluated by looking at volume, color, motility, and morphology. Evaluations are made in the context of age, breed, and species-specifics. Once breeding has occurred, measures of fertility generally are recorded.

b. Females

Evaluations of female reproductive function have not been formalized in the same manner as those for males, largely due to the role of the stud in modern agricultural practice. Nonetheless, there are standard reproductive function examinations that carry the same importance in female animals as do the male examinations. The female-specific examinations include behavioral and anatomic components. Evaluation of germ cell production is generally not measured except by fertility and fecundity. Behavioral observations include age at onset of estrous behavior (puberty), behavior at parturition, offspring acceptance, and mothering behavior. Anatomical evaluations include body condition, examination of external genitalia, and evaluation of the internal reproductive tract by rectal palpation, vaginal speculum, and ultrasound (if clinically indicated). Fertility is monitored by recording the number of coital or insemination attempts needed for conception, (although, in the event of conception failure, the male contribution is also considered). The producer often keeps records of abortion incidence, birthing interval, retained placenta (especially for cows), mastitis (inflamed mammary gland or udder), and metritis (infected uterus). The colostrum management program is often monitored and evaluated using a colostrometer for colostrum quality and blood levels of IgG in the calf for transfer of passive immunity.

D. Interpreting the Comprehensive Veterinary Examination for Animal Clones

The CVE was devised for sick animals, and works well when the patient has a chief complaint requiring diagnosis and therapy. Laboratory work is designed to gather evidence to support the clinical diagnosis. For animal clones, however, the CVE is attempting to prove the negative (*i.e.*, this animal is not materially different from a sexually reproduced animal), and therefore attempting to “prove” that the animal clone is healthy. Examination and diagnostic testing (such as blood work) as a screen for healthy animals (or animal clones) to determine if the animals have a general, non-specific occult abnormality is not straightforward. In the absence of the “complaint, diagnosis, and treatment” paradigm, if one value is out of range in an otherwise apparently healthy animal with an otherwise “normal” clinical chemistry screen, it is difficult to determine what level of concern to place on the anomalous value.

Reference ranges are based on a population of “normal” animals for a particular laboratory and species. In general, the reference ranges result from the collection of all of the measurements that have been taken by that laboratory, and calculation of the mean. The reference range is then set at the mean plus or minus two standard deviations, or a $p < 0.05$ or 95 percent confidence interval. In other words, of any 20 test runs, one will have a value that is out of the range based on the statistical cut-offs as previously defined.

There may also be biological explanations for “out of range” values. For example, alkaline phosphatase is an enzyme found in every tissue, and is released when the cells of that particular organ or tissue are damaged. Elevated levels can be associated with diseases of the bones such as rickets, osteomalacia, hyperparathyroidism, and bone tumors, or as part of such normal processes as healing fractures, pregnancy, or growth.

To place this discussion in the context of the whole animal, we refer to the adage that “one treats the patient, not the blood work.” CVM’s determination that there is a biologically relevant difference between an animal clone and a sexually reproduced animal was based on a combination of statistical validity, weight of evidence, biological plausibility, and clinical impression; that is, a reliance on the body of evidence and expert veterinary judgment.

E. Parameters Evaluated in the Comprehensive Veterinary Exam for a Risk Assessment

The following series of tables presents the parameters used in evaluating the health status of animal clones when incorporated into a CVE.

Table H-1: Clinical Chemistry Parameters Associated with Organs and Organ Systems <i>(adapted from Hayes Principles and Methods of Toxicology 1994)</i>	
Heart	Creatine kinase (creatine phosphokinase) and isoenzymes Lactate dehydrogenase and isoenzymes
Liver	Alanine aminotransferase Albumin Alkaline phosphatase Ammonia and Blood Urea Nitrogen Aspartate aminotransferase Bile acids Bilirubin Gamma glutamyl transferase Lactate dehydrogenase and isoenzymes Sorbitol dehydrogenase Total protein
Kidney	Albumin Blood Urea Nitrogen Chloride Creatine (urine and serum) Glucose (urine and serum) Potassium Protein Sodium
Pancreas	Amylase Calcium Glucose Lipase
Bone	Alkaline phosphatase and isoenzymes Calcium Phosphorus Uric Acid

Table H-2: Animal Health Measures for Evaluating Livestock, Including Animal Clones - General Health Status	
Measurement	Comments
Condition at Birth and Perinatal (+48 hours) Period	
Caesarian/Vaginal Delivery	Birthing ease
Examination of Placenta	Cotyledon numbers
Immediate Post-parturition assistance	Fetal-neonatal transition
Birth weight	LOS
Comprehensive Neonatal Examination <i>Generalized</i> <ul style="list-style-type: none"> - Demeanor - Posture/Gait - Respiratory rate - Vocalization - Nursing behavior - Urination/Defecation - Body condition - Body conformation - Skin/coat <i>System Specific</i> <ul style="list-style-type: none"> - Special Senses (ear, eyes, nose, throat) - Integumentary - Musculoskeletal - Cardiovascular - Respiratory - Oropharyngeal - Gastrointestinal - Genitourinary - Neurological - Peripheral lymph nodes 	<p>Take notice if LOS is present, especially in cattle and sheep.</p> <p>Umbilicus Contracted tendons</p> <p>Bloat, diarrhea</p>
IgG	at 24-48 hours after birth

Table H-3: Animal Health Measures for Evaluating Livestock, Including Animal Clones - Condition During Juvenile Post-Weaning Period (All Species)	
Measurement	Comments
<p>Comprehensive Juvenile Animal Examination</p> <p>Weaning weight</p> <p><i>Generalized</i></p> <ul style="list-style-type: none"> - Demeanor - Posture/Gait - Respiratory rate - Vocalization - Appetite/Feed consumption - Urination/Defecation - Body condition - Body conformation - Skin/coat - Body weight/gain <p><i>System Specific</i></p> <ul style="list-style-type: none"> - Special Senses (ear, eyes, nose, throat) - Integumentary - Musculoskeletal - Cardiovascular - Respiratory - Oropharyngeal - Gastrointestinal - Genitourinary - Neurological - Peripheral lymph nodes 	<p>Normal growth rate</p> <p>Contracted tendons</p>
<p>Health/therapeutic records, with special attention to</p> <ul style="list-style-type: none"> - Diarrhea - Pneumonia 	
Diabetes	previously noted in animal clones
Nervousness	previously noted in animal clones

Table H-4: Animal Health Measures for Evaluating Livestock, Including Animal Clones - Reproductive (for males and non-dairy females, during first breeding cycle; early in lactation for dairy animals)

Measurement	Comments
<p>Comprehensive Breeding Soundness Examination (Males)</p> <ul style="list-style-type: none"> -Comprehensive physical examination with special attention to <ul style="list-style-type: none"> -eyes, -feet/legs -prepuce -penis -palpation of testes and epididymides -Scrotal circumference -Rectal Examination, including <ul style="list-style-type: none"> -hernia, -spinal lesions, -internal abscesses, -accessory sex glands -Semen analysis (motility and morphology) -Libido 	<p>per established guidance on a species-specific basis</p> <p>For seasonal breeders, exam should be performed during breeding season.</p>
<p>Comprehensive Reproductive Exam (Females)</p> <ul style="list-style-type: none"> - Age of puberty onset (behavioral and/or physiological measures) -Comprehensive physical examination - Physical exam of genitalia <ul style="list-style-type: none"> -External - Internal <ul style="list-style-type: none"> -Rectal palpation, vaginal speculum, -Ultrasound if indicated by clinical findings -Record review for <ul style="list-style-type: none"> -calving assistance - abortion incidence - calving interval - retained placenta - metritis - mastitis - maternal behavioral traits - colostrum quality using colostrometer 	<p>Concomitant IgG measure in calf for measure of colostrum quality in beef calves, kids, and lambs. First parity dairy animals may not produce adequate quality colostrum. Comparisons should thus be made for appropriate parity level.</p>

Table H-5: Animal Health Measures for Evaluating Livestock, Including Animal Clones - Maturity, Aging, Lifespan (immediately before animal use (e.g., slaughter))	
Measurement	Comments
<p>Comprehensive Mature Animal Examination <i>Generalized</i></p> <ul style="list-style-type: none"> - Demeanor - Posture/Gait - Respiratory rate - Vocalization - Appetite/Feed consumption - Urination/Defecation - Body condition - Body conformation - Skin/coat <p><i>System Specific</i></p> <ul style="list-style-type: none"> - Special Senses (ear, eyes, nose, throat) - Integumentary - Musculoskeletal - Cardiovascular - Respiratory - Oropharyngeal - Gastrointestinal - Genitourinary - Neurological - Peripheral lymph nodes 	<p>Health records kept on all breeding animals, and on all primary animal clones. They include all veterinary diagnoses, therapies, and vital statistics such as birth weight, weaning weight, physical exam findings, etc.</p>
Growth Performance/ Weight gain	
<p>Signs and Symptoms Observed in Animal Clones</p> <ul style="list-style-type: none"> -Arthritis -Diabetes -Nervousness -Seizures -Neoplasms -Other unspecified signs of early aging 	

Table H-6: Animal Health Measures for Evaluating Livestock, Including Animal Clones - Clinical Measurements	
Measurement	Comments
Biochemistry	
Albumin	
Alkaline Phosphatase	
Amylase	
ALT (SGPT)	
AST (SGOT)	
Bile acids	
BUN	
CIAP (calf intestinal alkaline phosphatase)	for bovine species
Creatine phosphokinase	
Gamma glutamyl transferase	
Lipase	
Sorbitol Dehydrogenase	
Cholesterol	
Creatinine	
Glucose	
Serum Protein	
Calcium	
Chloride	
Potassium	
Sodium	
Hemo/Leukograms (CBC)	
Red Blood Cell count	
WBC count including Differential	
Platelet count	
Urinalysis	
Specific gravity	
Glucose	
Ketones	
Bilirubin	
pH	
Cells	
Protein	
Bacteria	
Blood (including leukocytes)	
Nitrate	

The following table describes the nature of the laboratory tests that have been performed during a CVE, and what they measure. Some common abbreviations, expanded explanations of the functional descriptions, and how the tests are used in differential diagnosis were added during the review of the submitted data as part of the risk assessment.

Table H-7: Standard Large Animal Panel (Blood Biochemistry) Often Performed During a CVE		
Test	Origin	Functional Description
Sodium (Na ⁺)	Diet	Principle cation of extracellular fluid (ECF or plasma). Maintains osmotic pressure of ECF. Cannot evaluate the electrolytes (Na ⁺ , K ⁺ , Cl ⁻ , HCO ₃ ⁻) by themselves. They are interdependent. Their regulation depends on hydration status, disease, aldosterone, renin-angiotensin, acid/base status, etc. Low Na ⁺ may indicate - diarrhea, vomiting, congestive heart failure, renal disease, ruptured bladder. Elevations may indicate - dehydration, vomiting and diarrhea, inadequate intake, renal failure, increased salt intake, artifact from improper sample handling.
Potassium (K ⁺)	Diet	Principle cation of intracellular fluid (ICF). Maintains osmotic pressure within the cell (osmotic balance). Elevations effect cardiac function. Plasma levels may be altered by diarrhea, renal failure, metabolic or respiratory acidosis (causes elevated K ⁺ , a.k.a. hyperkalemia), anorexia, hypoadrenocorticism (Addison's disease), ruptured bladder, artifact from hemolysis of sample.
Chloride (Cl ⁻)	Diet	Major plasma (ECF) anion. Used to calculate the anion gap. Also affected by acid/base status. Reductions seen with chronic vomiting.
Bicarbonate (HCO ₃ ⁻)	Metabolism	Indicative of the CO ₂ concentration in blood, buffers blood from radical pH changes. Used to diagnose metabolic or respiratory acidosis or alkalosis.
Anion Gap	Calculated	Calculated value to represent the unmeasured anions in the blood. High anion gap may indicate ketoacidosis, lactic acidosis, renal failure (uremic acidosis) and ethylene glycol toxicosis (organic acidosis). Low anion gap is RARE. It may indicate low plasma albumin, or high calcium
Blood Urea Nitrogen (BUN)	Liver	Urea is produced by liver and excreted by kidneys. High values may be pre-renal (dehydration), renal (disease of kidney), or post renal (disease of ureter, bladder or urethra). Low values may indicate chronic liver failure (lack of production) or dietary protein deficiency.
Creatinine	Muscle	Product of creatine metabolism by muscle tissue and excreted by kidneys. Used as an important measurement of kidney function. Resting level directly related to muscle mass. Higher in non-castrated males than females.
Calcium (Ca ⁺⁺)	Diet, bone	Regulated by parathyroid hormone and calcitonin (from the thyroid). Vitamin D synthesized in liver important in dietary absorption. Acid base status and albumin level can affect blood levels. Co-factor for many enzymes. Key role in bone

Table H-7: Standard Large Animal Panel (Blood Biochemistry) Often Performed During a CVE		
Test	Origin	Functional Description
		development, blood coagulation, cell growth, neuro-muscular transmission. Intestinal absorption may be affected by diarrhea or vitamin D deficiency.
Phosphorus (P)	Diet, bone	Similar to Ca ⁺⁺ Indicative of parathyroid and thyroid gland function, renal function. Important as a buffer for the blood. Blood level primarily regulated by the kidneys. Abnormalities related to dietary deficiency, renal excretion and hormonal imbalances that would also affect Ca ⁺⁺ .
Magnesium (Mg ⁺⁺)	Diet	Required for normal muscle and nervous tissue function. Co-factor for many enzymes, especially kinases and phosphatases. Influences the regulation of serum calcium. Mostly clinical relevance in a deficiency known as “Grass Tetany” usually from acutely feeding the cattle lush rye pasture which is notorious for Mg ⁺⁺ deficiency.
Total Protein (TP)	Liver, Immune System	Consists mostly of proteins produced by liver (albumin, carrier proteins) and immunoglobulins. Used as indicator of total plasma volume.
Albumin (alb)	Liver	Regulates plasma osmolarity. Binds certain molecules (some drugs, Ca ⁺⁺). Deficiency can be increased loss or decreased production. Loss may come from malfunctioning kidneys, intestine, or leakage into a body cavity. Decreased production may come from chronic liver failure.
Globulin (glob)	alpha-, beta-liver; gamma-B lymphocytes	Sum of globulins produced by the liver (transport proteins, like haptoglobin) and immunoglobulins (passive acquired from colostrum or endogenous production from B lymphocytes that have matured into plasma cells).
Albumin/Globulin Ratio (A/G)	Calculated	Used to determine if there is an overproduction of gamma globulin which may occur in autoimmune disease. Low values may be due to insufficient albumin (see albumin for discussion)
Glucose	Digestion, liver glycogen, synthesis	Indicative of the energy state of the animal. Can be reduced (hypoglycemia) in anorexia or due to artifact (sample handling), increased in diabetes mellitus (rare in cattle) or stress (common in cattle)
Alanine Aminotransferase (AST)	Liver, muscle	Present in liver and released if liver is damaged. May also indicate skeletal and cardiac muscle damage.
Sorbitol Dehydrogenase (SDH)	Liver	Present in cytosol of liver cells. High serum levels may indicate liver damage.
Alkaline Phosphatase (Alk Phos)	Organ Membranes	Present in hepatocytes, biliary epithelium, osteoblasts, placenta, intestine, and kidney. May be high in young animals due to bone growth. Also an indicator of cholestasis (impaired bile flow).
Gamma glutamyltransferase (GGT)	Organ Membranes	Present in hepatocytes, biliary epithelium, and kidney. Used to detect cholestasis. Has no involvement with the skeletal system to differentiate it from alkaline phosphatase. It is a better indicator of biliary stasis in large animals.
Total Bilirubin (T bili)	Hemoglobin Degradation	Increase in bilirubin can result in icterus (jaundice). Used as a measure of liver maturity and function.
Indirect Bilirubin	Hemoglobin	Bilirubin not conjugated to various carbohydrates for transport

Table H-7: Standard Large Animal Panel (Blood Biochemistry) Often Performed During a CVE		
Test	Origin	Functional Description
	Degradation (Non-Conjugated)	into bile in to the liver (elevations are from pre-hepatic sources). Value may be elevated by hemolysis or internal hemorrhage. Used to assess hepatobiliary function.
Direct Bilirubin	Hemoglobin Degradation (Conjugated)	Bilirubin conjugated to various carbohydrates for transport to allow for inclusion into micelles in bile for transport from the liver (elevated in post-hepatic obstruction). Elevated with biliary outflow obstruction. Used to assess hepatobiliary function.
Amylase	Pancreas	Used by GI system to aid in digestion of starch and sugars. Elevations indicate pancreatic inflammation. Not important in ruminant species.
Cholesterol	Liver Synthesis, diet	Precursor for synthesis of steroid hormones, bile acids, and vitamin D. Constituent of cell membranes and bile micelles. Variations may be secondary to endocrine, hepatic, or renal disease.
Creatinine Phosphokinase (CK or CPK)	Muscle (skeletal and cardiac), Brain	Intracellular enzyme in skeletal and cardiac muscle. Used to detect damage to muscle.
Iron (Fe ⁺⁺)	Diet	Ferric Iron associated with transferrin. Iron deficiency is often suspected as the cause of anemia.
TIBC (Total Iron Binding Capacity)	Transferrin production by Liver	Used as measurement of the total amount of transferrin.
Random Bile Acids (hBA)	Liver	Produced by liver and secreted into bile. Elevations indicate reduced liver function and not necessarily inflammation or biliary stasis.
Lipemia - Index	Lipids in plasma	Measure of the level of lipids in the circulation. Can be caused by diets high in fats and influenced by postprandial (after eating) sampling.
Hemolysis - Index	Lysis of red blood cells	Caused by lysis of red blood cells which results in release of hemoglobin into the plasma. Also used as an indicator of sample quality.
Icterus - Index	Bilirubin in plasma	Index measured by the color of plasma to indicate the amount of bilirubin. It is qualitatively measured by comparing to standard colors. It varies with labs, species, liver disease, dietary intake of carotene in cattle, among other things.
Insulin Like Growth Factor I (IGF-I)	Liver	Synthesized by the liver in response to growth hormone. Used as an indicator of the amount of growth hormone being produced. Elevations can be related to increased nutritional status, low values to negative energy balance. Higher in growing animals.
Estradiol (E ₂)	Ovary	Synthesis is controlled by gonadotropins. Synthesis rates related to ovarian function in females. Used to monitor follicular and luteal activity.
Cortisol	Adrenal Cortex	Synthesis is regulated by the hypothalamus and pituitary. Involved in normal metabolism. Elevated levels are associated with stress.

Table H-7: Standard Large Animal Panel (Blood Biochemistry) Often Performed During a CVE		
Test	Origin	Functional Description
Triiodothyronine (T3)	Thyroid Gland	Synthesis is regulated by the hypothalamus and pituitary. Involved in normal metabolism.
Complete Blood Count (Hemogram)		Description
Hematocrit (Hct)		Refers to the percent of blood that is occupied by red blood cells. Low values are good indicators of anemia.
Hemoglobin (Hb)		Protein used by red blood cells to distribute oxygen to other tissues and cells in the body. Low values are good indicators of anemia.
Red Blood Cells (RBC)		The absolute concentration of red blood cells in the blood. A low red blood cell count is defined as anemia. High count is polycythemia.
Mean Corpuscular Volume (MCV)		The actual volume of the red blood cells. Larger red blood cells may indicate anemia due to B ₁₂ or folic acid deficiency, also may be caused by increase in reticulocytes; smaller red blood cells may indicate anemia due to iron deficiency.
Mean Corpuscular Hemoglobin (MCH)		This test measures the amount of hemoglobin in red blood cells. Both hemoglobin and hematocrit are used to calculate this number. Low levels indicate anemia. Of limited diagnostic value.
Mean Corpuscular Hemoglobin Concentration (MCHC)		This test measures the amount of hemoglobin in red blood cells. Both hemoglobin and hematocrit are used to calculate this number. Low levels indicate anemia.
Red Cell Distribution Width(RDW)		RDW evaluates the range of sizes of RBCs in a blood sample. If anemia is suspected, based on other blood counts, RDW test results are often used together with MCV results to determine the cause of anemia.
White Blood Cells (WBC)		Leukocytes (WBCs) are produced by the immune system (in bone marrow) to help defend against infection. A high WBC count likely indicates an infection, whereas a low number might be an acute response where readily available cells are summoned to the site of infection or due to immunosuppression.
Segmented Neutrophils (segs)		Phagocytic cells present to guard against infection, particularly bacterial. Segmented neutrophils are mature neutrophils, and are the predominant white cell in non-ruminant mammals.
Banded Neutrophils (bands)		Immature neutrophils. Elevated levels occur in response to a recent infection.
Lymphocytes (lymphs)		T-cells, B-cells, and natural killer (NK) cells. Viral infections can either increase or decrease the total percentage of lymphocytes. It is the predominant white cell in ruminants.
Monocytes (monos)		Monocytes are a type of phagocyte that mature into macrophages. A low number can indicate a higher risk of bacterial infection.
Eosinophils (eos)		Active in killing parasites, can inhibit mast cells or release mediators of inflammation. A high number of eosinophils can indicate allergies or parasitic infections.
Basophils (basos)		Function unclear. A type of phagocyte that produces the anti-inflammatory protein histamine.
Platelets (Thrombocytes)		Tiny cells produced by the bone marrow to help blood clot

Table H-7: Standard Large Animal Panel (Blood Biochemistry) Often Performed During a CVE		
Test	Origin	Functional Description
		formation. High number might indicate a blood disease. A decreased platelet count is called thrombocytopenia. Used to measure immune system function.
Mean Platelet Volume (MPV)		Measures the average volume of platelets. May be artifactually high due to clumping of platelets in blood sample.
Total Protein-refractometer (TP-ref)		The total amount of protein in the plasma measured by a refractometer.
RBC Morphology		General morphology (shape) of red blood cells. Poikilocytes - RBCs of irregular shape. Schizocytes - poikilocytes from fragmentation due to flowing through damaged small vessels.
Parasites		The blood sample is examined for the presence of parasites.
Complete Blood Count (Hemogram)		Description
WBC Exam		Morphological appearance of the white blood cells.
Plasma Appearance		General appearance of the plasma.

Appendix I:

Investigation on the Attributes of Cloned Bovine Products

Appendix I

Investigation on the Attributes of Cloned Bovine Products

The Japanese Research Institute for Animal Science in Biochemistry and Toxicology provided an unpublished bound report “Investigation on the Attributes of Cloned Bovine Products” by the Japan Livestock Technology Association (Japan, 2002). The 489 page report, provided in the original Japanese, and was accompanied by an eight page August 2002 English-language summary.

This appendix contains a translation of the first three pages of the bound report and the eight page English summary. These are followed by tables from the original bound report. The tables present the results of a feeding study in which rats were fed diets containing freeze dried milk or freeze dried beef from ordinary cattle and clone cattle at concentrations of 0, 2.5, 5, or 10% of the diet for 28 days. General signs, body weight, food consumption, urinalysis, sensory and reflex function, spontaneous movement frequency, general function, reproductive cycle, hematology at autopsy, blood chemistry, organ weights, pathology and histopathology were compared between groups. English-language tables were provided in the original Japanese-language report with the results. These tables are included in this appendix.

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**English language translation of the first three
pages of the report “Investigation on the
Attributes of Cloned Bovine Products”**

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A project aided by the Agriculture and
Livestock Industries Corporation

PROJECT REPORT OF AN INVESTIGATION ON THE PROPERTIES
OF PRODUCTS FROM CLONED CATTLE
(An Urgent Study Project for the Utilization of Cloned Cattle)
(1999 – 2001)

September 2002

Japan Livestock Technology Association

Results of an investigation on the properties of products from cloned cattle

Rapid advances have been made in the application of cloning technology in cattle multiplication. Products from embryonic clones of cattle are already on the market as safe foods. The Ministry of Health, Labor and Welfare (MHLW) is currently gathering data on the safety of products from somatic cell cloned cattle and has released an interim report which states that so far there is no reason to anticipate safety-related problems.

Cloning technology is expected to advance further and come into wide use as a technology that can provide inexpensive meat, milk, etc to consumers. However, for this to happen, it is essential for the meat and milk of cloned cattle to become widely accepted by consumers as safe, high-quality commodities.

This Association conducted various investigations on cloned cattle, with grants from the Agriculture and Livestock Industries Corporation (ALIC), during 1999 to 2001. An investigation on the properties of products from cloned cattle, which was a part of these investigations, was commissioned to the Research Institute for Animal Science in Biochemistry and Toxicology (RIAS).

The objective of this investigation was to collect data that would confirm the safety of products from embryonic clones of cattle, which are already in use as food items, and also data that would be useful in evaluating the safety of products from somatic cell clones. For this purpose, the properties of the blood, and the composition of nutritional components such as proteins, lipids, amino acids and fatty acids of raw milk and meat were analyzed and compared among ordinary cattle, embryonic clones and somatic cell clones of Holstein and Black Japanese breeds. Digestibility studies with artificial digestive fluid and with rats, allergenicity and mutagenicity (micronucleus) tests with mice, and a 14-week feeding study with rats were also conducted and the digestibility, allergenicity, and mutagenicity of the products, and their effects on the growth, functions and morphology of the test animals were compared. The results of none of the analyses or tests showed any significant differences between products from ordinary cattle and the two types of cloned cattle. Also, no harmful effect attributable to the raw milk or meat of the two types of clones was observed.

September 10, 2002

Cloned Cattle Investigation Committee
Japan Livestock Technology Association

– Contents –

Results of an investigation on the properties of products from cloned cattle

Urgent study project on the utilization of cloned cattle – List of the “Cloned Cattle Investigation Committee” members

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2. Tests on the properties of blood of cattle covered by the investigation on the properties of products from cloned cattle
3. Analysis of nutritional components of raw milk and meat of cloned cattle
4. Allergenicity tests of raw milk and meat of cloned cattle by the mouse abdominal wall method
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**English language summary of the report
“Investigation on the Attributes of Cloned
Bovine Products”**

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August 13, 2002
Livestock Technology Division,
Livestock Industry Department,
Agricultural Production Bureau,
Ministry of Agriculture,
Forestry and Fisheries

RE: Outline of Investigative Results on the Attributes of Cloned Bovine Products

An interim report of the Ministry of Health and Welfare (MHW) released in June 2000 concerning the safety of foods made from cloned cattle states, "There is no scientific basis for fearing the safety of foods", and it recommended that foods derived from BNT cloned cattle be sold with labeling to that effect (optional labeling). The interim report also stated that, "It would be desirable to obtain data on a greater number of cloned cattle that would support safety." Because somatic cloning technology is a newer technology, the Ministry of Agriculture, Forestry and Fisheries (MAFF) is requesting self-restraint on the shipment of SCNT cloned cattle. The Ministry of Health, Labour and Welfare (MHLW: a new Ministry changed from MHW) are currently conducting an investigative study on safety.

