CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER:

125349Orig1s000

CLINICAL PHARMACOLOGY AND BIOPHARMACEUTICS REVIEW(S)

CLINICAL PHARMACOLOGY REVIEW

BLA: 125-349	Submission Date(s): 06/15/12
Drug	Raxibacumab
Trade Name	N/A
OCP Reviewers	Ryan P. Owen, Ph.D.
OCP Team Leader	Kimberly L. Bergman, Pharm.D.
PM Reviewer	Jerry Yu, Ph.D.
PM Team Leader	Yaning Wang, Ph.D.
OCP Division	DCP4
OND division	DAIP (520)
Sponsor	Human Genome Sciences
Relevant IND(s)	BB-IND 11,069
Submission Type; Code	BLA Resubmission
Formulation; Strength(s)	Each single-use vial contains 35.1 mL of raxibacumab solution at a concentration of 50 mg/mL (to allow delivery of 1700 mg/34 mL). Contains no preservative
Indication	(b) (4)
Dosage and Administration	Adult Dose: 40 mg/kg Pediatric Doses >15 kg to ≤ 50 kg: 60 mg/kg ≤ 15 kg: 80 mg/kg

BACKGROUND:

Raxibacumab (HGS1021, PA mAb, or ABthrax) is a fully human monoclonal antibody that specifically binds the Protective Antigen (PA) of *Bacillus anthracis*, thereby preventing its association with the anthrax toxin receptor on host cells. On January 30, 2009, the Agency reviewed raxibacumab efficacy, safety, and product quality data for inclusion of raxibacumab in the Strategic National Stockpile (SNS). These data were determined to be sufficient to support the use of a single 40 mg/kg IV raxibacumab dose for the treatment of inhalational anthrax under a CDC-held IND; raxibacumab is currently being stored in the SNS for use under this IND.

A BLA for raxibacumab was submitted on May 14, 2009 and sought approval under the "Animal Efficacy Rule" (21 CFR 601, Subpart H, "Approval of Biological Products when Human Efficacy Studies are not Ethical or Feasible") for the indication of therapeutic treatment of inhalation anthrax. The proposed dosage and route of administration was a single dose of 40 mg/kg administered as an intravenous infusion over approximately 2 hours, alone or in combination with antibiotics.

To allow selection of an effective dose in humans for therapeutic treatment of anthrax, the applicant submitted clinical pharmacology data for raxibacumab in humans and in the

two pivotal animal species, rabbits and cynomolgus monkeys (studies 682-G005758 and 724-G005829, respectively). The pharmacokinetics and safety of raxibacumab administered as the product proposed for licensure was evaluated in a Phase 1 antibiotic interaction study with ciprofloxacin (HGS1021-C-1064), a repeat dose immunogenicity study (HGS1021-C1069) and a Phase 2/3 safety study (HGS1021-C1063).

On October 20, 2009, the primary clinical pharmacology reviewer was made aware of significant findings from an audit of analytical sites used for studies submitted under BLA 125349. In September 2009, the Division of Scientific Investigations (DSI) inspections for the analytical portions of human studies HGS1021-C1063 and HGS1021-C1064 were conducted. During these inspections, multiple issues were identified and FDA Forms-483 were issued to the analytical sites responsible for analysis of raxibacumab and ciprofloxacin concentrations. Inspection findings during the audit conducted by DSI for clinical studies HGS1021-C1063 and HGS1021-C1064 raised concerns regarding the reliability of the pharmacokinetic data submitted in the application and utilized for selection of an effective dose in humans for therapeutic treatment of anthrax. The applicant submitted a follow-up correspondence dated October 29, 2009 (BLA 125349/Submission Number 017) for the purpose of addressing the inspection issues pertaining to the raxibacumab assay. Upon agency review of the applicant's submission and proposal to address the DSI inspection findings, it was determined that the revised assay validation, qualification, and SOP documents did not fully address the inspection issues identified in the Form FDA-483 and recommendations were conveyed to the applicant via facsimile on November 6, 2009. On November 14, 2009, the FDA issued a Complete Response letter to the applicant requesting that the applicant revise the analytical procedures for raxibacumab to address the DSI inspection findings, re-analyze the pharmacokinetic samples obtained from human studies HGS1021-C1063, HGS1021-C1064, and HGS1021-C1069, and re-calculate the PK parameters for raxibacumab from these re-analyses. At a Type A meeting held on January 29, 2010, the FDA and applicant agreed upon "equivalence" criteria to compare the results obtained with the original raxibacumab assay (CLI-2879) with the results obtained with modified assay (CLI-3192) for purposes of supporting data integrity. Upon revalidation of the assay for analysis of serum raxibacumab samples, the analytical sites underwent re-inspection by DSI focusing on the methods and data supporting Clinical Study HGS1021-C1064. Following inspection, DSI did not identify concerns with the modified raxibacumab assay as used for the reanalysis of Study HGS1021-C1063.