In response, the Research Institute for Animal Science in Biochemistry and Toxicology has been conducting a study on the attributes of cloned bovine products (emergency study project on cloned bovine usage). The results of this study have now been gathered so a summary is attached separately.

It is intended that the results of this study will be submitted as reference material for the "investigative research on the safety of animal foods that use cloning technology" currently being conducted at the MHLW.

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Outline of Investigation on the Attributes of Cloned Bovine Products

Commissioned by the Livestock Technology Association from FY1999, we conducted an investigation on the attributes of cloned bovine products. The following are the results of this investigation.

1. Objectives

To conduct an investigation on the blood attributes of cloned cattle (BNT cloned cattle or SCNT cloned cattle), and analyze the components of cloned bovine products (milk and beef), as well as to conduct a study on animal feeding of feed additives from cloned cattle, and obtain data comparing cloned bovine products and existing foods (products from ordinary cattle produced by artificial insemination, etc).

2. Outline of investigation

(1) Blood test

(Material and method)

Blood was sampled from ordinary cattle and cloned cattle at 3, 6, and 9 months of pregnancy and 3 and 6 weeks after birth in the case of dairy cattle (Holstein), and 3 to 4 times during a period from 21 to 28 weeks after birth in the case of beef cattle (*kuroge-wagyu*). The sampled blood was subject to hematological testing (12 items including red blood cell count, white blood cell count, and hemoglobin) and biochemical examination of blood (25 items including total protein, and total cholesterol) and compared.

(Results)

None of the animals showed abnormalities in performance status. There were also no biologically significant differences* in any of the test values between ordinary cattle and cloned cattle, for both the dairy and beef types.

* A biologically significant difference means a difference that could possibly have an effect on factors such as health and survival evident between the study groups. There are no problems if a biologically significant difference and statistically significant difference are in accord, but even if there is a statistically significant difference between the study groups in general, and that the difference is within the range of normal values it is unlikely that health would be affected, so one could not say that a biologically significant difference exists. In the investigative report, biologically significant difference was studied in addition to statistically significant difference. The same applies hereafter.

(2) Analytical study of milk and meat components

(Material and method)

The general components (water content, protein, lipids, and sugars), amino acids (18 types), and fatty acid (21 types) content (content per 100 g) in milk and slices of meat (9 sites) sampled from ordinary cattle and cloned cattle were measured and compared.

(Results)

Although there were slight variations seen among individual cattle, no biologically significant differences were evident in the general components, amino acids, and fatty acid content between ordinary cattle and cloned cattle for both milk and different sites of meat (Table 1).

(3) Milk and meat digestion study

(Material and method)

A study of digestion of pieces of meat sampled from ordinary cattle and cloned cattle by artificial gastric juice and intestinal juice, and a study of digestion of milk and meat that had been frozen, dried, and powdered (freeze-dried food) and added to feed, using rats were conducted, and the digestion rates that were regarded as parameters of protein were compared.

(Results)

There were no biologically significant differences in the rates of digestion of the feed additives due to artificial gastric juice and intestinal juice using rats between ordinary cattle and cloned cattle (Tables 2 and 3).

(4) Allergen testing of milk and meat by mouse abdominal wall method

(Material and method)

We sensitized mice with an intraperitoneal injection of extract from freeze-dried milk and meat slices sampled from ordinary cattle and cloned cattle. Fourteen days later we retracted the abdomen and induced an allergic reaction by re-injection in the abdominal wall. Allergen activity was compared based on the extent of vascular permeability (diameter of dye leakage) seen due to the sensitization treatment.

(Results)

For both milk and meat slices there were no statistically significant differences in allergen activity between ordinary cattle and cloned cattle (Table 4).

(5) Feeding test by the supply of a combination feed of milk and meat using rats

(Material and method)

Freeze-dried milk and meat of ordinary cattle and cloned cattle were each combined with basic feed at concentrations of 2.5%, 5%, and 10% in the case of freeze-dried milk, and 1%, 2.5%, and 5% in the case of freeze-dried meat, and fed to rats (20 per group (10 males and 10 females)) for 14 weeks.

The general sign, body weight, food consumption, urinalysis (8 items), sensory and reflex function, spontaneous movement frequency, general function, reproductive cycle, hematology at autopsy (11 items), blood chemistry (23 items), autopsy and organ weights (brain, pituitary gland, cerebral gland, thyroid gland, heart, lungs, liver, pancreas, adrenal bodies, and reproductive organs) of rats given the feed were compared among a basic feed group, ordinary cattle group, and cloned cattle group.

(Results)

There were no biologically significant differences in each of the items observed and tested over time in rats at any concentration of feed additive for milk and meat between ordinary cattle and cloned cattle (Table 5).

(6) Mutagenicity by milk and meat supply using mice (micronucleus test)

(Material and method)

Feed produced in the feed test by the supply of a combination feed of milk and meat in (5) was given to mice for 14 days whereupon the incidence of bone marrow micronucleus-possessing erythrocytes appearing was tested (micronucleus) and mutagenicity (clastogenicity) was studied.

(Results)

Clastogenicity was negative and mutagenicity was not evident for milk and meat feed additives from ordinary cattle and cloned cattle (Table 6).

3. Summary

The above results revealed no biologically significant differences in component analysis testing and feed additive animal testing between products of BNT cloned cattle and SCNT cloned cattle (milk and meat), and the products of ordinary cattle.

Table 1. General components**(1) Milk**

Classification	Cattle No.	Protein (g/100 g)	Fats (g/100 g)	Sugars (g/100 g)	Ash content (g/100 g)	Water content (g/100 g)	Calcium (mg/100 g)	Cholesterol (mg/100 g)
Ordinary cattle	Min. value	3.0	2.2	4.6	0.7	88.1	100	8
	Max value	3.4	3.3	4.6	0.7	89.7	110	10
	Mean value	3.3	2.7	4.6	0.7	88.9	105	9
BNT cloned cattle	No.1	2.9	2.3	3.0	0.8	91.1	95	9
	No.2	2.9	3.6	3.5	0.7	89.3	105	9
SCNT cloned cattle	No 1	3.1	4.3	4.6	0.7	87.4	120	9
	No.2	3.3	2.6	4.4	0.7	89.1	115	11
	No.3	3.3	3.1	4.5	0.7	88.5	115	10

Note: The analytical values for each animal are the mean of the analytical values for milk sampled at two points – 3 weeks and 6 weeks after delivery.

(2) Meat

Classification	Cattle No.	Protein (g/100 g)	Fats (g/100 g)	Sugars (g/100 g)	Ash content (g/100 g)	Water content (g/100 g)	Cholesterol (mg/100 g)
Ordinary cattle	Min. value	17.8	13.8	0.4	0.9	58.0	50
	Max value	19.6	22.9	0.8	1.0	64.8	68
	Mean value	18.4	19.3	0.6	0.9	60.8	59
BNT cloned cattle		17.4	21.2	0.4	0.9	60.2	56
SCNT cloned cattle		16.8	23.8	0.5	0.9	57.9	68

Note: The analytical value for each animal is the mean value of the analytical values of 9 sites: shoulder, chuck loin, rib loin, loin end, brisket, round, silver side, rump, and tender loin.

Table 2. Rates of meat digestion by artificial digestive juices

Digestive juice	Sample	Rate of digestion after the start of incubation						
		Course	Start	0.75 hr	1.5 hr	3 hr	6 hr	12 hr
Artificial gastric juice	Ordinary beef		0	68	79	-	95	90
	Somatic cloned beef		0	59	78	-	91	90
Artificial intestinal juice	Ordinary beef		0	-	20	40	66	67
	Somatic cloned beef		0	-	28	38	67	63

Note: The digestion rate shows the protein rate of digestion.

Table 3. Digestion rates of milk and meat in rats

Sample	Test group	Number of animals	Digestion rate (mean \pm standard deviation)
Milk	Ordinary cattle	5	83.0 \pm 2.6
	BNT cloned cattle	5	82.7 \pm 2.0
	SCNT cloned cattle	5	81.3 \pm 3.4
Meat	Ordinary cattle	5	83.8 \pm 6.6
	BNT cloned cattle	5	82.3 \pm 4.7
	SCNT cloned cattle	5	84.9 \pm 3.6

Note: Milk and meat were each freeze-dried and combined in feed. The digestion rate shows the protein digestion rate.

Table 4. Allergen study of milk and meat by mouse abdominal wall method

Sample	Test group		Number of animals	Diameter of dye leakage (mm) (mean \pm SD)
Milk	Ordinary cattle	Control group	7	7.0 \pm 3.7
		Test group	10	18.0 \pm 2.9
	BNT cloned cattle	Control group	7	4.7 \pm 3.2
		Test group	10	18.0 \pm 3.9
	SCNT cloned cattle	Control group	7	4.9 \pm 4.6
		Test group	10	17.9 \pm 4.2
Meat	Ordinary cattle	Control group	7	5.3 \pm 5.0
		Test group	10	13.0 \pm 5.9
	BNT cloned cattle	Control group	7	7.0 \pm 4.9
		Test group	10	12.5 \pm 3.5
	SCNT cloned cattle	Control group	7	5.7 \pm 4.2
		Test group	10	13.1 \pm 5.0

Note: Milk and meat were each freeze-dried and the extracts were used as samples. The test groups underwent sensitization treatment and elicitation, while the control groups underwent elicitation only.

Table 5. Feed study of milk and meat by formula feed supply using rats

(1) Changes in rat body weight by milk formula feed supply (mean \pm standard deviation)

Study group		Number of animals	Feeding period (weeks)									Amount of body weight increase in 1-14 weeks (g)	
			1	2	3	4	5	6	8	10	12		14
Male	Basal diet	10	146 \pm 6	189 \pm 20	255 \pm 34	299 \pm 45	344 \pm 59	373 \pm 58	428 \pm 56	448 \pm 58	516 \pm 47	547 \pm 77	401 \pm 76
	Ordinary cattle High concentration (10%)	10	146 \pm 6	194 \pm 11	260 \pm 13	304 \pm 17	350 \pm 19	378 \pm 22	426 \pm 36	475 \pm 43	515 \pm 43	544 \pm 48	398 \pm 48
	BNT cloned cattle High concentration (10%)	10	146 \pm 6	175 \pm 19	245 \pm 15	295 \pm 14	333 \pm 18	367 \pm 16	425 \pm 22	462 \pm 34	503 \pm 41	530 \pm 44	384 \pm 41
	SCNT cloned cattle High concentration (10%)	10	146 \pm 7	196 \pm 10	261 \pm 17	310 \pm 23	353 \pm 27	379 \pm 30	432 \pm 41	473 \pm 41	519 \pm 43	545 \pm 48	399 \pm 43
Female	Basal diet	10	117 \pm 5	150 \pm 6	184 \pm 12	208 \pm 9	229 \pm 10	242 \pm 10	267 \pm 16	283 \pm 15	304 \pm 18	310 \pm 20	193 \pm 18
	Ordinary cattle High concentration (10%)	10	118 \pm 5	152 \pm 7	181 \pm 7	209 \pm 12	234 \pm 16	247 \pm 15	273 \pm 20	298 \pm 21	316 \pm 28	329 \pm 40	211 \pm 42
	BNT cloned cattle High concentration (10%)	10	119 \pm 7	157 \pm 11	186 \pm 12	208 \pm 22	223 \pm 27	244 \pm 26	272 \pm 19	292 \pm 21	313 \pm 26	326 \pm 30	207 \pm 27
	SCNT cloned cattle High concentration (10%)	10	118 \pm 7	151 \pm 12	181 \pm 12	209 \pm 13	229 \pm 16	247 \pm 19	274 \pm 18	293 \pm 19	317 \pm 21	330 \pm 33	213 \pm 33

Note: Aside from studying a 10% milk powder concentration, 10 cattle each were also fed a low concentration (2.5%) and a medium concentration (5%), but no significant differences were noted.

(2) Changes in rat body weight by meat formula feed supply (mean \pm standard deviation)

Study group		Number of animals	Feeding period (weeks)									Amount of body weight increase in 1-14 weeks (g)	
			1	2	3	4	5	6	8	10	12		14
Male	Basal diet	10	143 \pm 5	216 \pm 12	279 \pm 14	336 \pm 16	382 \pm 18	426 \pm 22	489 \pm 31	534 \pm 37	560 \pm 42	590 \pm 50	447 \pm 51
	Ordinary cattle High concentration (5%)	10	143 \pm 5	221 \pm 9	284 \pm 17	337 \pm 24	386 \pm 30	432 \pm 36	492 \pm 44	541 \pm 55	575 \pm 62	604 \pm 65	462 \pm 65
	BNT cloned cattle High concentration (5%)	10	143 \pm 5	219 \pm 10	286 \pm 14	343 \pm 18	392 \pm 21	431 \pm 26	488 \pm 34	535 \pm 39	564 \pm 43	591 \pm 49	448 \pm 51
	SCNT cloned cattle High concentration (5%)	10	143 \pm 6	215 \pm 9	278 \pm 14	336 \pm 20	392 \pm 29	435 \pm 35	499 \pm 48	551 \pm 60	581 \pm 70	613 \pm 80	469 \pm 76
Female	Basal diet	10	120 \pm 4	167 \pm 14	198 \pm 15	228 \pm 18	253 \pm 26	272 \pm 26	290 \pm 26	313 \pm 33	330 \pm 38	338 \pm 35	218 \pm 38
	Ordinary cattle High concentration (5%)	10	120 \pm 4	169 \pm 11	200 \pm 12	230 \pm 15	254 \pm 19	274 \pm 18	297 \pm 23	316 \pm 28	331 \pm 31	341 \pm 30	221 \pm 30
	BNT cloned cattle High concentration (5%)	10	120 \pm 5	171 \pm 8	201 \pm 10	236 \pm 15	260 \pm 20	280 \pm 23	311 \pm 29	330 \pm 27	347 \pm 35	361 \pm 40	241 \pm 39
	SCNT cloned cattle High concentration (5%)	10	120 \pm 4	167 \pm 8	195 \pm 9	227 \pm 11	250 \pm 12	268 \pm 12	292 \pm 16	310 \pm 19	329 \pm 20	336 \pm 20	216 \pm 17

Note: Aside from studying a 5% meat powder concentration, 10 cattle each were also fed a low concentration (1.0%) and a medium concentration (2.5%), but no significant differences were noted.

Table 6. Mutagenicity by the supply (14 days) of milk and meat using mice (micronucleus test)

(1) Milk

Test group	Number of animals	Incidence (%) of micronucleus appearance (Min – max)		Polychromatic erythrocyte rate (%) (Min – max)		Assessment
Negative control group (basal diet)	6	0.27±0.10	(0.1 – 0.4)	49.2±6.6	(42.2 – 57.1)	
Ordinary cattle						
2.5% group	6	0.22±0.17	(0.0 – 0.4)	49.4±3.8	(43.1 – 53.1)	Negative
5% group	6	0.20±0.14	(0.0 – 0.4)	45.7±5.0	(36.8 – 50.3)	Negative
10% group	6	0.12±0.10	(0.0 – 0.2)	44.5±7.5	(35.4 – 56.9)	Negative
BNT cloned cattle						
2.5% group	6	0.30±0.14	(0.1 – 0.5)	44.0±6.7	(36.5 – 55.2)	Negative
5% group	6	0.25±0.12	(0.1 – 0.4)	47.4±8.1	(36.5 – 56.3)	Negative
10% group	6	0.17±0.08	(0.1 – 0.3)	44.7±8.4	(32.3 – 56.8)	Negative
SCNT cloned cattle						
2.5% group	6	0.22±0.13	(0.0 – 0.3)	49.7±7.4	(35.8 – 56.3)	Negative
5% group	6	0.28±0.15	(0.1 – 0.5)	49.5±7.8	(41.2 – 60.9)	Negative
10% group	6	0.25±0.05	(0.2 – 0.3)	44.0±6.2	(34.2 – 52.8)	Negative
Positive control group (Mitomycin C)	6	6.02±1.03**	(4.6 – 7.6)	34.6±5.5	(26.3 – 40.3)	Positive

(2) Meat

Test group	Number of animals	Incidence (%) of micronucleus appearance (Min – max)		Polychromatic erythrocyte rate (%) (Min – max)		Assessment
Negative control group (basal diet)	6	0.20±0.18	(0.0 – 0.5)	47.7±9.7	(30.2 – 59.7)	
Ordinary cattle						
1% group	6	0.17±0.12	(0.1 – 0.4)	50.0±9.1	(37.9 – 61.3)	Negative
2.5% group	6	0.13±0.08	(0.0 – 0.2)	47.3±13.1	(22.3 – 60.2)	Negative
5% group	6	0.12±0.15	(0.0 – 0.3)	46.8±10.5	(37.2 – 63.5)	Negative
BNT cloned cattle						
1% group	6	0.20±0.06	(0.1 – 0.3)	51.0±7.3	(41.3 – 59.3)	Negative
2.5% group	6	0.23±0.14	(0.0 – 0.4)	47.1±4.3	(40.6 – 51.1)	Negative
5% group	6	0.12±0.08	(0.1 – 0.2)	49.6±9.6	(37.9 – 61.9)	Negative
SCNT cloned cattle						
1% group	6	0.18±0.10	(0.1 – 0.3)	48.3±8.4	(35.6 – 55.1)	Negative
2.5% group	6	0.22±0.10	(0.1 – 0.4)	51.7±7.3	(44.3 – 63.9)	Negative
5% group	6	0.22±0.08	(0.1 – 0.3)	48.4±8.1	(38.4 – 58.1)	Negative
Positive control group (Mitomycin C)	6	6.95±1.56**	(4.1 – 8.4)	25.7±6.5	(19.4 – 36.4)	Positive

Note: Milk and meat were freeze-dried and powdered and combined in feed. The positive control group was administered a single dose of 2 mg/kg of mitomycin C intraperitoneally. Values were shown as mean ± standard deviation.

** denotes a significant difference at p<0.01 against the positive control group.

Results of a 28-Day Feeding Study in Rats Fed Diets Containing Freeze Dried Milk or Beef from Ordinary Cattle and Clone Cattle

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Table 1 Analyzed nutrient composition of the milk powder

Macronutrient (%)	Vitamin (mg/100g)	Mineral (mg/100g)
Crude protein 23.3	A 113IU/100g	Ca 1410
Crude fat 23.4	B ₁ 0.27	P 723
Carbohydrate 45.0	B ₂ 1.11	K 2010
Crude fiber 0.0	B ₆ 0.17	Na 195
Ash 5.4	B ₁₂ 0.0017	Mg 79.2
Water 2.9	D ₃ ND	Fe 0.326
	E 0.4	Zn 3.069
	K ₁ ND	Cu 0.064
	K ₂ 0.0070	S 220
	Niacin 0.86	Mn 0.073
	Pantotheinic acid 2.90	I 0.07
	Folic acid 0.05	Se 0.023
	Biotin 0.00919	Mo ND
	Choline 0.10 (%)	

Table 2-1 Composition of the milk powder-contained diets

(g/kg diets)

Ingredient	Basal diet	5% diet	10% diet	20% diet
Milk powder		50.000	100.000	200.000
Cornstarch	397.486	378.180	358.872	320.258
Casein (93.5% protein)	200.000	187.540	175.080	150.160
Alfa-cornstarch	132.000	125.588	119.177	106.355
Sucrose	100.000	100.000	100.000	100.000
Soybean oil	70.000	58.300	46.600	23.200
Cellulose	50.000	50.000	50.000	50.000
Mineral Mix *	35.000	35.000	35.000	35.000
Vitamin Mix *	10.000	10.000	10.000	10.000
L-Cystine	3.000	3.000	3.000	3.000
Choline Bitartrate (41.1% choline)	2.500	2.378	2.257	2.013
t-Butylhydroquinone	0.014	0.014	0.014	0.014
Caloric value (kcal)	395	396	397	398
Crude protein (%)	18.7	18.7	18.7	18.7
Crude fat (%)	7.0	7.0	7.0	7.0
Carbohydrate (%)	64.4	64.6	64.8	65.1
Crude fiber (%)	5.0	5.0	5.0	5.0

* : Ingredients of the mineral mix and vitamin mix are shown in table 2-2.

Table 2-2 Ingredients of the mineral mix and vitamin mix for the milk powder-contained diet

(g/kg Mix)

Mineral Mix						Vitamin Mix				
Ingredients	Contents (M)	Basal diet	5% diet	10% diet	20% diet	Ingredients	Basal diet	5% diet	10% diet	20% diet
ESSENTIAL MINERALS						Vitamine A Palmitate (500,000IU/g)	0.800	0.687	0.574	0.348
Calcium Carbonate	Ca: 40.04	357.00	306.683	256.326	155.652	Thiamine HCl (B ₁)	0.600	0.590	0.570	0.550
Potassium Phosphate (monobasic)	P: 22.78	196.00	161.722	127.570	59.390	Riboflavin (B ₂)	0.800	0.540	0.490	0.380
Potassium Citrate H ₂ O	K: 28.73	124.150 *	93.211	62.174		Pyridoxine HCl (B ₆)	0.700	0.690	0.680	0.670
Sodium Chloride	K: 44.87				45.140	Vitamine B ₁₂ (0.1%)	2.500	2.400	2.300	2.200
Potassium Sulfate	Na: 39.34	74.00	66.822	59.570		Vitamine D ₃ (400,000IU/g)	0.250	0.250	0.250	0.250
Magnesium Oxide	S: 18.39	68.35 *	51.263	34.175		Vitamine B Acetate (500IU/g)	15.000	15.000	15.000	15.000
Ferric Citrate	K: 44.87				16.585	Vitamine K ₁	0.075	0.075	0.074	0.074
Zinc Carbonate	Mg: 60.32	24.00	22.118	20.282		Ritacin	3.000	3.000	3.000	3.000
Manganous Carbonate	Fe: 16.50	6.06	6.06	6.06	5.887	Calcium Pantothenate	1.600	1.500	1.300	1.000
Capric Carbonate	Zn: 52.14	1.85	1.54	1.485	1.32	Folic Acid	0.200	0.200	0.200	0.190
Potassium Iodate	Mn: 47.79	0.63	0.630	0.630	0.624	Biotin	0.020	0.020	0.019	0.018
Sodium Selenate	Cu: 57.47	0.30	0.30	0.295	0.285	Sucrose finely powdered	74.655	975.048	975.543	978.320
Ammonium Paramolybdate 4H ₂ O	I: 59.30	0.01	0.009	0.007	0.003					
	Se: 41.79	0.01025	0.00957	0.00888	0.00752					
	Mo: 54.34	0.00795	0.00795	0.00795	0.00795					
NON-ESSENTIAL MINERALS										
Sodium Metasilicate 9H ₂ O	Si: 9.88	1.45	1.45	1.45	1.45					
Chromium Potassium Sulfate 12H ₂ O	Cr: 10.42	0.275	0.275	0.275	0.275					
Lithium Chloride	Li: 16.38	0.0174	0.0174	0.0174	0.0174					
Boric Acid	B: 17.50	0.0815	0.0815	0.0815	0.0815					
Sodium Fluoride	F: 45.24	0.0635	0.0635	0.0635	0.0635					
Nickel Carbonate	Ni: 45.00	0.0318	0.0318	0.0318	0.0318					
Ammonium Vanadate	V: 43.55	0.0066	0.0066	0.0066	0.0066					

* : Composition is differ from that of the AIN standard diet.

The mineral mix and vitamin mix for each composition was designed to reflect the mineral (essential minerals) and vitamin contents of the milk powder at each additional level.

Table 3 Body weights of male rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	Number of animals	Day	(g)					Gain 0~28
			1	7	14	21	28	
0%	6		125	187	239	290	331	205
		±	3	± 6	± 7	± 10	± 13	± 12
5%	6		125	183	241	297	340	215
		±	3	± 6	± 11	± 16	± 22	± 21
10%	6		125	183	239	290	332	208
		±	3	± 7	± 12	± 18	± 20	± 17
20%	6		125	183	238	281	320	196
		±	4	± 9	± 12	± 21	± 26	± 27

Each value is expressed as mean±S.D.

Table 4 Body weights of female rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	Number of animals	Day	(g)					Gain 0~28
			1	7	14	21	28	
0%	6		116	150	177	202	224	109
		±	5	± 5	± 6	± 7	± 12	± 11
5%	6		116	153	178	203	229	113
		±	5	± 16	± 17	± 21	± 29	± 25
10%	6		115	151	177	202	226	111
		±	4	± 9	± 11	± 13	± 14	± 10
20%	6		117	153	182	207	229	112
		±	8	± 10	± 17	± 22	± 25	± 18

Each value is expressed as mean±S.D.

Table 5 Food consumption of male rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	Number of animals	Week			
		1	2	3	4
0%	6	16 ± 0	19 ± 2	20 ± 1	20 ± 1
5%	6	± 16 2	± 19 1	± 20 1	± 20 0
10%	6	± 15 2	± 19 1	± 20 2	± 20 2
20%	6	15 ± 0	18 ± 0	17 ± 3	19 ± 1

Each value is expressed as mean±S.D.

Table 6 Food consumption of female rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	Number of animals	Week			
		1	2	3	4
0%	6	11 ± 1	14 ± 0	15 ± 1	16 ± 2
5%	6	± 12 1	± 14 1	± 15 3	± 16 3
10%	6	± 11 2	± 13 2	± 14 1	± 15 2
20%	6	11 ± 2	14 ± 2	15 ± 2	15 ± 1

Each value is expressed as mean±S.D.

Table 7 Urinary findings of male rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	No. of animals	Color		Cloudy		pH								Protein					Glucose				
		PY		-	+	5.0	6.0	6.5	7.0	7.5	8.0	8.5	-	±	+	++	+++	-	±	+	++	+++	
0%	6	6		6		3	2	1					1	3	2							6	
5%	6	6		6		4	2						1	3	2							6	
10%	6	6		6		1	5						2	4								6	
20%	6	6		6		4	2						1	5								6	

Conc. in diet	No. of animals	Ketone body					Occult blood					Urobilinogen					Bilirubin				
		-	±	+	++	+++	-	±	+	++	+++	0.1	1	2	4	8	-	+	++	+++	
0%	6	4	2				6					6					6				
5%	6	4	2				6					6					6				
10%	6	3	3				6					6					6				
20%	6	4	2				6					6					6				

Color : PY(pale yellow)
 Cloudy : - (negligible), + (cloudy)
 Protein : - (negligible), ±(15~30mg/dL), +(30mg/dL), ++(100mg/dL), +++(300mg/dL)
 Glucose : - (negligible), ±(0.1g/dL), +(0.25g/dL), ++(0.5g/dL), +++(1g/dL)
 Ketone body : - (negligible), ±(5mg/dL), +(15mg/dL), ++(40mg/dL), +++(80mg/dL)
 Occult blood : - (negligible), ±(trace), +(slight), ++(moderate), +++(marked)
 Urobilinogen : Ehrlich unit/dL
 Bilirubin : - (negligible), +(slight), ++(moderate), +++(marked)

Table 8 Urinary findings of female rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	No. of animals	Color		Cloudy		pH								Protein					Glucose				
		PY		-	+	5.0	6.0	6.5	7.0	7.5	8.0	8.5	-	±	+	++	+++	-	±	+	++	+++	
0%	6	6		6		2	2	2					4	2								6	
5%	6	6		6		2	2	2					1	5								6	
10%	6	6		6		1	3	2					5	1								6	
20%	6	6		6		2	2	2					5	1								6	

Conc. in diet	No. of animals	Ketone body					Occult blood					Urobilinogen					Bilirubin				
		-	±	+	++	+++	-	±	+	++	+++	0.1	1	2	4	8	-	+	++	+++	
0%	6	6					6					6					6				
5%	6	6					3	3				6					6				
10%	6	5	1				6					6					6				
20%	6	5	1				6					6					6				

Color : PY(pale yellow)
 Cloudy : - (negligible), + (cloudy)
 Protein : - (negligible), ±(15~30mg/dL), +(30mg/dL), ++(100mg/dL), +++(300mg/dL)
 Glucose : - (negligible), ±(0.1g/dL), +(0.25g/dL), ++(0.5g/dL), +++(1g/dL)
 Ketone body : - (negligible), ±(5mg/dL), +(15mg/dL), ++(40mg/dL), +++(80mg/dL)
 Occult blood : - (negligible), ±(trace), +(slight), ++(moderate), +++(marked)
 Urobilinogen : Ehrlich unit/dL
 Bilirubin : - (negligible), +(slight), ++(moderate), +++(marked)

Table 9 Hematological findings of male rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	No. of animals	RBC (10 ⁶ /μL)	Hb (g/dL)	Ht (%)	MCV (fL)	MCH (pg)	MCHC (%)	WBC (10 ³ /μL)	Plat. (10 ⁴ /μL)	PT (sec)	APTT (sec)
0%	6	752 ± 17	14.9 ± 0.5	42.4 ± 1.0	56 ± 1	19.8 ± 0.9	35.2 ± 0.9	60 ± 17	106 ± 11	12.9 ± 0.4	20.0 ± 1.5
5%	6	734 ± 31	14.6 ± 0.8	41.6 ± 2.3	57 ± 2	19.9 ± 0.8	35.0 ± 0.9	50 ± 7	121 ± 15	12.7 ± 0.4	19.2 ± 0.8
10%	6	760 ± 37	14.8 ± 0.6	42.2 ± 1.4	56 ± 4	19.5 ± 1.1	35.0 ± 0.7	46 ± 11	122 ± 14	13.0 ± 0.3	18.8 ± 1.2
20%	6	685 ± 136	13.4 ± 3.0	38.8 ± 7.5	57 ± 2	19.4 ± 1.1	34.3 ± 1.9	48 ± 12	122 ± 19	13.0 ± 0.2	19.3 ± 1.7

Each value is expressed as mean±S.D.

Table 10 Hematological findings of female rats fed the milk powder-contained diet in the 28-day repeat dose toxicity test

Conc. in diet	No. of animals	RBC (10 ⁶ /μL)	Hb (g/dL)	Ht (%)	MCV (fL)	MCH (pg)	MCHC (%)	PT (sec)	APTT (sec)	WBC (10 ³ /μL)
0%	6	758 ± 36	14.8 ± 0.2	41.8 ± 0.8	55 ± 2	19.6 ± 0.9	35.4 ± 0.6	13.0 ± 0.3	18.3 ± 1.1	43 ± 12
5%	6	741 ± 49	14.5 ± 0.7	41.2 ± 2.1	56 ± 2	19.6 ± 0.7	35.3 ± 0.3	13.1 ± 0.4	18.6 ± 0.6	31 ± 9
10%	6	744 ± 39	14.7 ± 0.7	41.5 ± 2.2	56 ± 2	19.8 ± 0.8	35.5 ± 0.4	13.2 ± 0.3	18.6 ± 1.2	26* ± 3
20%	6	742 ± 44	14.8 ± 0.6	41.1 ± 2.1	55 ± 2	20.0 ± 0.7	36.1 ± 0.6	13.0 ± 0.7	17.5 ± 1.0	33 ± 13

Conc. in diet	No. of animals	Differential leukocyte counts (%)								Plat. (10 ³ /μL)
		Baso.	Eosin.	Neutro.			Mono.	Other		
				Stab.	Seg.	Lymph.				
0%	6	0 ± 0	1 ± 1	0 ± 0	12 ± 4	86 ± 6	2 ± 1	0 ± 0	119 ± 16	
5%	6	0 ± 0	1 ± 1	0 ± 0	14 ± 3	84 ± 4	1 ± 1	0 ± 0	120 ± 19	
10%	6	0 ± 0	1 ± 1	0 ± 0	14 ± 4	84 ± 3	1 ± 1	0 ± 0	122 ± 12	
20%	6	0 ± 0	1 ± 1	0 ± 0	11 ± 2	87 ± 4	1 ± 1	0 ± 0	132 ± 19	

Each value is expressed as mean±S.D.

* : Significantly different from control at 5% level of probability

Table 11

Blood biochemical findings of male rats fed the milk powder-contained diet
in a 28day dose range-finding test

Conc. in diet	No. of animals	LDH (IU/L)	GOT (IU/L)	GPT (IU/L)	ALP (IU/L)	γ -GTP (IU/L)	ChE (IU/L)	CK (IU/L)	T.P. (g/dL)	Alb (%)	α_1 -G (%)	α_2 -G (%)	α_3 -G (%)	β -G (%)	γ -G (%)	A/G
0%	6	293 ± 77	71 ± 2	27 ± 2	717 ± 180	0.63 ± 0.35	54 ± 5	183 ± 24	6.16 ± 0.32	49.8 ± 1.3	20.7 ± 1.1	6.1 ± 1.1	5.4 ± 0.2	14.7 ± 1.4	3.9 ± 1.0	0.97 ± 0.05
5%	6	269 ± 81	77 ± 7	31 ± 3	663 ± 106	0.42 ± 0.14	55 ± 16	194 ± 46	5.94 ± 0.13	49.9 ± 1.2	22.1 ± 1.3	5.0 ± 0.7	5.5 ± 0.3	13.9 ± 1.1	3.7 ± 1.1	1.00 ± 0.05
10%	6	301 ± 59	80 ± 9	29 ± 5	693 ± 193	0.60 ± 0.20	60 ± 25	210 ± 25	5.87* ± 0.14	50.6 ± 1.4	21.1 ± 1.6	5.2 ± 1.0	5.6 ± 0.9	14.1 ± 1.1	3.4 ± 0.8	1.03 ± 0.06
20%	6	337 ± 137	73 ± 11	27 ± 4	543 ± 172	0.61 ± 0.37	40 ± 6	207 ± 42	6.14 ± 0.12	50.8 ± 2.7	20.7 ± 2.0	5.9 ± 0.8	4.8 ± 1.2	14.0 ± 2.1	3.9 ± 0.9	1.04 ± 0.11
Conc. in diet	No. of animals	T-Chol. (mg/dL)	T.G. (mg/dL)	PL (mg/dL)	Glu. (mg/dL)	BUN (mg/dL)	UA (mg/dL)	Crea. (mg/dL)	T-Bil. (mg/dL)	Ca (mg/dL)	P (mg/dL)	Na (mEq/L)	K (mEq/L)	Cl (mEq/L)		
0%	6	58 ± 8	71 ± 19	93 ± 12	131 ± 14	12.7 ± 1.4	0.85 ± 0.28	0.53 ± 0.06	0.27 ± 0.04	10.3 ± 0.2	7.2 ± 0.7	144 ± 1	4.47 ± 0.21	107 ± 1		
5%	6	55 ± 11	73 ± 31	89 ± 16	127 ± 8	12.8 ± 0.9	0.83 ± 0.16	0.50 ± 0.05	0.30 ± 0.03	10.2 ± 0.1	7.7 ± 0.7	144 ± 1	4.55 ± 0.22	106 ± 1		
10%	6	43 ± 11	58 ± 25	78 ± 14	134 ± 19	12.0 ± 0.6	0.86 ± 0.13	0.55 ± 0.04	0.27 ± 0.04	10.1 ± 0.2	7.5 ± 0.5	144 ± 1	4.70 ± 0.20	108 ± 2		
20%	6	55 ± 19	78 ± 24	94 ± 20	128 ± 6	10.7* ± 1.7	0.91 ± 0.24	0.52 ± 0.05	0.25 ± 0.02	10.3 ± 0.3	7.6 ± 0.5	145 ± 1	5.06** ± 0.50	108 ± 2		

Each value is expressed as mean±S.D.

* : Significantly different from control at 5% level of probability

** : Significantly different from control at 1% level of probability

Table 12

Blood biochemical findings of female rats fed the milk powder-contained diet
in a 28day dose range-finding test

Conc. in diet	No. of animals	LDH (IU/L)	GOT (IU/L)	GPT (IU/L)	ALP (IU/L)	γ -GTP (IU/L)	ChE (IU/L)	CK (IU/L)	T.P. (g/dL)	Alb (%)	α_1 -G (%)	α_2 -G (%)	α_3 -G (%)	β -G (%)	γ -G (%)	A/G
0%	6	320 ± 87	76 ± 8	25 ± 3	441 ± 137	0.39 ± 0.16	316 ± 97	171 ± 24	6.39 ± 0.28	56.2 ± 2.2	19.6 ± 1.4	5.4 ± 0.5	2.1 ± 0.3	12.6 ± 1.8	4.2 ± 0.6	1.28 ± 0.11
5%	6	453 ± 271	82 ± 15	25 ± 4	404 ± 89	0.38 ± 0.20	412 ± 125	198 ± 64	6.38 ± 0.24	56.1 ± 2.9	17.7 ± 2.6	5.8 ± 1.0	3.0 ± 0.6	12.9 ± 0.7	4.6 ± 0.3	1.29 ± 0.15
10%	6	347 ± 57	81 ± 7	23 ± 2	451 ± 62	0.35 ± 0.19	324 ± 146	185 ± 22	6.26 ± 0.38	57.2 ± 1.6	19.6 ± 1.0	4.4 ± 0.7	2.4 ± 0.4	13.0 ± 0.5	3.5 ± 1.0	1.34 ± 0.09
20%	6	430 ± 95	85 ± 14	27 ± 5	388 ± 94	0.32 ± 0.15	378 ± 105	195 ± 24	6.48 ± 0.26	56.6 ± 1.0	18.5 ± 1.2	5.5 ± 0.5	2.6 ± 1.2	12.6 ± 1.1	4.1 ± 0.4	1.31 ± 0.05
Conc. in diet	No. of animals	T-Chol. (mg/dL)	T.G. (mg/dL)	PL (mg/dL)	Glu. (mg/dL)	BUN (mg/dL)	UA (mg/dL)	Crea. (mg/dL)	T-Bil. (mg/dL)	Ca (mg/dL)	P (mg/dL)	Na (mEq/L)	K (mEq/L)	Cl (mEq/L)		
0%	6	64 ± 6	30 ± 10	111 ± 12	115 ± 9	14.4 ± 2.9	0.86 ± 0.22	0.58 ± 0.06	0.25 ± 0.01	9.9 ± 0.1	6.4 ± 0.4	146 ± 1	4.53 ± 0.24	107 ± 1		
5%	6	59 ± 16	27 ± 16	107 ± 26	120 ± 14	13.8 ± 1.4	0.94 ± 0.23	0.60 ± 0.05	0.25 ± 0.03	9.6 ± 0.2	5.7 ± 0.5	146 ± 1	4.36 ± 0.21	108 ± 2		
10%	6	52 ± 10	33 ± 9	100 ± 19	123 ± 11	13.5 ± 1.5	0.98 ± 0.25	0.61 ± 0.05	0.27 ± 0.02	9.6 ± 0.2	5.7 ± 0.7	147 ± 1	4.50 ± 0.24	108 ± 2		
20%	6	55 ± 7	25 ± 20	99 ± 14	120 ± 17	16.1 ± 2.1	0.97 ± 0.23	0.66 ± 0.07	0.27 ± 0.02	9.8 ± 0.4	6.1 ± 0.8	146 ± 1	4.49 ± 0.23	107 ± 2		

Each value is expressed as mean±S.D.

Table 13-1

Absolute and relative organ weights of male rats fed milk powder-contained diet in a 28-day dose range-finding study

	Conc. in diet (%)	No. of Animals	B.W. (g)	Brain (g)	Liver (g)	Kidney (g)	Spleen (g)	Heart (g)	Lung (g)	Thymus (g)	Thyr. (mg)	Pituit. (mg)
Absolute	0	6	312 ±12	1.89 ±0.10	8.72 ±0.69	2.37 ±0.20	0.63 ±0.07	1.09 ±0.06	1.21 ±0.10	0.62 ±0.10	21.0 ±1.9	10.0 ±1.5
	5	6	320 ±20	1.93 ±0.05	8.98 ±0.61	2.49 ±0.15	0.62 ±0.14	1.15 ±0.11	1.25 ±0.13	0.67 ±0.15	21.4 ±4.4	10.6 ±1.2
	10	6	312 ±20	1.94 ±0.06	8.65 ±0.73	2.41 ±0.17	0.58 ±0.03	1.11 ±0.10	1.18 ±0.09	0.53 ±0.06	21.3 ±2.9	10.6 ±0.8
	20	6	300 ±25	1.91 ±0.02	8.88 ±1.27	2.35 ±0.19	0.64 ±0.06	1.06 ±0.06	1.19 ±0.08	0.66 ±0.09	21.7 ±3.4	10.3 ±0.5
Relative ^{a)}	0	6	312 ±12	0.61 ±0.03	2.79 ±0.17	0.76 ±0.04	0.20 ±0.02	0.35 ±0.02	0.39 ±0.03	0.20 ±0.03	6.7 ±0.7	3.2 ±0.4
	5	6	320 ±20	0.60 ±0.04	2.81 ±0.05	0.78 ±0.06	0.19 ±0.04	0.36 ±0.01	0.39 ±0.05	0.21 ±0.05	6.7 ±1.1	3.3 ±0.4
	10	6	312 ±20	0.62 ±0.06	2.77 ±0.16	0.77 ±0.06	0.18 ±0.01	0.35 ±0.03	0.38 ±0.01	0.17 ±0.01	6.9 ±1.3	3.4 ±0.3
	20	6	300 ±25	0.64 ±0.06	2.96 ±0.33	0.79 ±0.05	0.21 ±0.03	0.35 ±0.02	0.40 ±0.02	0.22 ±0.02	7.2 ±0.9	3.5 ±0.2

Each value is expressed as mean ± S.D.

a): Relative organ weight per 100g body weight

Table 13-2

Absolute and relative organ weights of male rats fed dry beef-contained diet in a 28-day dose range-finding study

	Dose (mg/kg)	No. of Animals	B.W. (g)	Adrenal (mg)	Testis (g)	Prost. (g)	Semi.v (g)	Epidid. (g)
Absolute	0	6	312 ±12	53.7 ±5.3	2.70 ±0.24	0.49 ±0.09	1.23 ±0.18	0.77 ±0.06
	5	6	320 ±20	53.1 ±5.6	2.68 ±0.13	0.50 ±0.14	1.22 ±0.12	0.75 ±0.10
	10	6	312 ±20	52.1 ±6.1	2.69 ±0.18	0.50 ±0.05	1.30 ±0.21	0.75 ±0.04
	20	6	300 ±25	53.7 ±8.5	2.61 ±0.27	0.49 ±0.10	1.36 ±0.28	0.71 ±0.05
Relative ^{a)}	0	6	312 ±12	17.2 ±1.3	0.87 ±0.08	0.16 ±0.03	0.39 ±0.07	0.25 ±0.01
	5	6	320 ±20	16.7 ±2.1	0.84 ±0.04	0.16 ±0.04	0.38 ±0.03	0.23 ±0.02
	10	6	312 ±20	16.7 ±1.8	0.87 ±0.10	0.16 ±0.02	0.42 ±0.09	0.24 ±0.03
	20	6	300 ±25	17.9 ±2.1	0.88 ±0.10	0.17 ±0.04	0.45 ±0.09	0.24 ±0.02

Each value is expressed as mean ± S.D.

a): Relative organ weight per 100g body weight

Table 14

Absolute and relative organ weights of female rats fed milk powder-contained diet in a 28-day dose range-finding study

	Conc. in diet (%)	No. of Animals	B.W. (g)	Brain (g)	Liver (g)	Kidney (g)	Spleen (g)	Heart (g)	Lung (g)	Thymus (g)	Thyr. (mg)	Pituit. (mg)	Adrenal (mg)	Ovary (mg)	Uterus (g)
Absolute	0	6	211 ±10	1.85 ±0.10	5.89 ±0.25	1.63 ±0.14	0.42 ±0.04	0.75 ±0.06	0.98 ±0.03	0.52 ±0.10	21.0 ±2.4	12.9 ±1.2	54.9 ±6.0	78.3 ±10.7	0.47 ±0.13
	5	6	217 ±28	1.83 ±0.04	6.18 ±1.17	1.57 ±0.17	0.45 ±0.08	0.77 ±0.10	1.00 ±0.07	0.49 ±0.10	20.3 ±3.8	13.3 ±2.2	60.0 ±10.8	74.3 ±7.7	0.47 ±0.16
	10	6	211 ±14	1.82 ±0.09	5.88 ±0.60	1.64 ±0.19	0.42 ±0.04	0.76 ±0.07	0.95 ±0.03	0.46 ±0.05	20.3 ±1.9	11.8 ±1.4	52.4 ±2.9	86.1 ±17.0	0.48 ±0.10
	20	6	216 ±24	1.81 ±0.05	6.09 ±0.62	1.75 ±0.13	0.42 ±0.04	0.77 ±0.08	1.01 ±0.09	0.52 ±0.09	21.3 ±2.6	12.8 ±1.4	59.0 ±4.5	79.3 ±12.6	0.46 ±0.06
Relative ^{a)}	0	6	211 ±10	0.88 ±0.06	2.79 ±0.11	0.77 ±0.05	0.20 ±0.02	0.35 ±0.02	0.47 ±0.02	0.25 ±0.05	10.0 ±1.1	6.1 ±0.6	26.1 ±3.5	37.2 ±5.4	0.22 ±0.06
	5	6	217 ±28	0.86 ±0.10	2.84 ±0.21	0.73 ±0.06	0.21 ±0.03	0.36 ±0.02	0.47 ±0.06	0.23 ±0.03	9.5 ±1.9	6.2 ±1.0	27.8 ±5.0	34.7 ±5.6	0.22 ±0.08
	10	6	211 ±14	0.86 ±0.05	2.79 ±0.15	0.78 ±0.05	0.20 ±0.02	0.36 ±0.02	0.45 ±0.01	0.22 ±0.02	9.7 ±1.1	5.6 ±0.6	24.9 ±1.4	40.8 ±7.7	0.23 ±0.04
	20	6	216 ±24	0.84 ±0.07	2.82 ±0.10	0.81 ±0.05	0.20 ±0.03	0.36 ±0.01	0.47 ±0.02	0.25 ±0.05	9.9 ±1.3	6.0 ±0.5	27.5 ±3.5	36.8 ±5.7	0.22 ±0.04

Each value is expressed as mean ± S.D.

a) : Relative organ weight per 100g body weight

Appendix 1 Individual body weights of male rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	Number of animals	Day				
		0	7	14	21	28
0%	001	125	191	243	295	337
	002	120	183	243	290	326
	003	127	184	235	279	311
	004	130	193	249	306	345
	005	124	178	232	284	323
	006	126	190	231	285	341
	Mean		125	187	239	290
5%	007	125	193	258	324	376
	008	119	175	230	277	312
	009	124	180	237	289	333
	010	125	186	248	304	350
	011	129	178	239	296	337
	012	127	183	232	290	330
	Mean		125	183	241	297
10%	013	124	182	233	286	328
	014	126	182	250	307	353
	015	123	181	235	282	324
	016	120	171	221	260	299
	017	127	192	253	309	351
	018	129	189	243	294	339
	Mean		125	183	239	290
20%	019	125	186	237	287	331
	020	123	187	238	288	329
	021	127	192	255	309	354
	022	118	166	218	264	299
	023	124	180	236	289	327
	024	131	187	245	251	281
	Mean		125	183	238	281

Appendix 2 Individual body weights of female rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	Number of animals	Day				
		0	7	14	21	28
0%	501	114	154	179	205	231
	502	116	153	186	210	243
	503	112	144	168	194	216
	504	120	153	178	197	217
	505	110	143	173	195	211
	506	122	154	178	208	228
	Mean		116	150	177	202
5%	507	125	184	209	240	281
	508	112	144	169	198	221
	509	111	141	161	180	200
	510	118	151	174	190	208
	511	115	147	172	201	239
	512	116	151	181	208	225
	Mean		116	153	178	203
10%	513	110	139	161	188	212
	514	112	144	169	187	215
	515	116	153	185	213	232
	516	119	164	190	220	248
	517	121	156	184	207	232
	518	114	151	175	199	219
	Mean		115	151	177	202
20%	519	115	140	160	185	205
	520	110	152	183	205	225
	521	133	167	211	247	278
	522	119	161	189	211	228
	523	114	150	178	203	220
	524	112	148	173	192	216
	Mean		117	153	182	207

Appendix 3 Individual food consumption of male rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	Cage number	Week	(g/day/rat)			
			1	2	3	4
0%	1		16	21	20	20
	2		16	19	20	19
	3		16	17	19	21
	Mean		16	19	20	20
5%	4		17	20	21	20
	5		16	19	20	20
	6		14	19	20	20
	Mean		16	19	20	20
10%	7		15	19	20	20
	8		14	18	18	18
	9		17	20	21	21
	Mean		15	19	20	20
20%	10		15	18	19	19
	11		15	18	19	20
	12		15	18	14	19
	Mean		15	18	17	19

Appendix 4 Individual food consumption of female rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	Cage number	Week	(g/day/rat)			
			1	2	3	4
0%	13		12	14	16	17
	14		11	14	15	16
	15		11	14	14	14
	Mean		11	14	15	16
5%	16		13	15	19	19
	17		11	13	13	13
	18		11	15	14	16
	Mean		12	14	15	16
10%	19		10	12	13	13
	20		13	15	15	16
	21		11	13	14	15
	Mean		11	13	14	15
20%	22		10	13	14	14
	23		13	16	17	16
	24		11	12	13	14
	Mean		11	14	15	15

Appendix 5

Individual urinary findings of male rats fed the milk powder-contained diet
in a 28day dose range-finding test

Conc. in diet	Animal number	Color	Cloudy	pH	Protein	Glucose	Ketone body	Occult blood	Urobilinogen	Bilirubin
0%	001	PY	-	6.0	+	-	-	-	0.1	-
	002	PY	-	6.0	+	-	-	-	0.1	-
	003	PY	-	6.0	±	-	-	-	0.1	-
	004	PY	-	6.5	-	-	-	-	0.1	-
	005	PY	-	7.0	±	-	±	-	0.1	-
	008	PY	-	6.5	±	-	±	-	0.1	-
5%	007	PY	-	6.0	±	-	-	-	0.1	-
	008	PY	-	6.5	±	-	±	-	0.1	-
	009	PY	-	6.0	+	-	-	-	0.1	-
	010	PY	-	6.5	+	-	±	-	0.1	-
	011	PY	-	6.0	±	-	-	-	0.1	-
	012	PY	-	6.0	-	-	-	-	0.1	-
10%	013	PY	-	6.5	±	-	-	-	0.1	-
	014	PY	-	6.5	±	-	-	-	0.1	-
	015	PY	-	6.0	±	-	±	-	0.1	-
	016	PY	-	6.5	±	-	-	-	0.1	-
	017	PY	-	6.5	-	-	±	-	0.1	-
	018	PY	-	6.5	-	-	±	-	0.1	-
20%	019	PY	-	6.0	±	-	-	-	0.1	-
	020	PY	-	6.5	±	-	±	-	0.1	-
	021	PY	-	6.0	±	-	-	-	0.1	-
	022	PY	-	6.0	-	-	-	-	0.1	-
	023	PY	-	6.0	±	-	±	-	0.1	-
	024	PY	-	6.5	±	-	-	-	0.1	-

Color : PY (pale yellow)

Cloudy : - (negligible)

Protein : - (negligible), ±(15~30mg/dL), + (30mg/dL)

Glucose : - (negligible)

Ketone body : - (negligible), ±(5mg/dL)

Occult blood : - (negligible)

Urobilinogen : Ehrlich unit/dL

Bilirubin : - (negligible)

Appendix 6

Individual urinary findings of female rats fed the milk powder-contained diet
in a 28day dose range-finding test

Conc. in diet	Animal number	Color	Cloudy	pH	Protein	Glucose	Ketone body	Occult blood	Urobilinogen	Bilirubin
0%	501	PY	-	7.0	-	-	-	-	0.1	-
	502	PY	-	6.5	±	-	-	-	0.1	-
	503	PY	-	6.0	-	-	-	-	0.1	-
	504	PY	-	7.0	-	-	-	-	0.1	-
	505	PY	-	6.5	-	-	-	-	0.1	-
	508	PY	-	6.0	±	-	-	-	0.1	-
5%	507	PY	-	6.5	±	-	-	-	0.1	-
	508	PY	-	6.0	-	-	-	±	0.1	-
	509	PY	-	6.5	±	-	-	±	0.1	-
	510	PY	-	7.0	±	-	-	±	0.1	-
	511	PY	-	6.0	±	-	-	-	0.1	-
	512	PY	-	7.0	±	-	-	-	0.1	-
10%	513	PY	-	6.0	-	-	-	-	0.1	-
	514	PY	-	7.0	-	-	-	-	0.1	-
	515	PY	-	7.0	-	-	-	-	0.1	-
	516	PY	-	6.5	±	-	±	-	0.1	-
	517	PY	-	6.5	-	-	-	-	0.1	-
	518	PY	-	6.5	-	-	-	-	0.1	-
20%	519	PY	-	7.0	-	-	-	-	0.1	-
	520	PY	-	7.0	-	-	-	-	0.1	-
	521	PY	-	6.5	-	-	-	-	0.1	-
	522	PY	-	6.5	-	-	-	-	0.1	-
	523	PY	-	6.0	±	-	-	-	0.1	-
	524	PY	-	6.0	-	-	±	-	0.1	-

Color : PY (pale yellow)

Cloudy : - (negligible)

Protein : - (negligible), ±(15~30mg/dL)

Glucose : - (negligible)

Ketone body : - (negligible), ±(5mg/dL)

Occult blood : - (negligible), ±(trace)

Urobilinogen : Ehrlich unit/dL

Bilirubin : - (negligible)

Appendix 7

Individual hematological findings of male rats fed the milk powder-contained diet
in a 28day dose range finding test

Conc. in diet	Animal number	RBC (10 ⁶ /μL)	Hb (g/dL)	Ht (%)	MCV (fL)	MCH (pg)	MCHC (%)	WBC (10 ³ /μL)	Plat. (10 ⁴ /μL)	PT (sec)	APTT (sec)
0%	001	770	14.8	42.2	55	19.2	35.1	67	110	13.2	20.1
	002	764	15.5	43.7	57	20.3	35.5	61	95	12.9	22.4
	003	735	14.7	41.2	56	20.0	35.7	49	94	13.4	20.6
	004	754	14.2	41.4	55	18.8	34.3	56	124	12.3	18.2
	005	728	15.4	42.2	58	21.2	36.5	39	110	12.6	19.9
	006	762	14.8	43.5	57	19.4	34.0	90	102	12.8	18.6
	Mean	752	14.9	42.4	56	19.8	35.2	60	106	12.9	20.0
5%	007	714	13.8	39.7	56	19.3	34.8	53	148	12.5	19.1
	008	718	14.9	41.0	57	20.8	36.3	50	105	12.5	20.3
	009	739	14.2	42.1	57	19.2	33.7	61	120	12.6	18.6
	010	701	13.7	38.5	55	19.5	35.6	49	120	12.4	20.1
	011	739	15.5	44.4	60	21.0	34.9	38	124	13.4	18.2
	012	790	15.3	43.8	55	19.4	34.9	48	110	12.9	19.0
	Mean	734	14.6	41.6	57	19.9	35.0	50	121	12.7	19.2
10%	013	716	14.2	40.3	56	19.8	35.2	52	115	12.8	20.3
	014	781	15.5	42.9	55	19.8	36.1	52	118	12.7	19.1
	015	743	15.4	43.8	59	20.7	35.2	60	120	13.5	18.7
	016	822	14.2	40.6	49	17.3	35.0	28	150	12.9	18.6
	017	753	14.8	42.6	57	19.7	34.7	46	118	13.0	16.8
	018	747	14.7	43.2	58	19.7	34.0	39	110	12.8	19.5
	Mean	760	14.8	42.2	56	19.5	35.0	46	122	13.0	18.8
20%	019	749	14.7	41.3	55	19.6	35.6	52	129	12.8	20.7
	020	719	14.8	42.3	59	20.6	35.0	45	115	13.0	21.2
	021	718	14.6	42.0	58	20.3	34.8	30	129	13.1	18.4
	022	780	14.8	42.0	54	19.0	35.2	40	132	12.7	18.3
	023	734	14.4	41.6	57	19.6	34.6	57	142	13.2	20.3
	024	411	7.2	23.6	57	17.5	30.5	61	87	13.1	16.8
	Mean	685	13.4	38.8	57	19.4	34.3	48	122	13.0	19.3

Appendix 8-1

Individual hematological findings of female rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	Animal number	RBC (10 ⁶ /μL)	Hb (g/dL)	Ht (%)	MCV (fL)	MCH (pg)	MCHC (%)	PT (sec)	APTT (sec)	WBC (10 ³ /μL)
0%	501	769	14.9	41.8	54	19.4	35.6	13.3	18.3	64
	502	714	14.9	41.1	58	20.9	36.3	13.0	18.8	34
	503	797	14.7	42.3	53	18.4	34.8	12.5	19.8	39
	504	735	14.8	41.3	56	20.1	35.8	13.0	16.5	30
	505	800	15.0	43.2	54	18.8	34.7	13.2	18.3	47
	506	731	14.5	41.1	56	19.8	35.3	12.9	18.0	42
	Mean	758	14.8	41.8	55	19.6	35.4	13.0	18.3	43
5%	507	667	13.6	38.5	58	20.4	35.3	12.6	17.6	42
	508	806	15.4	43.8	54	19.1	35.2	12.8	19.3	39
	509	761	14.1	39.8	52	18.5	35.4	13.3	18.9	17
	510	748	15.1	42.2	56	20.2	35.8	12.6	18.0	30
	511	704	14.0	39.9	57	19.9	35.1	13.5	18.5	26
	512	762	15.0	42.8	56	19.7	35.0	13.5	19.0	31
	Mean	741	14.5	41.2	56	19.6	35.3	13.1	18.6	31
10%	513	795	14.9	42.3	53	18.7	35.2	12.6	18.8	27
	514	785	15.3	44.0	56	19.5	34.8	13.2	20.2	28
	515	718	14.1	39.6	56	19.8	35.6	13.5	18.4	22
	516	744	15.8	44.1	59	21.2	35.8	13.4	17.4	22
	517	699	13.9	39.2	56	19.9	35.5	13.0	17.1	27
	518	730	14.3	39.9	55	19.6	35.8	13.2	19.6	27
	Mean	744	14.7	41.5	56	19.8	35.5	13.2	18.6	26
20%	519	756	15.3	43.3	57	20.2	35.3	13.4	18.3	27
	520	687	14.4	38.7	56	21.0	37.2	12.9	18.7	37
	521	725	14.5	40.5	56	20.0	35.8	14.1	17.4	25
	522	806	15.1	42.1	52	18.7	35.9	12.6	16.4	22
	523	708	14.1	38.8	55	19.9	36.3	12.8	18.0	30
	524	772	15.6	43.1	56	20.2	36.2	11.9	16.4	57
	Mean	742	14.8	41.1	55	20.0	36.1	13.0	17.5	33

Appendix 8-2

Individual hematological findings of female rats fed the milk powder-contained diet in the 28-day repeat dose toxicity test

Conc. in diet	Animal number	Differential leukocyte counts (%)							Plat. (10 ⁴ /μL)
		Baso.	Eosin.	Neutro.		Lymph.	Mono.	Other	
				Stab.	Seg.				
0%	501	0	1	0	14	82	3	0	102
	502	0	0	0	9	89	2	0	96
	503	0	0	1	6	93	0	0	131
	504	0	0	0	14	86	0	0	129
	505	0	0	0	10	87	3	0	131
	506	0	3	0	18	76	3	0	127
	Mean	0	1	0	12	86	2	0	119
5%	507	0	0	0	12	86	2	0	90
	508	0	1	1	16	81	1	0	139
	509	0	3	0	18	78	1	0	120
	510	0	2	0	10	88	0	0	106
	511	0	1	0	14	83	2	0	127
	512	0	1	0	11	87	1	0	138
	Mean	0	1	0	14	84	1	0	120
10%	513	0	0	0	18	80	2	0	130
	514	0	1	0	10	89	0	0	128
	515	0	2	0	9	86	3	0	99
	516	0	2	0	13	84	1	0	122
	517	0	0	0	19	81	0	0	124
	518	0	2	0	12	85	1	0	131
	Mean	0	1	0	14	84	1	0	122
20%	519	0	0	0	10	90	0	0	120
	520	0	1	0	9	90	0	0	119
	521	0	2	0	15	80	3	0	147
	522	0	2	0	9	88	1	0	158
	523	0	0	0	10	90	0	0	108
	524	0	1	0	12	86	1	0	138
	Mean	0	1	0	11	87	1	0	132

Appendix 9 - 1

Individual blood biochemical findings of male rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	Animal number	LDH (U/L)	GOT (U/L)	GPT (U/L)	ALP (U/L)	γ-GTP (U/L)	CHE (U/L)	CK (U/L)	T.P. (g/dL)	Alb (%)	α ₁ -G (%)	α ₂ -G (%)	α ₃ -G (%)	β-G (%)	γ-G (%)	A/G
0%	001	314	70	27	701	0.88	52	216	6.04	47.4	20.4	6.0	5.0	16.9	4.3	0.90
	002	437	71	26	821	0.14	46	208	6.46	60.3	20.9	7.3	5.4	14.0	2.1	1.01
	003	221	69	24	936	0.59	54	174	5.91	50.3	18.9	7.3	5.5	13.3	4.7	1.01
	004	276	71	26	830	0.77	55	162	6.11	48.2	20.6	6.2	5.7	15.9	3.4	0.93
	005	241	73	30	482	0.95	53	160	5.80	49.0	22.2	5.0	5.3	14.3	4.2	0.96
	006	271	69	26	530	1.07	61	175	6.63	60.5	20.9	4.6	5.3	13.9	4.8	1.02
	Mean	293	71	27	717	0.63	54	183	6.16	49.3	20.7	6.1	5.4	14.7	3.9	0.97
5%	007	402	86	33	773	0.51	40	276	6.04	60.7	22.9	4.0	5.3	12.5	4.6	1.03
	008	197	78	32	610	0.44	51	175	6.04	51.2	21.6	5.0	5.1	13.0	4.1	1.05
	009	315	69	30	748	0.34	56	218	5.92	48.2	20.5	5.7	5.7	15.8	4.6	0.93
	010	280	84	34	606	0.64	81	181	6.04	50.2	24.3	4.3	5.8	13.7	1.7	1.01
	011	209	71	29	504	0.29	36	168	5.84	60.3	21.2	5.4	5.3	14.9	2.9	1.01
	012	208	73	25	734	0.27	64	157	5.73	48.5	22.0	5.7	5.9	13.8	4.1	0.94
	Mean	269	77	31	663	0.42	55	194	5.94	49.9	22.1	5.0	5.5	13.9	3.7	1.00
10%	013	330	84	33	522	0.64	39	220	5.80	51.4	21.5	4.5	5.1	13.3	4.2	1.03
	014	331	88	29	834	0.25	95	214	5.90	52.2	22.5	3.8	4.7	13.2	3.6	1.09
	015	309	80	32	953	0.64	86	247	5.83	50.8	21.9	4.8	5.9	13.0	3.6	1.08
	016	291	87	34	803	0.72	51	189	6.10	48.4	21.7	5.4	6.2	14.5	3.8	0.94
	017	355	73	25	510	0.83	38	213	5.90	49.5	18.0	6.3	6.9	16.0	3.3	0.98
	018	190	65	22	537	0.51	50	177	5.67	51.5	21.0	6.3	4.7	14.5	2.0	1.06
	Mean	301	80	29	693	0.60	60	210	5.87	50.6	21.1	5.2	5.6	14.1	3.4	1.03
20%	019	477	78	31	475	0.48	49	242	6.04	49.9	21.9	6.8	4.5	13.0	3.9	1.00
	020	323	71	28	747	0.39	42	188	5.98	60.1	19.4	6.5	5.1	14.8	4.1	1.00
	021	177	65	25	686	0.45	35	153	6.14	54.0	21.1	5.4	3.8	12.7	3.0	1.17
	022	321	72	32	453	0.67	32	220	6.26	62.6	23.1	4.6	3.7	11.6	4.4	1.11
	023	211	59	22	613	0.33	40	176	6.29	46.3	17.5	6.3	7.0	17.7	5.2	0.86
	024	515	90	26	288	1.32	40	264	6.12	52.0	21.0	5.7	4.4	14.1	2.8	1.08
	Mean	337	73	27	543	0.61	40	207	6.14	50.8	20.7	5.9	4.8	14.0	3.9	1.04

Appendix 9 - 2

Individual blood biochemical findings of male rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	Animal number	T-Chol. (mg/dL)	T.G. (mg/dL)	PL (mg/dL)	Glu. (mg/dL)	BUN (mg/dL)	UA (mg/dL)	Crea. (mg/dL)	T-Bil. (mg/dL)	Ca (mg/dL)	P (mg/dL)	Na (mEq/L)	K (mEq/L)	Cl (mEq/L)
0%	001	68	74	112	146	13.3	1.20	0.55	0.29	10.2	6.8	144	4.31	106
	002	52	76	92	146	14.4	1.16	0.64	0.22	10.5	7.3	145	4.26	106
	003	55	88	93	118	11.9	0.87	0.55	0.26	10.2	7.3	144	4.28	107
	004	51	63	81	132	13.3	0.68	0.52	0.22	10.2	7.8	145	4.61	108
	005	53	36	79	113	10.3	0.55	0.46	0.29	10.1	7.5	144	4.59	107
	006	66	86	98	132	12.9	0.64	0.48	0.32	10.6	7.7	144	4.74	106
	Mean	58	71	93	131	12.7	0.85	0.53	0.27	10.3	7.2	144	4.47	107
5%	007	66	125	109	122	12.0	1.05	0.54	0.31	10.2	7.0	143	4.62	106
	008	46	71	81	119	13.3	0.76	0.54	0.33	10.2	7.0	144	4.37	106
	009	66	86	106	122	11.4	0.93	0.54	0.27	10.0	7.2	143	4.71	106
	010	53	72	89	132	12.7	0.93	0.48	0.26	10.2	8.3	142	4.84	106
	011	40	38	66	127	14.0	0.68	0.42	0.31	10.3	7.9	146	4.24	107
	012	58	48	91	141	13.2	0.68	0.46	0.33	10.3	8.6	144	4.54	107
	Mean	55	73	89	127	12.8	0.83	0.50	0.30	10.2	7.7	144	4.55	106
10%	013	62	53	97	106	12.2	0.85	0.56	0.29	10.1	7.3	145	4.61	110
	014	52	98	90	137	11.8	0.78	0.61	0.24	10.2	6.9	144	4.48	107
	015	39	76	84	125	11.5	1.05	0.55	0.30	10.1	7.3	143	4.69	107
	016	35	31	66	158	12.3	0.79	0.50	0.21	9.7	7.5	144	5.01	110
	017	32	43	63	152	12.8	0.98	0.55	0.30	10.2	8.2	143	4.87	105
	018	39	44	68	123	11.3	0.69	0.50	0.27	10.2	8.0	144	4.55	108
	Mean	43	58	78	134	12.0	0.86	0.55	0.27	10.1	7.5	144	4.70	108
20%	019	47	71	83	131	12.7	0.92	0.81	0.22	10.4	8.0	145	5.22	106
	020	30	40	60	130	10.1	0.80	0.47	0.24	10.1	7.0	143	5.19	108
	021	61	66	100	130	10.8	0.79	0.47	0.29	10.4	7.9	145	4.72	105
	022	59	104	110	125	12.6	0.74	0.53	0.25	10.3	7.5	145	4.75	108
	023	48	96	93	135	8.6	0.80	0.50	0.24	10.7	6.9	145	4.55	108
	024	87	91	115	118	9.1	1.39	0.52	0.23	9.9	8.2	145	5.93	111
	Mean	55	78	94	128	10.7	0.91	0.52	0.25	10.3	7.6	145	5.06	108

Individual blood biochemical findings of female rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	Animal number	LDH (U/L)	GOT (U/L)	GPT (U/L)	ALP (U/L)	γ-GTP (U/L)	ChE (U/L)	CK (U/L)	T.P. (g/dL)	Alb (%)	α ₁ -G (%)	α ₂ -G (%)	α ₃ -G (%)	β-G (%)	γ-G (%)	A/G
0%	501	343	83	25	480	0.31	390	166	6.66	56.4	19.8	4.8	2.2	12.6	4.2	1.29
	502	300	66	21	330	0.68	199	156	6.44	57.5	18.9	5.6	2.2	11.6	4.3	1.36
	503	479	84	27	467	0.34	370	196	6.32	56.7	17.7	5.9	2.2	12.8	4.7	1.31
	504	239	81	26	648	0.43	189	168	6.37	52.9	19.1	5.4	1.9	16.0	4.7	1.12
	505	303	73	27	468	0.33	337	205	5.90	54.4	21.9	5.9	2.3	11.5	4.0	1.19
	506	253	67	21	252	0.24	410	147	6.64	59.1	20.2	5.0	1.6	10.9	3.2	1.44
Mean	320	76	25	441	0.39	316	171	6.39	56.2	19.6	5.4	2.1	12.6	4.2	1.28	
5%	507	293	68	26	412	0.38	642	146	6.57	57.2	17.9	4.7	3.0	12.4	4.8	1.34
	508	567	85	31	432	0.27	443	209	6.41	57.3	17.4	5.9	2.6	12.3	4.5	1.34
	509	236	79	22	396	0.53	400	117	6.70	51.7	20.4	7.3	3.9	12.3	4.4	1.07
	510	312	72	20	552	0.20	297	184	6.38	53.1	19.9	6.0	2.7	13.5	4.8	1.13
	511	355	75	23	329	0.20	370	241	6.15	58.8	13.0	5.9	3.5	13.9	4.9	1.43
	512	956	110	29	300	0.71	318	293	6.06	58.3	17.3	4.8	2.3	13.1	4.2	1.40
Mean	453	82	25	404	0.38	412	198	6.38	56.1	17.7	5.8	3.0	12.9	4.6	1.29	
10%	513	402	75	24	368	0.01	234	200	6.23	58.9	18.2	4.0	2.5	13.7	2.7	1.43
	514	252	71	21	467	0.35	161	154	6.02	55.2	19.9	4.8	2.8	12.5	4.8	1.23
	515	381	81	24	468	0.31	420	198	6.18	56.1	20.9	3.7	2.8	13.2	3.3	1.28
	516	392	87	25	392	0.41	217	201	5.75	57.6	19.5	3.7	2.1	13.4	3.7	1.36
	517	320	89	22	539	0.51	368	159	6.54	59.0	18.8	5.5	1.9	12.7	2.1	1.44
	518	334	81	24	471	0.51	545	195	6.84	56.4	20.1	4.4	2.1	12.7	4.3	1.29
Mean	347	81	23	451	0.35	324	185	6.26	57.2	19.6	4.4	2.4	13.0	3.5	1.34	
20%	519	370	99	27	404	0.36	369	198	6.27	55.6	18.8	5.7	4.1	11.6	4.2	1.26
	520	334	73	24	245	0.37	352	153	6.94	56.8	19.0	5.6	1.8	12.3	4.5	1.31
	521	412	99	35	357	0.46	206	187	6.23	57.2	17.3	4.7	2.0	14.6	4.2	1.34
	522	608	96	27	534	0.08	425	220	6.35	58.2	19.2	5.9	1.3	11.9	3.5	1.39
	523	423	67	20	371	0.19	526	218	6.58	55.9	20.0	5.1	2.2	12.8	4.0	1.27
	524	432	78	26	416	0.45	392	192	6.51	55.9	16.8	6.2	4.2	12.5	4.4	1.27
Mean	430	85	27	388	0.32	378	195	6.48	56.6	18.5	5.5	2.6	12.6	4.1	1.31	

Individual blood biochemical findings of female rats fed the milk powder-contained diet in a 28day dose range-finding test

Conc. in diet	Animal number	T-Chol. (mg/dL)	T.G. (mg/dL)	PL (mg/dL)	Glu. (mg/dL)	BUN (mg/dL)	UA (mg/dL)	Crea. (mg/dL)	T-Bil. (mg/dL)	Ca (mg/dL)	P (mg/dL)	Na (mEq/L)	K (mEq/L)	Cl (mEq/L)
0%	501	65	29	115	125	19.6	0.98	0.68	0.24	9.8	5.9	147	4.29	107
	502	65	38	118	124	15.2	0.96	0.58	0.27	10.1	6.2	146	4.27	107
	503	72	22	125	110	14.4	1.17	0.60	0.26	9.7	6.3	145	4.62	108
	504	60	16	106	101	11.6	0.57	0.53	0.24	9.8	6.9	145	4.79	108
	505	56	44	89	111	13.8	0.80	0.53	0.25	9.9	6.9	145	4.78	105
	506	67	28	114	120	11.9	0.68	0.55	0.24	9.8	6.0	147	4.40	108
Mean	64	30	111	115	14.4	0.86	0.58	0.25	9.9	6.4	146	4.53	107	
5%	507	83	33	144	145	15.3	0.83	0.52	0.23	9.8	5.5	145	4.01	106
	508	74	56	130	120	14.3	1.19	0.63	0.27	9.8	5.5	146	4.31	107
	509	53	15	100	112	12.8	0.55	0.57	0.23	9.7	5.6	146	4.38	108
	510	54	20	103	121	15.4	0.94	0.63	0.30	9.7	5.0	147	4.41	111
	511	43	25	80	111	13.0	1.02	0.63	0.21	9.4	5.9	145	4.68	107
	512	49	11	83	108	12.0	1.10	0.51	0.26	9.3	6.6	146	4.38	109
Mean	59	27	107	120	13.8	0.94	0.60	0.25	9.6	5.7	146	4.36	108	
10%	513	61	42	113	135	14.3	1.21	0.55	0.29	9.8	5.1	147	4.44	106
	514	37	25	78	119	13.8	1.27	0.61	0.27	9.5	5.6	148	4.40	110
	515	56	35	108	124	14.6	0.87	0.59	0.28	9.6	6.3	146	4.86	107
	516	47	33	82	114	10.9	0.97	0.52	0.28	9.2	4.9	147	4.23	111
	517	64	40	128	137	12.6	0.59	0.55	0.24	9.7	6.6	146	4.71	108
	518	49	20	93	109	14.7	0.94	0.61	0.28	9.7	5.9	148	4.34	107
Mean	52	33	100	123	13.5	0.98	0.61	0.27	9.6	5.7	147	4.50	108	
20%	519	51	21	87	104	15.1	1.16	0.57	0.27	9.3	5.5	148	4.31	109
	520	54	19	101	107	18.3	0.89	0.78	0.28	9.8	5.5	146	4.24	106
	521	52	12	85	134	18.5	1.08	0.68	0.28	9.4	5.7	145	4.58	106
	522	48	16	90	112	12.8	1.22	0.61	0.27	9.9	5.6	148	4.64	109
	523	57	66	117	115	16.0	0.60	0.62	0.25	10.4	7.1	146	4.41	105
	524	67	17	116	147	16.0	0.85	0.68	0.24	9.9	7.1	145	4.25	107
Mean	55	25	99	120	16.1	0.97	0.66	0.27	9.8	6.1	146	4.49	107	

Conc. in diet(%)	Animal numbers	B.W. (g)	Brain (g)	Liver (g)	Kidney (g)	Spleen (g)	Heart (g)	Lung (g)	Thymus (g)	Thyr. (mg)	Pitui. (mg)	Adrenal (mg)	Testis (g)	Prost. (g)	Semi.v (g)	Epidid. (g)
0	001	317	1.89	9.13	2.37	0.70	1.13	1.29	0.61	23.3	9.6	62.4	2.46	0.62	1.02	0.76
	002	306	1.85	9.36	2.27	0.56	1.02	1.20	0.73	19.5	8.8	50.0	2.61	0.51	1.47	0.76
	003	295	1.85	7.88	2.15	0.67	1.10	1.09	0.45	20.8	8.7	47.2	2.68	0.55	1.28	0.71
	004	325	1.81	9.11	2.43	0.69	1.14	1.31	0.68	19.6	12.7	54.4	2.56	0.45	1.15	0.79
	005	306	1.88	7.79	2.26	0.55	1.02	1.28	0.66	23.4	9.4	52.3	2.72	0.44	1.39	0.71
	006	323	2.08	9.02	2.73	0.58	1.15	1.09	0.56	19.1	10.7	56.0	3.16	0.36	1.08	0.88
	Mean		312	1.89	8.72	2.37	0.63	1.09	1.21	0.62	21.0	10.0	53.7	2.70	0.49	1.23
5	007	356	1.96	10.03	2.44	0.75	1.33	1.26	0.72	29.5	10.9	50.3	2.87	0.73	1.31	0.88
	008	296	1.97	8.30	2.41	0.66	1.03	1.17	0.90	21.1	9.9	53.4	2.56	0.34	1.14	0.64
	009	315	1.84	9.08	2.27	0.54	1.10	1.14	0.49	17.9	8.6	51.7	2.53	0.44	1.09	0.79
	010	327	1.94	9.14	2.70	0.80	1.20	1.21	0.74	17.8	11.9	61.0	2.64	0.58	1.40	0.76
	011	315	1.96	8.57	2.54	0.52	1.11	1.50	0.66	19.2	11.6	44.9	2.71	0.50	1.24	0.79
	012	311	1.90	8.73	2.59	0.45	1.10	1.22	0.53	23.0	10.8	57.4	2.78	0.40	1.12	0.62
	Mean		320	1.93	8.98	2.49	0.62	1.15	1.25	0.67	21.4	10.6	53.1	2.68	0.50	1.22
10	013	312	1.93	8.02	2.35	0.56	1.04	1.19	0.55	23.9	10.2	44.6	2.77	0.55	1.27	0.70
	014	331	1.87	9.18	2.32	0.57	1.19	1.19	0.56	17.4	9.7	60.7	2.38	0.43	1.11	0.71
	015	303	1.88	7.97	2.23	0.56	1.19	1.15	0.54	21.6	10.2	57.7	2.85	0.56	1.60	0.80
	016	279	2.01	8.23	2.36	0.54	0.99	1.04	0.40	25.2	10.4	47.9	2.78	0.45	1.44	0.79
	017	330	1.92	9.81	2.72	0.63	1.21	1.31	0.57	20.5	11.5	49.4	2.76	0.53	1.35	0.72
	018	319	2.00	8.69	2.50	0.61	1.03	1.18	0.55	19.3	11.6	52.0	2.57	0.50	1.05	0.76
	Mean		312	1.94	8.65	2.41	0.58	1.11	1.18	0.53	21.3	10.6	52.1	2.69	0.50	1.30
20	019	308	1.94	8.28	2.28	0.62	1.13	1.30	0.72	25.5	10.0	65.2	2.12	0.49	1.14	0.68
	020	309	1.88	8.50	2.38	0.69	1.08	1.21	0.71	22.6	10.7	54.3	2.65	0.37	1.89	0.75
	021	334	1.91	9.66	2.71	0.57	1.11	1.22	0.72	20.6	11.1	56.3	2.97	0.51	1.31	0.68
	022	278	1.91	8.33	2.17	0.56	1.02	1.05	0.51	19.8	10.3	41.2	2.64	0.60	1.43	0.76
	023	307	1.88	11.04	2.26	0.68	1.00	1.19	0.70	25.3	10.2	58.1	2.67	0.60	1.23	0.74
	024	265	1.93	7.48	2.31	0.71	0.99	1.14	0.61	16.6	9.7	46.8	2.63	0.37	1.16	0.64
	Mean		300	1.91	8.88	2.35	0.64	1.06	1.19	0.66	21.7	10.3	53.7	2.61	0.49	1.36

Conc. in diet(%)	Animal numbers	B.W. (g)	Brain (%)	Liver (%)	Kidney (%)	Spleen (%)	Heart (%)	Lung (%)	Thymus (%)	Thyr. (mg%)	Pitui. (mg%)	Adrenal (mg%)	Testis (%)	Prost. (%)	Semi.v (%)	Epidid. (%)
0	001	317	0.60	2.88	0.75	0.22	0.36	0.41	0.19	7.4	3.0	19.7	0.78	0.20	0.32	0.24
	002	306	0.60	3.06	0.74	0.18	0.33	0.39	0.24	6.4	2.9	16.3	0.85	0.17	0.48	0.25
	003	295	0.63	2.67	0.73	0.23	0.37	0.37	0.15	7.1	2.9	16.0	0.91	0.19	0.43	0.24
	004	325	0.56	2.80	0.75	0.21	0.35	0.40	0.21	6.0	3.9	16.7	0.79	0.14	0.35	0.24
	005	306	0.61	2.55	0.74	0.18	0.33	0.42	0.22	7.6	3.1	17.1	0.89	0.14	0.45	0.23
	006	323	0.64	2.79	0.85	0.18	0.36	0.34	0.17	5.9	3.3	17.3	0.98	0.11	0.33	0.27
	Mean		312	0.61	2.79	0.76	0.20	0.35	0.39	0.20	6.7	3.2	17.2	0.87	0.16	0.39
5	007	356	0.55	2.82	0.69	0.21	0.37	0.35	0.20	8.3	3.1	14.1	0.81	0.21	0.37	0.25
	008	296	0.67	2.80	0.81	0.22	0.35	0.40	0.30	7.1	3.3	18.0	0.86	0.11	0.39	0.22
	009	315	0.58	2.88	0.72	0.17	0.35	0.36	0.16	5.7	2.7	16.4	0.80	0.14	0.35	0.25
	010	327	0.59	2.80	0.83	0.24	0.37	0.37	0.23	5.4	3.6	18.7	0.81	0.18	0.43	0.23
	011	315	0.62	2.72	0.81	0.17	0.35	0.48	0.21	6.1	3.7	14.3	0.86	0.16	0.39	0.25
	012	311	0.61	2.81	0.83	0.14	0.35	0.39	0.17	7.4	3.5	18.5	0.89	0.13	0.36	0.20
	Mean		320	0.60	2.81	0.78	0.19	0.36	0.39	0.21	6.7	3.3	16.7	0.84	0.16	0.38
10	013	312	0.62	2.57	0.75	0.18	0.33	0.38	0.18	7.7	3.3	14.3	0.89	0.18	0.41	0.22
	014	331	0.56	2.77	0.70	0.17	0.36	0.36	0.17	5.3	2.9	18.3	0.72	0.13	0.34	0.21
	015	303	0.62	2.63	0.74	0.18	0.39	0.38	0.18	7.1	3.4	19.0	0.94	0.18	0.53	0.26
	016	279	0.72	2.95	0.85	0.19	0.35	0.37	0.14	9.0	3.7	17.2	1.00	0.16	0.52	0.28
	017	330	0.58	2.97	0.82	0.19	0.37	0.40	0.17	6.2	3.5	15.0	0.84	0.16	0.41	0.22
	018	319	0.63	2.72	0.78	0.19	0.32	0.37	0.17	6.1	3.6	16.3	0.81	0.16	0.33	0.24
	Mean		312	0.62	2.77	0.77	0.18	0.35	0.38	0.17	6.9	3.4	16.7	0.87	0.16	0.42
20	019	308	0.63	2.69	0.74	0.20	0.37	0.42	0.23	8.3	3.2	21.2	0.69	0.16	0.37	0.22
	020	309	0.61	2.75	0.77	0.22	0.35	0.39	0.23	7.3	3.5	17.6	0.86	0.12	0.61	0.24
	021	334	0.57	2.89	0.81	0.17	0.33	0.37	0.22	6.2	3.3	16.9	0.89	0.15	0.39	0.28
	022	278	0.69	3.00	0.78	0.20	0.37	0.38	0.18	7.1	3.7	14.8	0.95	0.22	0.51	0.27
	023	307	0.61	3.60	0.74	0.22	0.33	0.39	0.23	8.2	3.3	18.9	0.87	0.20	0.40	0.24
	024	265	0.73	2.82	0.87	0.27	0.37	0.43	0.23	6.3	3.7	17.7	0.99	0.14	0.44	0.24
	Mean		300	0.64	2.96	0.79	0.21	0.35	0.40	0.22	7.2	3.5	17.9	0.88	0.17	0.45

Conc. in diet (%)	Animal numbers	B.W. (g)	Brain (g)	Liver (g)	Kidney (g)	Spleen (g)	Heart (g)	Lung (g)	Thymus (g)	Thyr. (mg)	Pitui. (mg)	Adrenal (mg)	Ovary (mg)	Uterus (g)
0	501	218	1.92	5.91	1.61	0.46	0.76	0.96	0.46	25.7	14.4	50.2	76.3	0.53
	502	226	1.88	6.23	1.88	0.42	0.85	1.03	0.67	19.1	12.9	59.9	89.1	0.44
	503	201	2.01	6.05	1.64	0.45	0.75	0.97	0.49	19.4	13.4	60.1	91.8	0.47
	504	209	1.78	5.76	1.64	0.35	0.70	0.98	0.42	20.1	13.4	51.9	63.8	0.38
	505	199	1.74	5.50	1.52	0.44	0.69	0.95	0.61	21.5	12.2	60.4	78.0	0.31
	506	212	1.77	5.86	1.47	0.40	0.73	0.99	0.46	20.4	10.8	46.6	70.6	0.69
	Mean	211	1.85	5.89	1.63	0.42	0.75	0.98	0.52	21.0	12.9	54.9	78.3	0.47
5	507	265	1.82	8.51	1.88	0.58	0.95	1.09	0.55	19.2	17.1	71.7	77.9	0.63
	508	211	1.87	5.96	1.47	0.49	0.79	1.02	0.57	17.8	12.2	64.3	80.9	0.46
	509	186	1.79	5.39	1.47	0.37	0.67	1.02	0.33	17.3	13.8	63.3	70.6	0.66
	510	196	1.87	5.53	1.57	0.43	0.67	1.04	0.44	23.2	13.9	44.0	80.1	0.40
	511	227	1.83	6.05	1.42	0.37	0.75	0.90	0.58	26.7	11.0	49.3	60.5	0.24
	512	215	1.78	5.63	1.59	0.43	0.81	0.92	0.48	17.8	11.7	67.2	75.5	0.42
	Mean	217	1.83	6.18	1.57	0.45	0.77	1.00	0.49	20.3	13.3	60.0	74.3	0.47
10	513	193	1.70	5.26	1.41	0.44	0.70	0.91	0.51	22.4	9.9	52.2	60.8	0.53
	514	201	1.79	5.49	1.56	0.39	0.67	0.93	0.39	17.8	11.1	47.9	106.9	0.37
	515	218	1.93	5.86	1.80	0.46	0.79	0.97	0.47	22.5	11.2	50.7	76.2	0.43
	516	232	1.77	6.54	1.91	0.39	0.88	1.01	0.50	20.5	12.4	55.7	97.1	0.65
	517	217	1.92	6.69	1.52	0.48	0.75	0.95	0.49	19.6	14.1	55.1	96.6	0.45
	518	204	1.79	5.44	1.64	0.37	0.79	0.94	0.42	19.1	12.2	52.7	79.1	0.47
	Mean	211	1.82	5.88	1.64	0.42	0.76	0.95	0.46	20.3	11.8	52.4	86.1	0.48
20	519	198	1.76	5.64	1.64	0.44	0.69	0.99	0.63	20.9	10.9	65.6	64.2	0.56
	520	214	1.83	6.22	1.81	0.37	0.76	1.00	0.58	20.6	13.9	53.3	101.5	0.40
	521	263	1.89	7.29	1.93	0.43	0.91	1.18	0.55	23.4	14.6	61.7	84.4	0.40
	522	211	1.74	5.69	1.64	0.37	0.73	1.00	0.52	24.8	11.9	58.9	74.9	0.46
	523	211	1.81	5.78	1.85	0.42	0.77	1.00	0.38	17.1	12.6	55.0	74.2	0.48
	524	201	1.80	5.92	1.62	0.48	0.73	0.89	0.48	20.9	13.2	59.7	76.6	0.45
	Mean	216	1.81	6.09	1.75	0.42	0.77	1.01	0.52	21.3	12.8	59.0	79.3	0.46

Conc. in diet (%)	Animal numbers	B.W. (g)	Brain (%)	Liver (%)	Kidney (%)	Spleen (%)	Heart (%)	Lung (%)	Thymus (%)	Thyr. (mg%)	Pitui. (mg%)	Adrenal (mg%)	Ovary (mg%)	Uterus (%)
0	501	218	0.88	2.71	0.74	0.21	0.35	0.44	0.21	11.8	6.6	23.0	35.0	0.24
	502	226	0.83	2.76	0.83	0.19	0.38	0.46	0.30	8.5	5.7	26.5	39.4	0.19
	503	201	1.00	3.01	0.82	0.22	0.37	0.48	0.24	9.7	6.7	29.9	45.7	0.23
	504	209	0.85	2.76	0.78	0.17	0.33	0.47	0.20	9.6	6.4	24.8	30.5	0.18
	505	199	0.87	2.76	0.76	0.22	0.35	0.48	0.31	10.8	6.1	30.4	39.2	0.16
	506	212	0.83	2.76	0.69	0.19	0.34	0.47	0.22	9.6	5.1	22.0	33.3	0.33
	Mean	211	0.88	2.79	0.77	0.20	0.35	0.47	0.25	10.0	6.1	26.1	37.2	0.22
5	507	265	0.69	3.21	0.71	0.22	0.36	0.41	0.21	7.2	6.5	27.1	29.4	0.24
	508	211	0.89	2.82	0.70	0.23	0.37	0.48	0.27	8.4	5.8	30.5	38.3	0.22
	509	186	0.96	2.90	0.79	0.20	0.36	0.55	0.18	9.3	7.4	34.0	38.0	0.35
	510	196	0.95	2.82	0.80	0.22	0.34	0.53	0.22	11.8	7.1	22.4	40.9	0.20
	511	227	0.81	2.67	0.63	0.16	0.33	0.40	0.26	11.8	4.8	21.7	26.7	0.11
	512	215	0.83	2.62	0.74	0.20	0.38	0.43	0.22	8.3	5.4	31.3	35.1	0.20
	Mean	217	0.86	2.84	0.73	0.21	0.36	0.47	0.23	9.5	6.2	27.8	34.7	0.22
10	513	193	0.88	2.73	0.73	0.23	0.36	0.47	0.26	11.6	5.1	27.0	31.5	0.27
	514	201	0.89	2.73	0.78	0.19	0.33	0.46	0.19	8.9	5.5	23.8	53.2	0.18
	515	218	0.89	2.69	0.83	0.21	0.36	0.44	0.22	10.3	5.1	23.3	35.0	0.20
	516	232	0.76	2.82	0.82	0.17	0.38	0.44	0.22	8.8	5.3	24.0	41.9	0.28
	517	217	0.88	3.08	0.70	0.22	0.35	0.44	0.23	9.0	6.5	25.4	44.5	0.21
	518	204	0.88	2.67	0.80	0.18	0.39	0.46	0.21	9.4	6.0	25.8	38.8	0.23
	Mean	211	0.86	2.79	0.78	0.20	0.36	0.45	0.22	9.7	5.6	24.9	40.8	0.23
20	519	198	0.89	2.85	0.83	0.22	0.35	0.50	0.32	10.6	5.4	33.1	32.4	0.28
	520	214	0.86	2.91	0.85	0.17	0.36	0.47	0.27	9.6	6.5	24.9	47.4	0.19
	521	263	0.72	2.77	0.73	0.16	0.35	0.45	0.21	8.9	5.6	23.5	32.1	0.15
	522	211	0.82	2.70	0.78	0.18	0.35	0.47	0.25	11.8	5.6	27.9	35.5	0.22
	523	211	0.86	2.74	0.88	0.20	0.36	0.47	0.18	8.1	6.0	26.1	35.2	0.23
	524	201	0.90	2.95	0.81	0.24	0.36	0.44	0.24	10.4	6.6	29.7	38.1	0.22
	Mean	216	0.84	2.82	0.81	0.20	0.36	0.47	0.25	9.9	6.0	27.5	36.8	0.22

Table 1 Analyzed nutrient composition of the dried beef

Macronutrient (%)		Vitamin (mg/100g)		Mineral (mg/100g)	
Crude protein	42.8	A	ND	Ca	17.1
Crude fat	50.0	B ₁	0.23	P	386
Carbohydrate	4.3	B ₂	0.50	K	1120
Crude fiber	0.0	B ₆	0.54	Na	767
Ash	2.0	B ₁₂	0.0031	Mg	47.3
Water	0.9	D ₃	ND	Fe	6.26
		E	0.2	Zn	9.99
		K ₁	ND	Cu	0.14
		K ₂	0.010	S	370
		Niacin	12.1	Mn	0.03
		Pantotheinic acid	2.55	I	ND
		Folic acid	0.019	Se	0.03
		Biotin	0.0042	Mo	ND
		Choline	0.13 (%)		

Table 2-1 Composition of the dried beef-contained diets

Ingredient	(g/kg diets)				
	Basal diet I	5% diet	10% diet	Basal diet II	20% diet
Dried beef		50.000	100.000		200.000
Cornstarch	397.486	396.019	394.552	374.965	369.097
Casein (93.5% protein)	200.000	177.112	154.225	200.000	108.449
Alfa-cornstarch	132.000	131.513	131.027	124.521	122.573
Sucrose	100.000	100.000	100.000	100.000	100.000
Soybean oil	70.000	45.000	20.000	100.000	
Cellulose	50.000	50.000	50.000	50.000	50.000
Mineral Mix *	35.000	35.000	35.000	35.000	35.000
Vitamin Mix *	10.000	10.000	10.000	10.000	10.000
L-Cystine	3.000	3.000	3.000	3.000	3.000
Choline Bitartrate (41.1% choline)	2.500	2.342	2.184	2.500	1.867
t-Butylhydroquinone	0.014	0.014	0.014	0.014	0.014
Caloric value (kcal)	397	398	399	411	415
Crude protein (%)	18.7	18.7	18.7	18.7	18.7
Crude fat (%)	7.0	7.0	7.0	10.0	10.0
Carbohydrate (%)	64.7	65.0	65.2	61.5	62.5
Crude fiber (%)	5.0	5.0	5.0	5.0	5.0

*: Ingredients of the mineral mix and vitamin mix are shown in table 2-2.

Table 2-2 Ingredients of the mineral mix and vitamin mix for the dried beef-contained diet

Mineral Mix							Vitamin Mix				
Ingredients	Contents (%)	Basal diet I	5% diet	10% diet	Basal diet II	20% diet	Ingredients	Basal diet I/II	5% diet	10% diet	20% diet
ESSENTIAL MINERALS							Vitamine A Palmitate (500,000IU/g)	0.800	0.800	0.800	0.800
Calcium Carbonate	Ca: 40.04	357.00	356.357	355.786	357.00	354.572	Thiamine HCl (B ₁)	0.600	0.590	0.580	0.550
Potassium Phosphate (monobasic)	P: 22.78	196.00	192.359	188.843	196.00	181.688	Riboflavin (B ₂)	0.600	0.580	0.550	0.500
Potassium Citrate H ₂ O	K: 28.73	57.29*	51.612	45.832	122.844*		Pyridoxine HCl (B ₆)	0.700	0.670	0.650	0.590
Sodium Chloride	Na: 39.34	74.00	45.584	17.242	114.00*		Vitamine B ₁₂ (0.1%)	2.500	2.300	2.200	1.900
Potassium Sulfate	S: 18.39	57.47*	28.735		115.00*		Vitamine D ₃ (400,000IU/g)	0.250	0.250	0.250	0.250
Magnesium Oxide	Mg: 44.87	24.00	22.871	21.788	24.00	19.529	Vitamine E Acetate (500IU/g)	15.000	15.000	15.000	15.000
Ferric Citrate	Fe: 16.50	6.08	5.541	5.021	6.08	3.809	Vitamine K ₁	0.075	0.075	0.074	0.073
Zinc Carbonate	Zn: 52.14	1.65	1.375	1.100	1.65	0.550	Niacin	3.000	3.000	3.000	3.000
Manganous Carbonate	Mn: 47.79	0.63	0.630	0.630	0.63	0.620	Calcium Pantothenate	1.600	1.500	1.300	1.100
Cupric Carbonate	Cu: 57.47	0.30	0.295	0.295	0.30	0.285	Folic Acid	0.200	0.200	0.200	0.200
Potassium Iodate	I: 59.30	0.01	0.01	0.01	0.01	0.01	Biotin	0.020	0.020	0.019	0.018
Sodium Selenate	Se: 41.79	0.01025	0.00957	0.00820	0.01025	0.00615	Sucrose finely powdered	974.655	975.015	975.376	976.018
Ammonium Paramolybdate 4H ₂ O	Mo: 54.34	0.00795	0.00795	0.00795	0.00795	0.00795					
NON-ESSENTIAL MINERALS											
Sodium Metaalicate 9H ₂ O	Si: 9.88	1.45	1.45	1.45	1.45	1.45					
Chromium Potassium Sulfate 12H ₂ O	Cr: 10.42	0.275	0.275	0.275	0.275	0.275					
Lithium Chloride	Li: 16.38	0.0174	0.0174	0.0174	0.0174	0.0174					
Boric Acid	B: 17.50	0.0815	0.0815	0.0815	0.0815	0.0815					
Sodium Fluoride	F: 45.24	0.0635	0.0635	0.0635	0.0635	0.0635					
Nickel Carbonate	Ni: 45.00	0.0318	0.0318	0.0318	0.0318	0.0318					
Ammonium Vanadate	V: 43.55	0.0066	0.0066	0.0066	0.0066	0.0066					
Sucrose finely powdered		223.648	292.688	381.511	160.562	436.999					

*: Composition is differ from that of the AIN standard diet. The mineral mix and vitamin mix for each composition was designed to reflect the mineral (essential minerals) and vitamin contents of the dried beef at each additional level.

Table 3 Body weights of male rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	Number of animals	Day					Gain 0~28
		1	7	14	21	28	
0% ^{a)}	6	121	176	236	291	335	214
		± 4	± 6	± 8	± 7	± 11	± 14
5%	6	122	181	239	296	340	218
		± 6	± 8	± 14	± 24	± 31	± 28
10%	6	121	172	226	280	308	188
		± 6	± 7	± 12	± 20	± 23	± 20
0% ^{b)}	6	122	180	240	286	319	198
		± 5	± 8	± 12	± 23	± 40	± 37
20%	6	121	164 *	208 *	246 *	271 *	150 *
		± 5	± 12	± 22	± 30	± 34	± 30

Each value is expressed as mean±S.D.

a):Control I b):Control II

*:Significantly different from control II at 5% level of probability

Table 4 Body weights of female rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	Number of animals	Day					Gain 0~28
		1	7	14	21	28	
0% ^{a)}	6	116	146	174	198	218	102
		± 5	± 6	± 10	± 12	± 16	± 16
5%	6	115	149	171	201	223	107
		± 7	± 15	± 15	± 15	± 17	± 11
10%	6	115	145	175	195	210	94
		± 5	± 7	± 9	± 14	± 18	± 15
0% ^{b)}	6	116	148	180	207	231	115
		± 7	± 11	± 12	± 13	± 18	± 13
20%	6	116	145	174	197	217	101
		± 6	± 11	± 17	± 17	± 20	± 18

Each value is expressed as mean±S.D.

a):Control I b):Control II

Table 5 Food consumption of male rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	Number of animals	(g/day/rat)			
		Week 1	2	3	4
0% ^{a)}	6	18	21	22	22
		± 1	± 1	± 2	± 1
5%	6	18	19	21	21
		± 2	± 2	± 3	± 3
10%	6	17	19	20	18
		± 1	± 1	± 2	± 1
0% ^{b)}	6	17	19	20	17
		± 1	± 2	± 2	± 0
20%	6	16	18	17	15
		± 1	± 3	± 3	± 5

Each value is expressed as mean±S.D.
a):Control I b):Control II

Table 6 Food consumption of female rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	Number of animals	(g/day/rat)			
		Week 1	2	3	4
0% ^{a)}	6	13	14	15	15
		± 1	± 2	± 1	± 2
5%	6	13	13	15	15
		± 2	± 3	± 1	± 1
10%	6	14	14	14	13
		± 1	± 2	± 3	± 1
0% ^{b)}	6	14	14	15	15
		± 1	± 1	± 0	± 2
20%	6	13	14	13	14
		± 1	± 1	± 1	± 1

Each value is expressed as mean±S.D.
a):Control I b):Control II

Table

Conc. diet

0'

5'

10%

0'

20%

Conc. diet

0'

5'

10%

0'

20%

Color:
Cloud:
Protei:
Glucos:
Keton:
Occult:
Urobil:
Bilirub:
a): Co

Table

Conc diet

0

5

10

0

20

Conc diet

0

5

10'

0

20'

Color:
Cloud:
Protei:
Glucos:
Keton:
Occul:
Urobi:
Bilirub:
a): Co

Table 7 Urinary findings of male rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	No. of animals	Color		Cloudy		pH						Protein					Glucose					
		PY		-	+	5.0	6.0	6.5	7.0	7.5	8.0	8.5	-	±	+	++	+++	-	±	+	++	+++
0% ^{a)}	6	6		6		5	1					2	4				6					
5%	6	6		6		6						3	3				6					
10%	6	6		6		4	1	1				1	4	1			6					
0% ^{b)}	6	6		6		6						1	4	1			6					
20%	6	6		6		5	1					3	2	1			6					

Conc. in diet	No. of animals	Ketone body					Occult blood					Urobilinogen				Bilirubin					
		-	±	+	++	+++	-	±	+	++	+++	0.1	1	2	4	8	-	+	++	+++	
0% ^{a)}	6	4	2				6					6					6				
5%	6	5	1				6					6					6				
10%	6	5	1				4	2				6					6				
0% ^{b)}	6	6					6					6					6				
20%	6	6					5	1				6					6				

Color: PY(pale yellow)
 Cloudy: - (negligible), + (cloudy)
 Protein: - (negligible), ±(15~30mg/dL), + (30mg/dL), ++(100mg/dL), +++(300mg/dL)
 Glucose: - (negligible), ±(0.1g/dL), +(0.25g/dL), ++(0.5g/dL), +++(1g/dL)
 Ketone body: - (negligible), ±(5mg/dL), +(15mg/dL), ++(40mg/dL), +++(80mg/dL)
 Occult blood: - (negligible), ±(trace), +(slight), ++(moderate), +++(marked)
 Urobilinogen: Ehrlich unit/dL
 Bilirubin: - (negligible), +(slight), ++(moderate), +++(marked)
 a): Control I, b): Control II

Table 8 Urinary findings of female rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	No. of animals	Color		Cloudy		pH						Protein					Glucose					
		PY		-	+	5.0	6.0	6.5	7.0	7.5	8.0	8.5	-	±	+	++	+++	-	±	+	++	+++
0% ^{a)}	6	6		6		5	1					2	4				6					
5%	6	6		6		3	2	1				5	1				6					
10%	6	6		6		3	2	1				3	2	1			6					
0% ^{b)}	6	6		6		4	1	1				2	4				6					
20%	6	6		6		4	1	1				4	2				6					

Conc. in diet	No. of animals	Ketone body					Occult blood					Urobilinogen				Bilirubin					
		-	±	+	++	+++	-	±	+	++	+++	0.1	1	2	4	8	-	+	++	+++	
0% ^{a)}	6	6					6					6					6				
5%	6	6					3	2	1			6					6				
10%	6	6					6					6					6				
0% ^{b)}	6	6					5	1				6					6				
20%	6	6					6					6					6				

Color: PY(pale yellow)
 Cloudy: - (negligible), + (cloudy)
 Protein: - (negligible), ±(15~30mg/dL), + (30mg/dL), ++(100mg/dL), +++(300mg/dL)
 Glucose: - (negligible), ±(0.1g/dL), +(0.25g/dL), ++(0.5g/dL), +++(1g/dL)
 Ketone body: - (negligible), ±(5mg/dL), +(15mg/dL), ++(40mg/dL), +++(80mg/dL)
 Occult blood: - (negligible), ±(trace), +(slight), ++(moderate), +++(marked)
 Urobilinogen: Ehrlich unit/dL
 Bilirubin: - (negligible), +(slight), ++(moderate), +++(marked)
 a): Control I, b): Control II

Table 9

Hematological findings of male rats fed the dry beef-contained diet in the 28-day repeat dose toxicity test

Conc. in diet	No. of animals	RBC (10 ⁶ /μL)	Hb (g/dL)	Ht (%)	MCV (fL)	MCH (pg)	MCHC (%)	PT (sec)	APTT (sec)	WBC (10 ³ /μL)
0% ^{a)}	6	737 ± 24	14.0 ± 0.7	40.7 ± 1.9	55 ± 4	19.0 ± 1.2	34.4 ± 0.7	12.8 ± 0.3	19.4 ± 1.7	60 ± 7
5%	6	773 ± 20	14.6 ± 0.6	42.5 ± 1.1	55 ± 2	18.8 ± 0.7	34.2 ± 0.6	13.3 ± 0.4	18.8 ± 1.1	62 ± 22
10%	6	731 ± 65	14.2 ± 1.3	40.9 ± 3.4	56 ± 2	19.4 ± 0.8	34.6 ± 1.2	13.6 ± 0.8	19.1 ± 1.4	46 ± 5
0% ^{b)}	6	756 ± 48	14.6 ± 0.9	42.8 ± 2.5	57 ± 4	19.3 ± 1.6	34.0 ± 0.4	13.4 ± 1.0	19.7 ± 2.9	51 ± 17
20%	6	822 ± 64	14.3 ± 2.1	41.4 ± 4.5	51 # ± 4	17.3 ± 2.0	34.4 ± 1.7	13.6 ± 0.3	18.2 ± 1.1	38 ± 10
		Differential leukocyte counts (%)								Plat. (10 ³ /μL)
Conc. in diet	No. of animals	Baso.	Eosin.	Neutro.					Other	Plat. (10 ³ /μL)
				Stab.	Seg.	Lymph.	Mono.			
0% ^{a)}	6	0 ± 0	1 ± 1	0 ± 0	14 ± 5	83 ± 6	2 ± 1	0 ± 0	132 ± 9	
5%	6	0 ± 0	1 ± 1	0 ± 0	14 ± 4	84 ± 4	1 ± 1	0 ± 0	119 ± 16	
10%	6	0 ± 0	1 ± 2	0 ± 0	12 ± 4	86 ± 5	2 ± 1	0 ± 0	143 ± 47	
0% ^{b)}	6	0 ± 0	0 ± 1	0 ± 0	15 ± 4	83 ± 5	1 ± 1	0 ± 0	124 ± 18	
20%	6	0 ± 0	1 ± 1	0 ± 0	12 ± 3	86 ± 4	1 ± 1	0 ± 0	134 ± 50	

Each value is expressed as mean ± S.D.

a) : Control I, b) : Control II

: Significantly different from control II at 5% level of probability

Table 10

Hematological findings of female rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	No. of animals	RBC (10 ⁶ /μL)	Hb (g/dL)	Ht (%)	MCV (fL)	MCH (pg)	MCHC (%)	WBC (10 ³ /μL)	Plat. (10 ³ /μL)	PT (sec)	APTT (sec)
0% ^{a)}	6	762 ± 28	15.0 ± 0.3	42.0 ± 1.2	55 ± 2	19.7 ± 0.6	35.8 ± 0.6	37 ± 6	104 ± 7	12.8 ± 0.3	18.1 ± 1.4
5%	6	767 ± 21	14.8 ± 0.4	42.2 ± 0.6	55 ± 2	19.4 ± 0.7	35.2 ± 0.7	46 ± 30	120 * ± 13	12.9 ± 0.3	17.7 ± 1.4
10%	6	787 ± 38	15.3 ± 0.4	43.1 ± 0.8	55 ± 2	19.5 ± 0.8	35.5 ± 0.7	29 ± 6	126 ** ± 10	13.0 ± 0.4	18.0 ± 0.5
0% ^{b)}	6	748 ± 23	15.3 ± 0.4	43.0 ± 0.9	58 ± 1	20.4 ± 0.5	35.6 ± 0.7	34 ± 9	114 ± 15	13.2 ± 0.6	18.1 ± 1.6
20%	6	765 ± 28	14.2 ± 1.2	40.8# ± 2.2	53## ± 2	18.6## ± 1.2	34.9 ± 1.6	33 ± 10	124 ± 17	12.9 ± 0.3	18.3 ± 0.5

Each value is expressed as mean ± S.D.

a) : Control I, b) : Control II

* : Significantly different from control I at 5% level of probability

** : Significantly different from control I at 1% level of probability

: Significantly different from control II at 5% level of probability

: Significantly different from control II at 1% level of probability

Table 11

Conc. in diet

0%^{a)}

5%

10%

0%^{b)}

20%

Conc. in diet

0%^{a)}

5%

10%

0%^{b)}

20%

Each val.

a) : Contr

* : Signi

** : Signi

: Signi

: Signi

Table

Conc diet

0%

5%

10%

0%

20%

Conc diet

0

5

10%

0

20%

Each

a) : C

* : S

** : S

: S

: S

Table 11

Blood biochemical findings of male rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	No. of animals	LDH (U/L)	GOT (U/L)	GPT (U/L)	ALP (U/L)	γ -GTP (U/L)	ChE (U/L)	CK (U/L)	T.P. (g/dL)	Alb (%)	α_1 -G (%)	α_2 -G (%)	α_3 -G (%)	β -G (%)	γ -G (%)	A/G
0% ^{a)}	6	323 ± 119	82 ± 8	30 ± 3	666 ± 164	0.56 ± 0.26	50 ± 10	223 ± 34	5.84 ± 0.22	50.7 ± 2.1	22.8 ± 1.6	5.5 ± 0.9	2.9 ± 0.5	14.3 ± 1.0	3.9 ± 0.9	1.03 ± 0.09
5%	6	238 ± 84	75 ± 8	28 ± 2	736 ± 136	0.47 ± 0.12	49 ± 5	220 ± 87	5.88 ± 0.32	52.6 ± 1.6	21.9 ± 0.9	6.0 ± 1.0	2.8 ± 0.5	13.5 ± 1.0	3.2 ± 0.7	1.11 ± 0.07
10%	6	344 ± 98	74 ± 12	32 ± 3	799 ± 164	0.20** ± 0.11	41 ± 7	288 ± 42	5.83 ± 0.19	52.2 ± 6.0	23.2 ± 2.9	5.2 ± 1.3	2.7 ± 0.7	13.4 ± 3.8	3.3 ± 1.4	1.12 ± 0.25
0% ^{b)}	6	403 ± 57	82 ± 6	28 ± 3	710 ± 306	0.41 ± 0.25	51 ± 10	249 ± 43	5.89 ± 0.12	50.3 ± 1.7	24.0 ± 5.5	4.9 ± 1.2	3.4 ± 0.8	12.9 ± 3.0	4.5 ± 0.5	1.01 ± 0.07
20%	6	339 ± 170	96 ± 18	32 ± 5	1086 ± 187	0.55 ± 0.42	41 ± 4	228 ± 69	5.66# ± 0.14	51.5 ± 2.6	21.0 ± 2.1	5.6 ± 1.2	3.7 ± 1.1	13.9 ± 1.4	4.3 ± 1.0	1.07 ± 0.11
Conc. in diet	No. of animals	T-Chol. (mg/dL)	T.G. (mg/dL)	PL (mg/dL)	Glu. (mg/dL)	BUN (mg/dL)	UA (mg/dL)	Crea. (mg/dL)	T-Bil. (mg/dL)	Ca (mg/dL)	P (mg/dL)	Na (mEq/L)	K (mEq/L)	Cl (mEq/L)		
0% ^{a)}	6	52 ± 12	55 ± 28	84 ± 18	115 ± 11	12.8 ± 2.0	0.96 ± 0.21	0.49 ± 0.05	0.28 ± 0.02	9.9 ± 0.3	7.6 ± 0.5	144 ± 1	4.22 ± 0.19	105 ± 1		
5%	6	47 ± 14	60 ± 18	85 ± 15	136* ± 15	10.6 ± 2.5	1.07 ± 0.31	0.53 ± 0.07	0.25 ± 0.04	9.9 ± 0.5	8.3 ± 0.7	144 ± 1	4.53 ± 0.39	105 ± 1		
10%	6	42 ± 4	41 ± 17	75 ± 8	126 ± 10	11.3 ± 2.3	1.26 ± 0.15	0.53 ± 0.09	0.27 ± 0.03	9.6 ± 0.1	8.1 ± 0.4	145 ± 1	4.65 ± 0.67	107 ± 1		
0% ^{b)}	6	43 ± 8	34 ± 14	71 ± 12	120 ± 6	11.1 ± 2.1	0.94 ± 0.10	0.47 ± 0.03	0.28 ± 0.03	9.7 ± 0.4	8.2 ± 0.6	146 ± 1	4.59 ± 0.36	106 ± 2		
20%	6	39 ± 6	31 ± 10	75 ± 7	129 ± 15	12.8 ± 1.6	1.37 ± 0.30	0.52 ± 0.07	0.28 ± 0.03	9.4 ± 0.3	7.3 ± 0.6	144 ± 1	4.25 ± 0.65	104 ± 3		

Each value is expressed as mean±S.D.

a): Control I, b): Control II

*: Significantly different from control I at 5% level of probability

**: Significantly different from control I at 1% level of probability

: Significantly different from control II at 5% level of probability

Table 12

Blood biochemical findings of female rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	No. of animals	LDH (U/L)	GOT (U/L)	GPT (U/L)	ALP (U/L)	γ -GTP (U/L)	ChE (U/L)	CK (U/L)	T.P. (g/dL)	Alb (%)	α_1 -G (%)	α_2 -G (%)	α_3 -G (%)	β -G (%)	γ -G (%)	A/G
0% ^{a)}	6	378 ± 106	84 ± 5	26 ± 5	415 ± 55	0.49 ± 0.19	257 ± 85	221 ± 73	6.33 ± 0.32	57.4 ± 1.3	18.5 ± 1.9	5.9 ± 0.8	2.7 ± 0.3	12.6 ± 0.7	3.1 ± 0.4	1.35 ± 0.07
5%	6	474 ± 115	78 ± 11	25 ± 2	416 ± 70	0.44 ± 0.25	319 ± 104	205 ± 46	6.34 ± 0.44	56.5 ± 2.2	19.1 ± 1.7	5.9 ± 0.4	2.5 ± 0.6	12.3 ± 0.9	3.7 ± 0.3	1.30 ± 0.12
10%	6	551 ± 134	87 ± 11	24 ± 2	498 ± 153	0.54 ± 0.32	349 ± 79	219 ± 34	6.19 ± 0.25	58.0 ± 2.5	17.8 ± 2.3	4.1* ± 1.3	2.8 ± 1.0	13.1 ± 0.9	4.2** ± 0.6	1.39 ± 0.14
0% ^{b)}	6	609 ± 200	81 ± 6	26 ± 2	413 ± 42	0.44 ± 0.21	253 ± 72	196 ± 32	6.40 ± 0.39	59.7 ± 2.0	18.6 ± 0.6	4.0 ± 0.7	2.1 ± 0.9	12.3 ± 1.0	3.3 ± 1.3	1.49 ± 0.12
20%	6	495 ± 229	78 ± 4	26 ± 2	462 ± 55	0.23# ± 0.08	371# ± 67	228 ± 88	6.37 ± 0.42	56.8 ± 2.9	18.8 ± 1.3	5.8## ± 1.0	2.3 ± 0.7	12.8 ± 0.8	8.6 ± 1.6	1.32 ± 0.15
Conc. in diet	No. of animals	T-Chol. (mg/dL)	T.G. (mg/dL)	PL (mg/dL)	Glu. (mg/dL)	BUN (mg/dL)	UA (mg/dL)	Crea. (mg/dL)	T-Bil. (mg/dL)	Ca (mg/dL)	P (mg/dL)	Na (mEq/L)	K (mEq/L)	Cl (mEq/L)		
0% ^{a)}	6	54 ± 12	20 ± 6	99 ± 15	117 ± 12	16.5 ± 2.7	0.89 ± 0.22	0.64 ± 0.09	0.25 ± 0.03	9.7 ± 0.4	6.7 ± 0.9	143 ± 1	4.11 ± 0.32	106 ± 2		
5%	6	51 ± 9	20 ± 12	93 ± 16	122 ± 11	14.7 ± 3.6	1.20 ± 0.29	0.63 ± 0.10	0.27 ± 0.03	9.8 ± 0.2	6.8 ± 1.0	144 ± 1	4.14 ± 0.37	107 ± 1		
10%	6	52 ± 12	13 ± 5	95 ± 16	107 ± 7	15.1 ± 3.0	1.13 ± 0.11	0.68 ± 0.07	0.27 ± 0.03	9.6 ± 0.2	6.6 ± 0.6	145 ± 1	4.28 ± 0.22	110* ± 2		
0% ^{b)}	6	51 ± 8	14 ± 4	91 ± 10	122 ± 15	14.2 ± 2.2	0.92 ± 0.16	0.60 ± 0.05	0.26 ± 0.03	9.8 ± 0.3	6.4 ± 0.7	145 ± 1	4.27 ± 0.27	108 ± 2		
20%	6	45 ± 8	20 ± 13	90 ± 16	127 ± 6	17.8 ± 5.2	1.18 ± 0.29	0.68# ± 0.06	0.27 ± 0.02	9.8 ± 0.3	6.7 ± 0.6	145 ± 2	4.46 ± 0.87	108 ± 2		

Each value is expressed as mean±S.D.

a): Control I, b): Control II

*: Significantly different from control I at 5% level of probability

**: Significantly different from control I at 1% level of probability

: Significantly different from control II at 5% level of probability

: Significantly different from control II at 1% level of probability

Table 13-1 Absolute and relative organ weights of male rats fed dry beef-contained diet in a 28-day dose range-finding study

	Conc. in diet(%)	No. of Animals	B.W. (g)	Brain (g)	Liver (g)	Kidney (g)	Spleen (g)	Heart (g)	Lung (g)	Thymus (g)	Thyr. (mg)	Pitui. (mg)
Absolute	0 ^a	6	314 ±9	1.85 ±0.08	8.94 ±0.43	2.33 ±0.15	0.60 ±0.06	1.11 ±0.03	1.32 ±0.07	0.57 ±0.07	25.5 ±4.2	10.4 ±1.2
	5	6	318 ±30	1.92 ±0.08	8.97 ±1.21	2.56 ±0.11	0.56 ±0.10	1.10 ±0.12	1.28 ±0.04	0.67 ±0.15	19.3 ** ±2.4	10.0 ±1.4
	10	6	288 ±22	1.94 ±0.09	7.57 ±0.60	2.27 ±0.22	0.56 ±0.08	1.00 ±0.10	1.21 ±0.10	0.52 ±0.10	20.5 * ±1.9	10.1 ±1.0
	0 ^b	6	299 ±36	1.94 ±0.08	8.51 ±1.55	2.39 ±0.24	0.56 ±0.13	1.06 ±0.14	1.21 ±0.10	0.49 ±0.09	23.2 ±3.0	10.9 ±1.4
	20	6	257 ±29	1.90 ±0.08	6.32 # ±0.76	2.07 # ±0.12	0.50 ±0.08	0.96 ±0.14	1.13 ±0.06	0.47 ±0.06	21.4 ±2.3	9.6 ±1.0
	Relative ^c	0 ^a	6	314 ±9	0.59 ±0.03	2.84 ±0.12	0.74 ±0.03	0.19 ±0.01	0.35 ±0.01	0.42 ±0.02	0.18 ±0.02	8.2 ±1.5
5		6	318 ±30	0.61 ±0.05	2.82 ±0.20	0.81 ±0.05	0.18 ±0.01	0.35 ±0.02	0.40 ±0.03	0.21 ±0.04	6.1 * ±0.8	3.2 ±0.2
10		6	288 ±22	0.68 ±0.06	2.63 ±0.08	0.79 ±0.06	0.19 ±0.03	0.35 ±0.02	0.42 ±0.03	0.18 ±0.03	7.2 ±0.8	3.5 ±0.3
0 ^b		6	299 ±36	0.65 ±0.06	2.83 ±0.20	0.80 ±0.04	0.19 ±0.02	0.35 ±0.01	0.41 ±0.03	0.16 ±0.02	8.0 ±1.8	3.7 ±0.2
20		6	257 ±29	0.75 # ±0.08	2.47 # ±0.29	0.81 ±0.05	0.19 ±0.02	0.37 ±0.04	0.44 ±0.04	0.18 ±0.02	8.4 ±1.0	3.7 ±0.2

Each value is expressed as mean ± S.D.
a) : Control I ; b) : Control II
c) : Relative organ weight per 100g body weight
* : Significantly different from control I at 5% level of probability
** : Significantly different from control I at 1% level of probability
: Significantly different from control II at 5% level of probability

Table 13-2 Absolute and relative organ weights of male rats fed dry beef-contained diet in a 28-day dose range-finding study

	Conc. in diet(%)	No. of Animals	B.W. (g)	Adrenal (mg)	Testis (g)	Prost. (g)	Semi.v (g)	Epidid. (g)
Absolute	0 ^a	6	314 ±9	53.5 ±5.1	2.65 ±0.15	0.46 ±0.07	1.46 ±0.17	0.72 ±0.05
	5	6	318 ±30	51.0 ±7.2	2.64 ±0.15	0.43 ±0.07	1.33 ±0.17	0.68 ±0.02
	10	6	288 ±22	52.3 ±5.2	2.74 ±0.19	0.42 ±0.05	1.25 ±0.16	0.69 ±0.07
	0 ^b	6	299 ±36	51.2 ±6.5	2.65 ±0.17	0.49 ±0.11	1.40 ±0.25	0.71 ±0.06
	20	6	257 ±29	43.7 # ±4.4	2.58 ±0.17	0.30 ## ±0.04	1.09 # ±0.11	0.63 # ±0.03
	Relative ^c	0 ^a	6	314 ±9	17.0 ±1.3	0.84 ±0.04	0.15 ±0.02	0.46 ±0.04
5		6	318 ±30	16.1 ±2.0	0.84 ±0.09	0.14 ±0.02	0.42 ±0.05	0.22 ±0.02
10		6	288 ±22	18.3 ±2.5	0.95 ±0.08	0.15 ±0.02	0.44 ±0.06	0.24 ±0.01
0 ^b		6	299 ±36	17.2 ±1.5	0.89 ±0.09	0.16 ±0.03	0.47 ±0.06	0.24 ±0.03
20		6	257 ±29	17.1 ±2.0	1.02 ±0.11	0.12 ## ±0.02	0.42 ±0.03	0.25 ±0.03

Each value is expressed as mean ± S.D.
a) : Control I ; b) : Control II
c) : Relative organ weight per 100g body weight
: Significantly different from control II at 5% level of probability
: Significantly different from control II at 1% level of probability

Table 14

Co
d

Absoluta

Relative^c

Each value is
a) : Control
b) : Control
c) : Relative o
: Significantly

Absolute and relative organ weights of female rats fed dry beef-contaminated diet in a 28-day dose range-finding study

	Conc. in diet (%)	No. of Animals	B.W. (g)	Brain (g)	Liver (g)	Kidney (g)	Spleen (g)	Heart (g)	Lung (g)	Thymus (g)	Thyr. (mg)	Pitui. (mg)	Adrenal (mg)	Ovary (mg)	Uterus (g)
absolute	0 ^a	6	204 ±14	1.78 ±0.08	6.09 ±0.75	1.75 ±0.14	0.43 ±0.08	0.74 ±0.07	0.97 ±0.05	0.49 ±0.11	20.5 ±2.2	13.1 ±2.2	55.8 ±6.3	67.4 ±5.8	0.47 ±0.11
	5	6	208 ±16	1.80 ±0.07	6.16 ±0.88	1.67 ±0.18	0.45 ±0.08	0.77 ±0.10	1.03 ±0.06	0.46 ±0.06	20.5 ±3.5	13.3 ±2.0	52.5 ±9.4	73.8 ±14.9	0.48 ±0.14
	10	6	198 ±15	1.83 ±0.07	5.40 ±0.56	1.65 ±0.19	0.43 ±0.06	0.71 ±0.03	0.98 ±0.05	0.45 ±0.05	20.6 ±1.6	13.2 ±1.3	56.9 ±7.5	76.6 ±13.2	0.46 ±0.11
	0 ^b	6	215 ±17	1.83 ±0.06	6.31 ±0.81	1.74 ±0.20	0.42 ±0.05	0.76 ±0.08	1.00 ±0.09	0.52 ±0.08	21.4 ±2.2	13.5 ±2.7	62.5 ±8.4	73.9 ±7.4	0.54 ±0.19
	20	6	203 ±19	1.81 ±0.03	5.39 ±0.74	1.63 ±0.11	0.43 ±0.14	0.75 ±0.09	0.99 ±0.10	0.49 ±0.10	20.1 ±2.0	12.9 ±1.8	53.8 ±8.6	69.0 ±10.4	0.45 ±0.12
	relative ^c	0 ^a	6	204 ±14	0.88 ±0.07	2.98 ±0.19	0.86 ±0.07	0.21 ±0.03	0.36 ±0.03	0.48 ±0.04	0.24 ±0.05	10.1 ±1.4	6.4 ±0.9	27.4 ±2.2	33.1 ±2.7
5		6	208 ±16	0.87 ±0.04	2.96 ±0.26	0.80 ±0.04	0.22 ±0.03	0.37 ±0.02	0.50 ±0.04	0.22 ±0.04	10.0 ±1.9	6.4 ±0.6	25.2 ±3.3	35.5 ±6.5	0.23 ±0.07
10		6	198 ±15	0.93 ±0.09	2.72 ±0.13	0.83 ±0.05	0.22 ±0.02	0.36 ±0.02	0.50 ±0.02	0.23 ±0.02	10.5 ±1.1	6.7 ±0.5	28.8 ±2.9	38.8 ±6.7	0.23 ±0.05
0 ^b		6	215 ±17	0.86 ±0.04	2.93 ±0.17	0.81 ±0.05	0.20 ±0.03	0.36 ±0.03	0.47 ±0.03	0.25 ±0.04	10.1 ±1.8	6.3 ±0.8	29.1 ±3.2	34.4 ±1.9	0.25 ±0.08
20		6	203 ±19	0.90 ±0.08	2.65 # ±0.18	0.81 ±0.06	0.21 ±0.06	0.37 ±0.02	0.49 ±0.03	0.24 ±0.05	10.0 ±0.8	6.4 ±0.5	26.6 ±4.0	34.0 ±3.1	0.22 ±0.06

Each value is expressed as mean ± S.D.

a): Control I; b): Control II

c): Relative organ weight per 100g body weight

Significantly different from control II at 5% level of probability

Appendix 2 Individual body weights of female rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	Animal number	(g)				
		Day 0	7	14	21	28
0% ^{a)}	501	111	143	167	187	203
	502	112	143	172	197	223
	503	121	144	169	197	210
	504	114	155	185	215	242
	505	122	151	187	210	229
	506	116	141	161	183	201
	Mean	116	146	174	198	218
5%	507	104	132	154	181	196
	508	120	151	166	216	235
	509	113	145	168	196	220
	510	113	140	162	188	212
	511	124	176	196	219	245
	512	118	148	178	206	227
	Mean	115	149	171	201	223
10%	513	112	142	165	180	186
	514	109	139	162	179	188
	515	122	150	180	191	214
	516	114	137	177	206	217
	517	116	149	180	200	221
	518	119	154	185	214	231
	Mean	115	145	175	195	210
0% ^{b)}	519	105	134	164	193	210
	520	120	159	194	218	248
	521	111	136	168	189	206
	522	124	160	187	217	240
	523	116	151	182	208	235
	524	118	150	187	219	246
	Mean	116	148	180	207	231
20%	525	113	145	179	208	229
	526	125	150	182	210	224
	527	118	140	166	191	202
	528	109	143	172	198	216
	529	111	131	146	165	187
	530	120	163	196	210	244
	Mean	116	145	174	197	217

a):Control I b):Control II

Appendix 1 Individual body weights of male rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	Animal number	(g)				
		Day 0	7	14	21	28
0% ^{a)}	001	117	172	233	285	334
	002	121	176	228	285	328
	003	125	184	249	298	341
	004	122	171	231	287	330
	005	115	170	234	300	352
	006	126	180	241	290	322
	Mean	121	176	236	291	335
5%	007	122	177	228	274	308
	008	113	173	223	275	317
	009	129	196	264	336	385
	010	125	184	242	301	349
	011	122	179	241	309	365
	012	118	179	235	281	314
	Mean	122	181	239	296	340
10%	013	117	166	215	257	285
	014	124	174	221	274	311
	015	111	163	213	259	296
	016	129	182	241	300	336
	017	121	171	232	299	286
	018	122	176	236	292	335
	Mean	121	172	226	280	308
0% ^{b)}	019	116	166	227	285	321
	020	121	182	238	293	330
	021	125	190	258	323	375
	022	116	185	247	254	252
	023	124	178	228	271	306
	024	127	180	239	291	330
	Mean	122	180	240	286	319
20%	025	123	176	225	256	258
	026	121	158	194	221	241
	027	125	166	211	259	295
	028	128	180	242	296	329
	029	116	152	191	225	252
	030	114	152	187	221	249
	Mean	121	164	208	246	271

a):Control I b):Control II

Appendix 4 Individual food consumption of female rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	Cage number	(g/day/rat)			
		week 1	2	3	4
0% ^{a)}	16	13	13	15	15
	17	13	14	16	16
	18	14	16	14	13
	Mean	13	14	15	15
5%	19	12	10	16	14
	20	12	15	15	16
	21	16	14	16	14
	Mean	13	13	15	15
10%	22	13	12	11	14
	23	14	15	14	12
	24	14	14	17	14
	Mean	14	14	14	13
0% ^{b)}	25	14	14	15	16
	26	14	14	15	13
	27	13	15	15	16
	Mean	14	14	15	15
20%	28	14	15	14	15
	29	12	14	13	13
	30	13	13	12	13
	Mean	13	14	13	14

a):Control I b):Control II

Appendix 3 Individual food consumption of male rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	Cage number	(g/day/rat)			
		week 1	2	3	4
0% ^{a)}	1	17	20	21	21
	2	18	21	22	22
	3	18	22	24	22
	Mean	18	21	22	22
5%	4	16	17	18	18
	5	19	20	24	23
	6	18	21	21	22
	Mean	18	19	21	21
10%	7	17	18	18	17
	8	17	19	20	19
	9	18	20	22	19
	Mean	17	19	20	18
0% ^{b)}	10	16	18	21	17
	11	18	21	20	17
	12	17	17	18	17
	Mean	17	19	20	17
20%	13	17	18	15	9
	14	17	21	21	17
	15	15	15	16	18
	Mean	16	18	17	15

a):Control I b):Control II

Appendix 5 - 1

Individual urinary findings of male rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	Animal number	Color	Cloudy	pH	Protein	Glucose	Ketone body	Occult blood	Urobilinogen	Bilirubin
0% ^{a)}	001	PY	-	6.5	±	-	±	-	0.1	-
	002	PY	-	6.0	±	-	-	-	0.1	-
	003	PY	-	6.0	-	-	-	-	0.1	-
	004	PY	-	6.0	-	-	-	-	0.1	-
	005	PY	-	6.0	±	-	±	-	0.1	-
	006	PY	-	6.0	±	-	-	-	0.1	-
5%	007	PY	-	6.0	±	-	-	-	0.1	-
	008	PY	-	6.0	±	-	-	-	0.1	-
	009	PY	-	6.0	±	-	-	-	0.1	-
	010	PY	-	6.0	-	-	-	-	0.1	-
	011	PY	-	6.0	-	-	±	-	0.1	-
	012	PY	-	6.0	-	-	-	-	0.1	-
10%	013	PY	-	6.5	±	-	-	-	0.1	-
	014	PY	-	6.0	±	-	±	-	0.1	-
	015	PY	-	6.0	±	-	-	-	0.1	-
	016	PY	-	6.0	±	-	-	-	0.1	-
	017	PY	-	7.0	+	-	-	+	0.1	-
	018	PY	-	6.0	-	-	-	+	0.1	-

Color : PY (pale yellow)
 Cloudy : - (negligible)
 Protein : - (negligible), ± (15~30mg/dL), + (30mg/dL)
 Glucose : - (negligible)
 Ketone body : - (negligible), ± (5mg/dL)
 Occult blood : - (negligible), ± (trace), + (slight)
 Urobilinogen : Ehrlich unit/dL
 Bilirubin : - (negligible)
 a) : Control I

Appendix

Conc. in diet
0%^{a)}

5%

10%

Color : PY
 Cloudy : -
 Protein : -
 Glucose : -
 Ketone body : -
 Occult blood : -
 Urobilinogen : -
 Bilirubin : -
 a) : Control I

Appendix 5 - 2

Individual urinary findings of male rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	Animal number	Color	Cloudy	pH	Protein	Glucose	Ketone body	Occult blood	Urobilinogen	Bilirubin
0% ^{b)}	019	PY	-	6.0	±	-	-	-	0.1	-
	020	PY	-	6.0	±	-	-	-	0.1	-
	021	PY	-	6.0	+	-	-	-	0.1	-
	022	PY	-	6.0	±	-	-	-	0.1	-
	023	PY	-	6.0	-	-	-	-	0.1	-
	024	PY	-	6.0	±	-	-	-	0.1	-
20%	025	PY	-	6.5	-	-	-	±	0.1	-
	026	PY	-	6.0	±	-	-	-	0.1	-
	027	PY	-	6.0	±	-	-	-	0.1	-
	028	PY	-	6.0	-	-	-	-	0.1	-
	029	PY	-	6.0	-	-	-	-	0.1	-
	030	PY	-	6.0	+	-	-	-	0.1	-

b) : Control II

Appendix

Conc. in diet
0%^{b)}

20%

b) : Control II

Appendix 6 - 1

Individual urinary findings of female rats fed the dry beef-contained diet
in a 28day dose range-finding test

Conc. in diet	Animal number	Color	Cloudy	pH	Protein	Glucose	Ketone body	Occult blood	Urobilinogen	Bilirubin
0% ^{a)}	501	PY	-	6.0	±	-	-	-	0.1	-
	502	PY	-	6.0	±	-	-	-	0.1	-
	503	PY	-	6.0	-	-	-	-	0.1	-
	504	PY	-	6.5	±	-	-	-	0.1	-
	505	PY	-	6.0	-	-	-	-	0.1	-
	506	PY	-	6.0	±	-	-	-	0.1	-
5%	507	PY	-	7.0	-	-	-	-	0.1	-
	508	PY	-	6.0	-	-	-	-	0.1	-
	509	PY	-	6.5	-	-	-	-	0.1	-
	510	PY	-	6.5	-	-	-	+	0.1	-
	511	PY	-	6.0	±	-	-	±	0.1	-
	512	PY	-	6.0	-	-	-	±	0.1	-
10%	513	PY	-	7.5	+	-	-	-	0.1	-
	514	PY	-	6.0	-	-	-	-	0.1	-
	515	PY	-	6.0	±	-	-	-	0.1	-
	516	PY	-	7.0	-	-	-	-	0.1	-
	517	PY	-	6.0	±	-	-	-	0.1	-
	518	PY	-	7.0	-	-	-	-	0.1	-

Color : PY(pale yellow)

Cloudy : - (negligible)

Protein : - (negligible), ±(15~30mg/dL), + (30mg/dL)

Glucose : - (negligible)

Ketone body : - (negligible)

Occult blood : - (negligible), ±(trace), + (slight)

Urobilinogen : Ehrlich unit/dL

Bilirubin : - (negligible)

a) : Control I

Appendix 6 - 2

Individual urinary findings of female rats fed the dry beef-contained diet
in a 28day dose range-finding test

Conc. in diet	Animal number	Color	Cloudy	pH	Protein	Glucose	Ketone body	Occult blood	Urobilinogen	Bilirubin
0% ^{b)}	519	PY	-	7.5	±	-	-	-	0.1	-
	520	PY	-	6.0	-	-	-	-	0.1	-
	521	PY	-	6.0	±	-	-	-	0.1	-
	522	PY	-	6.0	-	-	-	-	0.1	-
	523	PY	-	6.5	±	-	-	±	0.1	-
	524	PY	-	6.0	±	-	-	-	0.1	-
20%	525	PY	-	6.5	-	-	-	-	0.1	-
	526	PY	-	6.0	-	-	-	-	0.1	-
	527	PY	-	6.0	-	-	-	-	0.1	-
	528	PY	-	7.5	±	-	-	-	0.1	-
	529	PY	-	6.0	-	-	-	-	0.1	-
	530	PY	-	6.0	±	-	-	-	0.1	-

b) : Control II

Appendix 7 - 1

Individual hematological findings of male rats fed the dry beef-contained diet in a 28-day dose range-finding test

Conc. in diet	Animal number	RBC (10 ⁶ /μL)	Hb (g/dL)	Ht (%)	MCV (fL)	MCH (pg)	MCHC (%)	PT (sec)	APTT (sec)	WBC (10 ⁶ /μL)
0% ^{a)}	001	711	13.7	39.3	55	19.3	34.9	13.2	21.7	69
	002	777	13.2	38.6	50	17.0	34.2	12.9	17.9	55
	003	749	15.0	42.3	56	20.0	35.5	12.9	20.2	58
	004	739	13.3	39.2	53	18.0	33.9	12.3	18.6	67
	005	721	14.3	42.4	59	19.8	33.7	13.0	17.4	55
	006	722	14.4	42.5	59	19.9	33.9	12.6	20.7	53
	Mean		737	14.0	40.7	55	19.0	34.4	12.8	19.4
5%	007	794	14.8	42.2	53	18.8	35.1	12.9	18.5	85
	008	784	14.3	42.0	54	18.2	34.0	13.6	19.0	32
	009	739	13.8	40.8	55	18.7	33.8	13.8	18.7	57
	010	780	14.5	43.1	55	18.6	33.6	13.5	17.5	46
	011	761	15.4	44.0	58	20.2	35.0	13.2	20.8	88
	012	780	14.5	42.8	55	18.6	33.9	13.0	18.4	64
	Mean		773	14.6	42.5	55	18.8	34.2	13.3	18.8
10%	013	792	15.1	42.3	53	19.1	35.7	13.0	20.8	45
	014	699	14.6	40.1	57	20.9	36.4	14.4	20.2	46
	015	727	14.4	42.1	58	19.8	34.2	13.3	19.9	54
	016	776	14.7	42.8	55	18.9	34.3	12.8	18.3	39
	017	618	11.5	34.3	56	18.6	33.5	14.7	18.4	46
	018	771	14.7	43.6	57	19.1	33.7	13.2	17.1	42
	Mean		731	14.2	40.9	56	19.4	34.6	13.6	19.1
0% ^{b)}	019	794	13.1	39.1	49	18.5	33.5	12.8	16.8	49
	020	758	14.3	41.4	55	18.9	34.5	12.4	16.7	69
	021	696	14.6	42.5	61	21.0	34.4	13.5	18.6	67
	022	820	15.8	46.5	57	19.3	34.0	15.2	23.4	32
	023	707	14.7	43.2	61	20.8	34.0	13.3	22.7	31
	024	762	14.8	44.2	58	19.4	33.5	13.4	20.1	57
	Mean		756	14.6	42.8	57	19.3	34.0	13.4	19.7
20%	025	718	10.0	32.2	46	13.9	31.1	13.9	18.7	32
	026	820	15.4	44.1	54	18.8	34.9	14.1	16.3	25
	027	822	15.5	43.2	53	18.9	35.9	13.4	19.3	47
	028	916	14.6	42.4	46	15.9	34.4	13.4	18.1	51
	029	807	14.7	42.5	53	18.2	34.6	13.6	18.8	31
	030	847	15.4	43.8	52	18.2	35.2	13.4	17.8	43
	Mean		822	14.3	41.4	51	17.3	34.4	13.6	18.2

a): Control I, b): Control II

Appendix 7 - 2

Individual hematological findings of male rats fed the dry beef-contained diet in the 28-day repeat dose toxicity test

Conc. in diet	Animal number	Differential leukocyte counts (%)							Plat. (10 ⁶ /μL)
		Baso.	Eosin.	Neutro.		Lymph.	Mono.	Other	
				Stab.	Seg.				
0% ^{a)}	001	0	1	0	14	83	2	0	130
	002	1	0	0	10	87	2	0	138
	003	0	0	0	16	83	1	0	128
	004	0	2	0	22	74	2	0	146
	005	0	1	0	16	80	3	0	119
	006	0	0	0	8	92	0	0	130
	Mean		0	1	0	14	83	2	0
5%	007	0	0	0	20	78	2	0	137
	008	0	0	0	10	87	3	0	126
	009	0	1	0	10	88	1	0	96
	010	0	0	0	12	87	1	0	131
	011	0	1	0	19	80	0	0	105
	012	0	1	0	13	86	0	0	116
	Mean		0	1	0	14	84	1	0
10%	013	0	1	0	12	85	1	0	125
	014	0	0	0	8	92	0	0	122
	015	0	1	0	7	90	2	0	127
	016	0	0	0	12	87	1	0	131
	017	0	0	0	19	77	4	0	239
	018	0	4	0	12	83	1	0	113
	Mean		0	1	0	12	86	2	0
0% ^{b)}	019	0	0	0	12	88	0	0	128
	020	0	0	0	11	87	2	0	110
	021	0	1	0	22	76	1	0	97
	022	0	1	0	17	80	2	0	142
	023	0	0	1	18	81	0	0	142
	024	0	0	0	12	87	1	0	124
	Mean		0	0	0	15	83	1	0
20%	025	0	0	0	7	92	1	0	234
	026	0	0	0	11	89	0	0	126
	027	0	2	0	13	84	1	0	104
	028	0	1	0	15	83	1	0	102
	029	0	0	0	15	83	2	0	123
	030	0	1	0	12	87	0	0	114
	Mean		0	1	0	12	86	1	0

a): Control I, b): Control II

Appendix

Conc. in diet

0%^{a)}

5%

10%

0%^{b)}

20%

a): Control

Appendix 8

Individual hematological findings of female rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	Animal number	RBC (10 ⁶ /μL)	Hb (g/dL)	Ht (%)	MCV (fL)	MCH (pg)	MCHC (%)	WBC (10 ³ /μL)	Plat. (10 ⁹ /μL)	PT (sec)	APTT (sec)
0% ^{a)}	501	765	14.7	40.7	53	19.2	36.1	29	111	12.9	17.1
	502	717	14.7	40.7	57	20.5	36.1	31	103	13.1	20.5
	503	780	15.0	42.4	54	19.2	35.4	42	100	12.6	16.6
	504	761	14.9	42.6	57	19.8	35.0	42	107	12.3	18.3
	505	758	15.4	42.1	56	20.3	36.6	35	94	12.7	17.7
	506	800	15.5	43.7	55	19.4	35.5	43	111	13.2	18.4
	Mean	762	15.0	42.0	55	19.7	35.8	37	104	12.8	18.1
5%	507	785	15.1	42.0	54	19.4	36.0	38	137	12.9	17.4
	508	766	14.2	41.2	54	18.5	34.5	28	123	13.2	17.6
	509	762	14.6	42.4	56	19.2	34.4	19	121	12.6	18.1
	510	797	14.9	42.7	54	18.7	34.9	44	129	13.3	15.3
	511	761	15.4	43.0	57	20.2	35.8	104	100	12.8	18.1
	512	734	14.8	41.9	57	20.2	35.3	44	111	12.7	19.6
	Mean	767	14.8	42.2	55	19.4	35.2	46	120	12.9	17.7
10%	513	827	15.1	43.0	52	18.3	35.1	28	125	12.7	17.8
	514	821	15.6	44.0	54	19.0	35.5	23	123	12.4	17.3
	515	751	15.4	42.2	56	20.5	36.5	22	115	13.1	18.1
	516	772	15.0	43.3	56	19.4	34.8	33	131	13.4	18.8
	517	811	15.9	44.1	54	19.6	36.1	38	143	12.7	18.4
	518	739	14.9	42.2	57	20.2	35.3	28	118	13.4	18.0
	Mean	787	15.3	43.1	55	19.5	35.5	29	126	13.0	18.0
0% ^{b)}	519	766	15.4	44.0	58	20.4	36.0	25	105	13.4	19.0
	520	718	14.8	42.2	59	20.6	35.1	46	121	12.5	17.3
	521	776	15.8	43.2	56	20.4	36.6	38	141	14.2	16.8
	522	738	15.5	43.5	59	21.0	35.6	21	110	12.9	16.1
	523	769	15.1	43.3	56	19.6	34.9	39	101	13.1	20.5
	524	728	15.0	41.6	57	20.6	36.1	32	106	13.3	18.8
	Mean	748	15.3	43.0	58	20.4	35.6	34	114	13.2	18.1
20%	525	759	13.8	38.6	51	18.2	35.8	25	125	12.4	18.5
	526	804	15.3	43.3	54	19.0	35.3	48	132	13.1	18.9
	527	794	15.0	42.0	53	18.9	35.7	25	113	13.2	17.6
	528	748	14.3	40.3	54	19.1	35.5	23	123	12.7	18.7
	529	753	15.0	42.5	56	19.9	35.3	43	100	13.1	17.9
	530	730	12.0	38.0	52	16.4	31.6	34	151	13.0	18.2
	Mean	765	14.2	40.8	53	18.6	34.9	33	124	12.9	18.3

a): Control I, b): Control II

Appendix 9 - 1

Individual blood biochemical findings of male rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	Animal number	LDH (U/L)	GOT (U/L)	GPT (U/L)	ALP (U/L)	γ-GTP (U/L)	ChE (U/L)	CK (U/L)	T.P. (g/dL)	Alb (%)	α ₁ -G (%)	α ₂ -G (%)	α ₃ -G (%)	β-G (%)	γ-G (%)	A/G
0% ^{a)}	001	337	88	34	843	0.99	59	229	5.58	49.7	21.1	5.6	3.4	15.7	4.5	0.99
	002	344	87	33	830	0.93	45	252	6.06	49.4	22.6	6.6	3.5	13.8	4.1	0.98
	003	267	74	29	472	0.38	45	242	6.05	51.8	21.2	5.4	2.6	14.7	4.3	1.07
	004	537	82	29	807	0.68	66	239	5.61	48.9	25.3	4.1	3.1	14.5	4.1	0.96
	005	267	90	30	764	0.53	45	218	5.76	54.5	23.4	5.1	2.3	12.6	2.1	1.20
	006	188	69	26	480	0.23	42	157	5.98	49.6	23.1	6.4	2.6	14.3	4.0	0.98
	Mean	323	82	30	666	0.56	50	223	5.84	50.7	22.8	5.5	2.9	14.3	3.9	1.03
5%	007	377	79	30	737	0.58	50	254	5.92	49.8	21.2	7.2	3.1	14.6	4.1	0.99
	008	227	84	30	871	0.40	48	378	5.53	54.4	21.8	4.7	3.2	12.5	3.4	1.19
	009	129	77	27	530	0.32	54	154	5.54	52.5	21.4	5.7	2.3	15.0	3.1	1.11
	010	242	66	25	785	0.45	45	164	5.93	53.8	23.2	5.4	2.4	13.1	2.1	1.16
	011	183	65	28	865	0.43	42	181	6.39	53.0	21.0	7.0	3.5	12.8	2.7	1.13
	012	270	77	29	627	0.66	56	179	5.97	52.2	22.9	5.8	2.3	13.1	3.7	1.09
	Mean	238	75	28	736	0.47	49	220	5.88	52.6	21.9	6.0	2.8	13.5	3.2	1.11
10%	013	462	80	30	733	0.13	36	255	5.96	54.1	21.4	4.6	3.0	12.6	4.3	1.18
	014	344	89	31	757	0.22	48	320	5.84	54.7	23.1	4.8	2.8	11.9	2.7	1.21
	015	223	79	29	933	0.35	45	327	5.80	49.3	19.3	7.6	3.4	15.2	5.2	0.97
	016	443	66	32	825	0.20	48	239	5.79	41.7	27.7	5.0	2.5	20.0	3.1	0.72
	017	244	56	37	541	0.29	32	328	6.06	59.1	25.1	3.8	1.4	9.6	1.0	1.44
	018	349	74	34	1005	0.03	36	253	5.50	54.2	22.7	5.5	3.1	10.8	3.7	1.18
	Mean	344	74	32	799	0.20	41	288	5.83	52.2	23.2	5.2	2.7	13.4	3.3	1.12
0% ^{b)}	019	347	81	33	674	0.52	61	277	5.88	48.3	28.2	5.4	3.8	10.2	4.1	0.93
	020	393	79	25	650	0.31	48	246	6.08	49.8	26.5	3.5	3.1	12.5	4.6	0.99
	021	409	72	26	706	0.30	39	202	5.78	51.9	23.0	5.2	2.9	11.6	5.4	1.08
	022	337	90	31	1277	0.83	41	307	5.83	51.6	24.0	4.2	3.7	11.9	4.6	1.07
	023	449	84	26	351	0.09	52	196	5.80	48.2	28.6	4.4	2.5	12.2	4.1	0.93
	024	484	83	28	604	0.38	63	263	5.97	51.7	13.7	6.9	4.6	18.8	4.3	1.07
	Mean	403	82	28	710	0.41	51	249	5.89	50.3	24.0	4.9	3.4	12.9	4.5	1.01
20%	025	265	84	30	1151	0.15	35	299	5.60	50.2	18.0	6.3	6.2	14.6	5.7	1.01
	026	573	74	24	1124	0.54	43	210	5.60	48.6	19.7	6.8	4.6	15.0	5.3	0.95
	027	483	89	31	954	0.11	42	319	5.74	50.9	23.8	6.2	2.9	13.2	3.0	1.04
	028	150	114	37	1145	0.55	45	172	5.53	54.6	21.7	4.8	2.6	12.6	3.7	1.20
	029	177	94	31	799	0.67	39	145	5.60	54.9	20.3	5.8	2.9	12.4	3.7	1.22
	030	385	121	39	1341	1.28	40	222	5.91	49.8	22.6	3.6	3.7	15.7	4.6	0.99
	Mean	339	96	32	1086	0.55	41	228	5.66	51.5	21.0	5.6	3.7	13.9	4.3	1.07

a): Control I, b): Control II

Individual blood biochemical findings of male rats fed the dry beef contained diet in a 28day dose range finding test

Conc. in diet	Animal number	T-Chol. (mg/dL)	T.G. (mg/dL)	PL (mg/dL)	Glu. (mg/dL)	BUN (mg/dL)	UA (mg/dL)	Crea. (mg/dL)	T-Bil. (mg/dL)	Ca (mg/dL)	P (mg/dL)	Na (mEq/L)	K (mEq/L)	Cl (mEq/L)
0% ^{a)}	001	45	39	81	102	13.3	1.10	0.50	0.24	9.3	7.1	145	4.29	106
	002	70	104	111	124	11.6	1.10	0.57	0.29	10.3	7.2	144	4.23	105
	003	64	71	101	119	15.1	1.20	0.52	0.26	9.9	7.3	144	3.97	105
	004	46	38	70	114	9.9	0.94	0.45	0.29	9.9	8.3	144	4.50	105
	005	46	39	72	129	14.8	0.69	0.44	0.31	10.1	8.1	144	4.29	103
	006	41	36	71	102	12.1	0.74	0.45	0.28	10.0	7.6	145	4.08	103
	Mean		52	55	84	115	12.8	0.96	0.49	0.28	9.9	7.6	144	4.22
5%	007	47	40	82	124	12.9	1.36	0.54	0.28	9.4	8.0	145	5.08	106
	008	32	42	65	131	11.1	1.25	0.56	0.28	9.3	7.3	144	4.90	107
	009	31	65	73	121	10.4	1.43	0.55	0.23	10.0	8.0	145	4.06	105
	010	52	69	88	131	13.3	0.85	0.50	0.29	10.1	9.1	144	4.36	104
	011	53	88	97	151	9.8	0.89	0.49	0.21	10.5	8.9	144	4.27	103
	012	68	63	104	157	6.3	0.66	0.42	0.21	9.8	8.2	144	4.50	105
	Mean		47	60	85	136	10.6	1.07	0.53	0.25	9.9	8.3	144	4.53
10%	013	37	41	80	138	7.5	1.09	0.52	0.28	9.3	7.3	146	4.42	108
	014	48	69	86	118	13.8	1.26	0.69	0.29	9.8	7.9	146	3.99	108
	015	40	28	75	122	12.5	1.42	0.49	0.27	9.4	7.8	143	4.64	106
	016	46	51	75	136	10.9	1.13	0.43	0.24	9.8	8.8	144	4.85	105
	017	43	29	66	114	10.0	1.46	0.53	0.31	9.5	8.1	145	5.86	105
	018	39	26	67	125	13.3	1.18	0.50	0.25	9.5	8.4	146	4.16	107
	Mean		42	41	75	126	11.3	1.26	0.53	0.27	9.6	8.1	145	4.65
0% ^{b)}	019	39	40	66	129	9.9	1.10	0.50	0.25	9.6	7.7	146	5.21	107
	020	55	37	87	115	8.0	0.94	0.49	0.27	9.8	7.7	148	4.35	104
	021	51	54	85	115	11.6	0.78	0.44	0.29	10.2	8.5	147	4.54	105
	022	38	18	61	121	10.8	0.97	0.49	0.31	9.2	7.7	146	4.71	108
	023	38	19	61	119	14.3	0.89	0.47	0.23	9.8	8.5	145	4.56	105
	024	35	36	65	118	11.7	0.96	0.44	0.30	10.0	9.0	147	4.17	105
	Mean		43	34	71	120	11.1	0.94	0.47	0.28	9.7	8.2	146	4.59
20%	025	35	25	69	124	13.7	1.33	0.57	0.33	9.5	7.6	143	5.12	103
	026	45	49	77	157	13.1	1.40	0.47	0.28	9.5	7.3	144	4.53	106
	027	46	25	82	122	11.1	1.22	0.42	0.29	9.2	7.4	143	4.53	106
	028	34	36	74	132	14.4	1.38	0.61	0.23	9.8	8.2	144	3.28	99
	029	38	28	84	115	10.5	1.12	0.51	0.25	9.2	6.6	145	4.29	105
	030	34	24	66	126	14.1	1.18	0.51	0.29	9.0	6.6	145	3.77	106
	Mean		39	31	75	129	12.8	1.37	0.52	0.28	9.4	7.3	144	4.25

a) : Control I, b) : Control II

Individual blood biochemical findings of female rats fed the dry beef contained diet in a 28day dose range finding test

Conc. in diet	Animal number	LDH (U/L)	GOT (U/L)	GPT (U/L)	ALP (U/L)	γ-GTP (U/L)	ChE (U/L)	CK (U/L)	T.P. (g/dL)	Alb (%)	α ₁ -G (%)	α ₂ -G (%)	α ₃ -G (%)	β-G (%)	γ-G (%)	A/G
0% ^{a)}	501	465	86	26	470	0.28	216	264	6.23	58.2	19.5	4.7	2.7	11.7	3.2	1.39
	502	319	79	22	340	0.46	187	148	5.95	58.4	15.9	6.5	2.5	13.1	3.6	1.40
	503	537	86	34	361	0.38	413	236	6.65	59.1	16.5	6.0	2.9	12.1	3.4	1.44
	504	247	88	29	410	0.51	260	125	6.40	56.3	20.6	5.7	3.0	11.9	2.5	1.29
	505	334	76	22	438	0.48	275	318	6.73	56.3	18.2	6.8	2.4	13.1	3.2	1.29
	506	368	88	25	472	0.85	189	237	6.02	56.2	20.0	5.4	2.4	13.4	2.6	1.28
	Mean		378	84	26	415	0.49	257	221	6.33	57.4	18.5	5.9	2.7	12.6	3.1
5%	507	557	78	27	484	0.63	252	290	6.96	57.3	18.0	6.2	2.6	12.4	3.5	1.34
	508	598	98	25	508	0.57	202	195	6.80	57.4	20.0	6.0	1.4	12.0	3.2	1.35
	509	341	73	25	366	0.30	440	170	6.74	58.5	18.5	5.7	2.8	10.6	3.9	1.41
	510	364	77	26	325	0.73	225	216	6.06	58.1	16.4	6.4	2.4	12.8	3.9	1.39
	511	573	72	22	397	0.33	392	192	6.70	52.7	21.2	5.9	3.1	13.1	4.0	1.11
	512	411	68	22	413	0.07	404	165	6.77	55.0	20.2	5.3	2.8	13.0	3.7	1.22
	Mean		474	78	25	416	0.44	319	205	6.34	56.5	19.1	5.9	2.5	12.3	3.7
10%	513	505	82	22	563	0.28	420	173	5.88	59.2	13.2	5.2	3.5	14.9	4.0	1.45
	514	632	99	25	505	0.37	375	213	6.08	57.3	18.1	4.5	3.9	12.4	3.8	1.34
	515	645	73	27	355	0.46	193	239	6.08	59.0	19.4	2.9	2.2	12.9	3.6	1.44
	516	517	80	21	359	0.30	378	214	6.45	61.7	17.8	2.5	1.2	13.0	3.8	1.61
	517	321	99	26	764	0.69	371	199	6.10	54.4	18.8	5.7	3.1	13.3	4.7	1.19
	518	685	88	23	444	1.11	356	273	6.52	56.6	19.3	3.9	2.7	12.3	5.2	1.30
	Mean		551	87	24	498	0.54	349	219	6.19	58.0	17.8	4.1	2.8	13.1	4.2
0% ^{b)}	519	672	86	26	460	0.37	299	192	5.90	60.1	17.5	4.7	2.2	12.1	3.4	1.51
	520	731	78	22	414	0.29	297	190	7.04	67.1	18.5	4.1	3.8	12.4	4.1	1.33
	521	219	74	27	377	0.48	190	142	6.17	62.4	19.2	4.7	1.8	10.9	1.2	1.66
	522	588	90	25	355	0.42	352	207	6.37	61.3	18.6	4.1	1.8	11.7	2.5	1.58
	523	676	80	27	459	0.83	186	204	6.34	58.8	19.0	3.3	1.8	13.0	4.1	1.43
	524	766	75	25	414	0.24	194	240	6.56	58.5	18.7	2.9	1.5	13.7	4.7	1.41
	Mean		609	81	26	413	0.44	253	196	6.40	59.7	18.6	4.0	2.1	12.3	3.3
20%	525	715	75	23	378	0.17	402	236	6.84	67.4	18.9	4.2	3.0	13.2	5.3	1.35
	526	649	74	26	504	0.11	336	233	6.63	59.7	18.5	5.0	1.4	13.5	1.9	1.48
	527	174	81	28	466	0.34	382	110	5.70	58.2	19.9	6.0	2.1	11.3	2.5	1.39
	528	642	85	25	537	0.21	483	332	6.63	58.7	17.9	6.7	2.0	12.3	2.4	1.42
	529	244	76	28	434	0.24	294	145	6.12	51.8	20.2	5.9	3.1	13.3	5.7	1.07
	530	546	78	24	452	0.28	329	314	6.30	55.1	19.2	6.9	2.0	12.9	3.9	1.23
	Mean		495	78	26	462	0.23	371	228	6.37	56.8	18.8	5.8	2.3	12.8	3.6

a) : Control I, b) : Control II

Individual blood biochemical findings of female rats fed the dry beef-contained diet in a 28day dose range-finding test

Conc. in diet	Animal number	T.Chol. (mg/dL)	T.G. (mg/dL)	PL (mg/dL)	Gluc. (mg/dL)	BUN (mg/dL)	UA (mg/dL)	Crea. (mg/dL)	T.Bil. (mg/dL)	Ca (mg/dL)	P (mg/dL)	Na (mEq/L)	K (mEq/L)	Cl (mEq/L)
0% ^{a)}	501	50	16	95	116	19.5	1.12	0.74	0.21	9.6	6.9	144	3.81	103
	502	36	14	77	133	18.3	0.87	0.67	0.29	9.3	6.9	142	4.29	108
	503	69	20	103	114	17.4	1.17	0.74	0.26	9.7	6.1	141	4.09	105
	504	63	30	113	113	12.3	0.67	0.57	0.27	10.2	8.0	144	3.66	104
	505	70	24	118	99	14.3	0.79	0.56	0.23	10.1	7.5	144	4.49	106
	506	49	15	89	129	16.9	0.82	0.56	0.26	9.6	6.7	144	4.33	109
	Mean	54	20	99	117	16.6	0.89	0.64	0.25	9.7	6.7	143	4.11	106
5%	507	48	13	81	108	18.0	1.49	0.70	0.26	9.6	6.3	142	4.46	109
	508	38	11	68	121	20.3	1.14	0.79	0.22	9.6	6.6	145	3.58	107
	509	80	42	110	135	11.7	0.97	0.61	0.27	10.0	5.2	145	4.45	105
	510	62	28	110	115	12.4	0.90	0.56	0.27	10.1	8.2	144	3.90	106
	511	50	14	96	128	12.8	1.62	0.52	0.31	9.8	7.4	144	4.46	107
	512	48	13	94	129	13.0	1.10	0.58	0.27	9.9	7.1	144	4.02	108
	Mean	51	20	93	122	14.7	1.20	0.63	0.27	9.8	6.8	144	4.14	107
10%	513	64	19	110	115	17.8	1.34	0.74	0.27	9.4	6.0	143	4.12	107
	514	66	12	114	106	18.9	1.07	0.72	0.24	9.4	6.2	145	4.19	111
	515	40	10	80	113	14.8	1.05	0.58	0.28	9.6	6.5	145	4.40	108
	516	45	11	81	106	10.6	1.06	0.59	0.22	9.7	6.7	145	4.40	109
	517	68	20	106	96	13.3	1.15	0.61	0.30	9.8	7.6	146	3.98	109
	518	41	8	80	109	15.2	1.13	0.70	0.29	9.4	6.8	146	4.57	113
	Mean	52	13	95	107	15.1	1.13	0.66	0.27	9.6	6.6	145	4.28	110
0% ^{b)}	519	64	16	96	94	13.4	0.91	0.59	0.21	9.5	6.6	146	4.84	109
	520	63	14	107	119	11.4	0.81	0.64	0.26	10.2	6.1	145	4.22	106
	521	49	19	86	125	17.6	0.75	0.59	0.26	9.6	6.2	144	4.07	107
	522	40	10	77	134	13.1	1.03	0.52	0.25	9.6	6.0	146	4.57	111
	523	50	9	86	124	13.8	0.84	0.61	0.26	9.7	7.5	146	4.15	107
	524	47	14	94	134	16.6	1.18	0.66	0.29	9.9	6.8	145	3.98	108
	Mean	51	14	91	122	14.2	0.92	0.60	0.26	9.8	6.4	145	4.27	108
20%	525	62	45	116	125	10.1	0.89	0.66	0.26	10.2	6.6	143	4.46	106
	526	44	15	91	123	16.7	1.06	0.77	0.26	9.9	7.2	148	3.57	106
	527	41	10	76	136	25.6	1.16	0.70	0.27	9.3	6.6	146	3.81	110
	528	30	12	69	126	15.1	1.22	0.60	0.24	9.9	6.1	145	4.86	109
	529	62	15	94	134	19.1	1.03	0.65	0.29	9.4	6.2	146	4.32	111
	530	48	20	94	119	20.0	1.73	0.71	0.30	9.8	7.7	144	5.92	108
	Mean	45	20	90	127	17.8	1.18	0.68	0.27	9.8	6.7	145	4.46	108

a): Control I, b): Control II

Appendix 11 Absolute organ weights of individual male rats fed dry beef-contained diet in a 28-day dose range-finding study

Conc. in diet (%)	Animal number	B.W. (g)	Brain (g)	Liver (g)	Kidney (g)	Spleen (g)	Heart (g)	Lung (g)	Thymus (g)	Thyr. (mg)	Pitui. (mg)	Adrenal (mg)	Testis (g)	Prost. (g)	Semiv. (g)	Epidid. (g)
0 ^{a)}	001	312	1.90	8.66	2.43	0.61	1.16	1.41	0.66	26.7	10.1	48.8	2.62	0.40	1.51	0.73
	002	308	1.76	9.45	2.15	0.61	1.08	1.24	0.61	30.5	9.5	47.1	2.46	0.51	1.51	0.72
	003	315	1.89	9.00	2.28	0.59	1.11	1.22	0.57	28.4	8.9	58.1	2.56	0.53	1.37	0.72
	004	313	1.73	8.59	2.24	0.57	1.10	1.36	0.58	20.9	10.6	52.6	2.60	0.35	1.30	0.74
	005	331	1.89	9.43	2.58	0.69	1.09	1.35	0.53	19.9	10.7	60.1	2.85	0.49	1.76	0.63
	006	307	1.93	8.48	2.32	0.51	1.11	1.32	0.45	26.7	12.3	54.1	2.83	0.48	1.31	0.76
	Mean	314	1.85	8.94	2.33	0.60	1.11	1.32	0.57	25.5	10.4	53.5	2.65	0.46	1.46	0.72
5	007	287	1.94	7.88	2.49	0.48	1.02	1.22	0.48	18.7	8.8	51.3	2.68	0.40	1.26	0.67
	008	296	1.91	7.60	2.49	0.48	0.97	1.29	0.58	16.1	9.5	49.5	2.63	0.37	1.32	0.65
	009	362	2.03	9.72	2.59	0.73	1.29	1.34	0.61	18.1	12.3	53.1	2.45	0.47	1.40	0.71
	010	329	1.97	9.45	2.62	0.57	1.18	1.26	0.85	19.1	10.1	45.6	2.75	0.54	1.61	0.67
	011	341	1.85	10.76	2.73	0.62	1.05	1.31	0.86	22.9	10.8	63.6	2.83	0.37	1.30	0.67
	012	295	1.82	8.41	2.45	0.49	1.09	1.25	0.61	21.0	8.7	42.8	2.49	0.43	1.10	0.71
	Mean	318	1.92	8.97	2.56	0.56	1.10	1.28	0.67	19.3	10.0	51.0	2.64	0.43	1.33	0.68
10	013	269	1.84	7.05	2.05	0.44	0.89	1.09	0.52	23.8	9.1	54.0	2.60	0.43	1.06	0.62
	014	295	1.90	7.92	2.07	0.48	0.98	1.14	0.48	19.2	10.7	54.1	2.57	0.32	1.24	0.71
	015	272	1.92	7.32	2.26	0.58	0.91	1.18	0.51	18.7	8.5	44.5	2.57	0.46	1.09	0.61
	016	310	1.87	8.39	2.62	0.66	1.06	1.37	0.68	20.8	10.5	52.6	2.77	0.44	1.27	0.73
	017	267	2.06	6.81	2.22	0.60	1.00	1.23	0.38	19.2	10.7	59.6	2.92	0.40	1.46	0.67
	018	315	2.05	7.91	2.42	0.59	1.16	1.25	0.52	21.3	11.1	48.7	3.01	0.45	1.38	0.78
	Mean	288	1.94	7.57	2.27	0.56	1.00	1.21	0.52	20.5	10.1	52.3	2.74	0.42	1.25	0.69
0 ^{b)}	019	300	1.93	7.89	2.28	0.56	1.02	1.16	0.48	20.9	11.1	48.1	2.52	0.55	1.10	0.67
	020	313	1.93	9.09	2.44	0.70	1.14	1.20	0.58	21.8	10.9	50.9	2.68	0.51	1.54	0.73
	021	348	2.02	10.95	2.65	0.70	1.25	1.39	0.53	19.0	11.7	54.4	2.96	0.65	1.62	0.70
	022	238	1.79	6.39	2.04	0.40	0.85	1.10	0.36	25.4	8.3	40.3	2.55	0.45	1.07	0.65
	023	285	2.00	7.73	2.26	0.44	0.96	1.26	0.55	25.6	11.3	54.4	2.50	0.34	1.55	0.69
	024	310	1.95	9.03	2.65	0.55	1.13	1.17	0.41	26.5	12.2	58.9	2.66	0.42	1.51	0.83
	Mean	299	1.94	8.51	2.39	0.56	1.06	1.21	0.49	23.2	10.9	51.2	2.65	0.49	1.40	0.71
20	025	252	1.98	5.54	2.07	0.55	1.14	1.07	0.49	21.2	9.8	37.9	2.62	0.32	1.06	0.64
	026	237	1.84	7.18	2.06	0.41	0.81	1.10	0.44	24.7	8.6	44.3	2.62	0.34	1.09	0.62
	027	274	1.79	6.72	2.16	0.52	1.08	1.15	0.47	21.6	9.8	45.3	2.55	0.28	1.19	0.57
	028	308	2.01	7.16	2.22	0.61	1.05	1.20	0.56	23.1	11.1	48.4	2.71	0.33	1.21	0.65
	029	233	1.90	5.70	1.87	0.43	0.86	1.20	0.37	19.0	9.5	47.6	2.72	0.22	0.92	0.65
	030	236	1.87	5.59	2.03	0.46	0.83	1.05	0.49	18.7	8.5	38.9	2.27	0.28	1.04	0.62
	Mean	257	1.80	6.32	2.07	0.50	0.96	1.13	0.47	21.4	9.6	43.7	2.58	0.30	1.09	0.63

a): Control I; b): Control II

Appendix 12 Relative organ weights of individual male rats fed dry beef-contained diet in a 28-day dose range-finding study

Conc. in diet (%)	Animal numbers	B.W. (g)	Brain (%)	Liver (%)	Kidney (%)	Spleen (%)	Heart (%)	Lung (%)	Thymus (%)	Thyr. (mg%)	Pitul. (mg%)	Adrenal (mg%)	Testis (%)	Prost. (%)	SemLv (%)	Epidid. (%)
0 ^a	001	312	0.61	2.78	0.78	0.20	0.37	0.45	0.21	8.6	3.2	15.6	0.84	0.13	0.48	0.23
	002	308	0.57	3.07	0.70	0.20	0.35	0.40	0.20	9.9	3.1	15.3	0.80	0.17	0.49	0.23
	003	315	0.60	2.86	0.72	0.19	0.35	0.39	0.18	9.0	2.8	18.4	0.81	0.17	0.43	0.23
	004	313	0.55	2.74	0.72	0.18	0.35	0.43	0.19	6.7	3.4	16.8	0.83	0.11	0.42	0.24
	005	331	0.57	2.85	0.78	0.21	0.33	0.41	0.16	6.0	3.2	18.2	0.86	0.15	0.53	0.19
	006	307	0.63	2.76	0.76	0.17	0.36	0.43	0.15	8.7	4.0	17.6	0.92	0.16	0.43	0.25
	Mean	314	0.59	2.84	0.74	0.19	0.35	0.42	0.18	8.2	3.3	17.0	0.84	0.15	0.46	0.23
5	007	287	0.68	2.75	0.87	0.17	0.36	0.43	0.17	6.5	3.1	17.9	0.93	0.14	0.44	0.23
	008	296	0.65	2.57	0.84	0.16	0.33	0.44	0.20	5.4	3.2	16.7	0.89	0.13	0.45	0.22
	009	362	0.56	2.69	0.72	0.20	0.36	0.37	0.17	5.0	3.4	14.7	0.68	0.13	0.39	0.20
	010	329	0.60	2.87	0.80	0.17	0.36	0.38	0.26	5.8	3.1	13.9	0.84	0.16	0.49	0.20
	011	341	0.54	3.16	0.80	0.18	0.31	0.38	0.25	6.7	3.2	18.7	0.83	0.11	0.38	0.20
	012	295	0.62	2.85	0.83	0.17	0.37	0.42	0.21	7.1	2.9	14.5	0.84	0.15	0.37	0.24
	Mean	318	0.61	2.82	0.81	0.18	0.35	0.40	0.21	6.1	3.2	16.1	0.84	0.14	0.42	0.22
10	013	269	0.68	2.62	0.76	0.16	0.33	0.41	0.19	8.8	3.4	20.1	0.97	0.16	0.39	0.23
	014	295	0.64	2.68	0.70	0.16	0.33	0.39	0.16	6.5	3.6	18.3	0.87	0.11	0.42	0.24
	015	272	0.71	2.69	0.83	0.21	0.33	0.43	0.19	6.9	3.1	16.4	0.94	0.17	0.40	0.22
	016	310	0.60	2.71	0.85	0.21	0.34	0.44	0.22	6.7	3.4	17.0	0.89	0.14	0.41	0.24
	017	267	0.77	2.55	0.83	0.22	0.37	0.46	0.14	7.2	4.0	22.3	1.09	0.15	0.55	0.25
	018	315	0.65	2.51	0.77	0.19	0.37	0.40	0.17	6.8	3.5	15.5	0.96	0.14	0.44	0.25
	Mean	288	0.68	2.63	0.79	0.19	0.35	0.42	0.18	7.2	3.5	18.3	0.95	0.15	0.44	0.24
0 ^b	019	300	0.64	2.63	0.76	0.19	0.34	0.39	0.16	7.0	3.7	16.0	0.84	0.18	0.37	0.22
	020	313	0.62	2.90	0.78	0.22	0.36	0.38	0.19	7.0	3.5	16.3	0.86	0.16	0.49	0.23
	021	348	0.58	3.15	0.76	0.20	0.36	0.40	0.15	5.5	3.4	15.6	0.85	0.19	0.47	0.20
	022	238	0.75	2.68	0.86	0.17	0.36	0.46	0.15	10.7	3.5	16.9	1.07	0.19	0.45	0.27
	023	285	0.70	2.71	0.79	0.15	0.34	0.44	0.19	9.0	4.0	19.1	0.88	0.12	0.54	0.24
	024	310	0.63	2.91	0.85	0.18	0.36	0.38	0.13	8.5	3.9	19.0	0.86	0.14	0.49	0.27
	Mean	299	0.65	2.83	0.80	0.19	0.35	0.41	0.16	8.0	3.7	17.2	0.89	0.16	0.47	0.24
20	025	252	0.79	2.20	0.82	0.22	0.45	0.42	0.19	8.4	3.9	15.0	1.04	0.13	0.42	0.25
	026	237	0.78	3.03	0.87	0.17	0.34	0.46	0.19	10.4	3.6	18.7	1.11	0.14	0.46	0.26
	027	274	0.65	2.45	0.79	0.19	0.39	0.42	0.17	7.9	3.6	16.5	0.93	0.10	0.43	0.21
	028	308	0.65	2.32	0.72	0.20	0.34	0.39	0.18	7.5	3.6	15.7	0.88	0.11	0.39	0.21
	029	233	0.82	2.45	0.80	0.18	0.37	0.52	0.16	8.2	4.1	20.4	1.17	0.09	0.39	0.28
	030	236	0.79	2.37	0.86	0.19	0.35	0.44	0.21	7.9	3.6	16.5	0.96	0.12	0.44	0.26
	Mean	257	0.75	2.47	0.81	0.19	0.37	0.44	0.18	8.4	3.7	17.1	1.02	0.12	0.42	0.25

a): Control I ; b): Control II

Appendix
Conc. in diet (%)
0^a
5
10
0^b
20
a): Cont

Appendix 13 Absolute organ weights of individual female rats fed dry beef-contained diet in a 28-day dose range-finding study

Conc. in diet (%)	Animal numbers	B.W. (g)	Brain (g)	Liver (g)	Kidney (g)	Spleen (g)	Heart (g)	Lung (g)	Thymus (g)	Thyr. (mg)	Pitul. (mg)	Adrenal (mg)	Ovary (mg)	Uterus (g)
0 ^a	501	191	1.82	5.76	1.68	0.40	0.67	0.95	0.40	17.5	14.5	48.1	60.4	0.43
	502	205	1.67	6.16	1.81	0.38	0.74	1.01	0.38	21.6	13.9	63.3	77.1	0.44
	503	201	1.80	6.02	1.93	0.47	0.75	1.04	0.65	22.2	12.2	52.3	64.2	0.36
	504	224	1.75	7.38	1.70	0.56	0.71	0.95	0.55	18.1	15.5	57.7	67.7	0.44
	505	215	1.90	6.13	1.83	0.39	0.87	0.91	0.55	20.7	13.4	62.4	69.9	0.45
	506	187	1.75	5.06	1.54	0.36	0.67	0.93	0.40	22.9	9.1	51.2	64.8	0.68
	Mean	204	1.78	6.09	1.75	0.43	0.74	0.97	0.49	20.5	13.1	55.8	67.4	0.47
5	507	186	1.70	5.06	1.39	0.39	0.64	0.99	0.45	18.4	10.8	40.2	76.3	0.36
	508	221	1.82	6.26	1.90	0.46	0.89	1.06	0.36	17.5	14.5	61.6	96.3	0.48
	509	201	1.76	6.57	1.59	0.52	0.78	1.12	0.47	24.4	12.9	57.4	73.3	0.65
	510	197	1.78	5.34	1.65	0.41	0.70	0.95	0.54	20.6	11.3	47.2	55.6	0.28
	511	228	1.88	7.50	1.83	0.55	0.87	1.05	0.49	17.0	14.7	62.9	82.0	0.50
	512	213	1.87	6.25	1.63	0.36	0.75	0.98	0.42	25.2	15.6	45.5	59.5	0.61
	Mean	208	1.80	6.16	1.87	0.45	0.77	1.03	0.46	20.5	13.3	52.5	73.8	0.48
10	513	181	1.77	4.62	1.47	0.46	0.68	0.92	0.40	19.5	11.7	50.2	61.1	0.35
	514	179	1.91	5.01	1.53	0.33	0.68	0.92	0.42	21.5	12.1	54.1	77.7	0.44
	515	201	1.89	5.62	1.64	0.48	0.75	1.03	0.46	21.8	14.9	57.9	100.5	0.57
	516	204	1.76	5.23	1.61	0.42	0.72	0.98	0.48	21.0	13.5	48.2	70.3	0.33
	517	204	1.77	5.74	1.63	0.42	0.67	1.03	0.40	17.9	12.3	65.6	77.4	0.50
	518	219	1.85	6.17	2.01	0.49	0.73	1.02	0.54	21.9	14.5	65.6	72.8	0.57
	Mean	198	1.83	5.40	1.65	0.43	0.71	0.98	0.45	20.6	13.2	56.9	76.6	0.46
0 ^b	519	198	1.77	5.50	1.60	0.42	0.73	0.96	0.57	21.1	10.7	52.5	69.7	0.40
	520	234	1.89	7.24	2.10	0.46	0.74	1.04	0.62	20.7	17.8	67.7	86.7	0.46
	521	190	1.76	5.17	1.52	0.41	0.66	0.86	0.47	25.5	10.9	59.4	65.9	0.36
	522	223	1.84	6.46	1.78	0.48	0.80	1.02	0.58	22.1	12.6	75.7	71.7	0.81
	523	221	1.86	6.93	1.76	0.35	0.88	1.14	0.50	19.6	15.2	63.4	71.2	0.46
	524	224	1.88	6.54	1.69	0.37	0.73	0.99	0.40	19.6	14.0	56.3	78.1	0.73
	Mean	215	1.83	6.31	1.74	0.42	0.76	1.00	0.52	21.4	13.5	62.5	73.9	0.54
20	525	218	1.80	6.25	1.70	0.38	0.90	0.95	0.46	22.6	15.4	55.6	82.5	0.35
	526	209	1.86	5.07	1.51	0.36	0.73	0.99	0.58	22.3	11.7	44.5	61.2	0.37
	527	191	1.78	4.82	1.67	0.35	0.67	0.92	0.44	19.8	11.6	49.6	68.8	0.36
	528	201	1.82	5.68	1.71	0.41	0.73	1.07	0.63	18.3	13.1	67.7	70.1	0.43
	529	172	1.78	4.40	1.46	0.35	0.65	0.88	0.38	17.7	11.1	46.9	54.1	0.54
	530	226	1.79	6.10	1.71	0.72	0.81	1.15	0.44	19.8	14.7	58.3	77.5	0.66
	Mean	203	1.81	5.39	1.63	0.43	0.75	0.99	0.49	20.1	12.9	53.8	69.0	0.45

a): Control I ; b): Control II

Conc. in diet(%)	Animal numbers	B.W. (g)	Brain (%)	Liver (%)	Kidney (%)	Spleen (%)	Heart (%)	Lung (%)	Thymus (%)	Thyr. (mg%)	Pitui. (mg%)	Adrenal (mg%)	Ovary (mg%)	Uterus (%)
0 ^a	501	191	0.95	3.02	0.88	0.21	0.35	0.50	0.21	9.2	7.6	25.2	31.6	0.23
	502	205	0.81	3.00	0.88	0.19	0.36	0.49	0.19	10.5	6.8	30.9	37.6	0.21
	503	201	0.90	3.00	0.96	0.23	0.37	0.52	0.32	11.0	6.1	26.0	31.9	0.18
	504	224	0.78	3.29	0.76	0.25	0.32	0.42	0.25	8.1	6.9	25.8	30.2	0.20
	505	215	0.88	2.85	0.85	0.18	0.40	0.42	0.26	9.6	6.2	29.0	32.5	0.21
	506	187	0.94	2.71	0.82	0.19	0.36	0.50	0.21	12.2	4.9	27.4	34.7	0.36
	Mean	204	0.88	2.98	0.86	0.21	0.36	0.48	0.24	10.1	6.4	27.4	33.1	0.23
5	507	186	0.91	2.72	0.75	0.21	0.34	0.53	0.24	9.9	5.8	21.6	41.0	0.19
	508	221	0.82	2.83	0.86	0.21	0.40	0.48	0.16	7.9	6.6	27.9	43.6	0.22
	509	201	0.88	3.27	0.79	0.26	0.39	0.56	0.23	12.1	6.4	28.6	36.5	0.32
	510	197	0.90	2.71	0.84	0.21	0.36	0.48	0.27	10.5	5.7	24.0	28.2	0.14
	511	228	0.82	3.29	0.80	0.24	0.38	0.46	0.21	7.5	6.4	27.6	36.0	0.22
	512	213	0.88	2.93	0.77	0.17	0.35	0.46	0.20	11.8	7.3	21.4	27.9	0.29
	Mean	208	0.87	2.96	0.80	0.22	0.37	0.50	0.22	10.0	6.4	25.2	35.5	0.23
10	513	181	0.98	2.55	0.81	0.25	0.38	0.51	0.22	10.8	6.5	27.7	33.8	0.19
	514	179	1.07	2.80	0.85	0.18	0.38	0.51	0.23	12.0	6.8	30.2	43.4	0.25
	515	201	0.94	2.80	0.82	0.24	0.37	0.51	0.23	10.8	7.4	28.8	50.0	0.28
	516	204	0.86	2.56	0.79	0.21	0.35	0.48	0.24	10.3	6.6	23.6	34.5	0.16
	517	204	0.87	2.81	0.80	0.21	0.33	0.50	0.20	8.8	6.0	32.2	37.9	0.25
	518	219	0.84	2.82	0.92	0.22	0.33	0.47	0.25	10.0	6.6	30.0	33.2	0.26
	Mean	198	0.93	2.72	0.83	0.22	0.36	0.50	0.23	10.5	6.7	28.8	38.8	0.23
0 ^b	519	198	0.89	2.78	0.81	0.21	0.37	0.48	0.29	10.7	5.4	26.5	35.2	0.20
	520	234	0.81	3.09	0.90	0.20	0.32	0.44	0.26	8.8	7.6	28.9	37.1	0.20
	521	190	0.93	2.72	0.80	0.22	0.35	0.45	0.25	13.4	5.7	31.3	34.7	0.19
	522	223	0.83	2.90	0.80	0.22	0.36	0.46	0.26	9.9	5.7	33.9	32.2	0.36
	523	221	0.84	3.14	0.80	0.16	0.40	0.52	0.23	8.9	6.9	28.7	32.2	0.21
	524	224	0.84	2.92	0.75	0.17	0.33	0.44	0.18	8.8	6.3	25.1	34.9	0.33
	Mean	215	0.86	2.93	0.81	0.20	0.36	0.47	0.25	10.1	6.3	29.1	34.4	0.25
20	525	218	0.83	2.87	0.78	0.17	0.41	0.44	0.21	10.4	7.1	25.5	37.8	0.16
	526	209	0.89	2.43	0.72	0.17	0.35	0.47	0.28	10.7	5.6	21.3	29.3	0.18
	527	191	0.93	2.52	0.87	0.18	0.35	0.48	0.23	10.4	6.1	26.0	36.0	0.19
	528	201	0.91	2.83	0.85	0.20	0.36	0.53	0.31	9.1	6.5	33.7	34.9	0.21
	529	172	1.03	2.56	0.85	0.20	0.38	0.51	0.22	10.3	6.5	27.3	31.5	0.31
	530	226	0.79	2.70	0.76	0.32	0.36	0.51	0.19	8.8	6.5	25.8	34.3	0.29
	Mean	203	0.90	2.65	0.81	0.21	0.37	0.49	0.24	10.0	6.4	26.6	34.0	0.22

a): Control I ; b): Control II

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