On April 23, 2010, the applicant responded with a submission that contains information supporting the revalidated assay for concentrations of raxibacumab in human serum. Included in this submission are PK comparison reports which compare the results obtained with the original raxibacumab assay (CLI-2879) with the results obtained with the modified assay (CLI-3192) and relevant bioanalytical validation and quantitation reports.

In the opinion of the clinical pharmacology reviewer (see review by Dr. Kimberly Bergman dated 3/25/11 under IND 11,069 in DARRTS), the April 23, 2010 submission adequately addressed the request to: 1) revise the analytical procedures for raxibacumab

to address the DSI inspection findings, 2) re-analyze the pharmacokinetic samples obtained from human studies HGS1021-C1064, and 3) re-calculate the PK parameters for raxibacumab from this re-analysis. The statistical analyses presented in Study Report HGS1021-C1064-PK support the acceptability of the revised assay for determination of serum concentrations of raxibacumab and the overall data integrity of purposes of extrapolating animal efficacy findings to humans by bridging pharmacokinetic data between animals and humans. The recommendations from the clinical pharmacology reviewer pertaining to the April 23, 2010 submission were as follows:

"The pharmacokinetic data for raxibacumab obtained by both the original and modified bioanalytical methods should be considered reliable and sufficient for extrapolation of animal efficacy findings to humans for determination of the efficacious dose in humans for therapeutic treatment of anthrax. Therefore, pharmacokinetic data for raxibacumab obtained by the original bioanalytical method will be used for the purposes of labeling. "

The applicant proposed a pediatric dosing recommendation (BB-IND 11069**,** cover letter dated December 14, 2011**)** in response to a request made by the agency. At the same time, Division of Pharmacometrics developed pediatric dosing regimen to assist pre-Emergency Use Authorization (preEUA) activities.

Simulations were performed for a population with body weight range from 5-100 kg at a dose of 40 mg/kg. The predicted AUC and C_{max} in the simulated subjects were compared with those observed in healthy adults with body weight > 45 kg. The 90% prediction interval of AUC and C_{max} for simulated adult subjects was comparable with those seen in adults at 40 mg/kg. However, the 40 mg/kg dose does not provide similar exposure for a population with body weight less than 45 kg. To match the exposure level in pediatric population with that observed in adults, several dose regimens for pediatric subjects were explored. FDA's final dose recommendations in pediatric subjects have been accepted by the applicant and are shown in Table 1. Please see the original pharmacometrics review by Dr. Jerry Yu dated 11/6/12 (attached as Appendix 2.

Table 1: Raxibacumab dosing recommendations for pediatric patients based on body weight

Body Weight (in kg)	Raxibacumab dose (in mg/kg)
≤ 15	80
>15 to ≤ 50	60
> 50	40

In the current BLA resubmission (June 15, 2012), the applicant has included the results of two studies that were requested in the complete response letter of November 14, 2009: Study 1141-CG920871 (also referred to as the added benefit study) and Study 1103-G923704 (also referred to as the CNS penetration study). Additionally, the applicant has submitted the ciprofloxacin reanalysis that was requested in the original complete response letter.

This clinical pharmacology review is divided into three sections. The first section will address the remaining review issues related to clinical pharmacology. The second section (Appendix 1) contains study reports that were reviewed for the current submission. The third section (Appendix 2) is the pharmacometric review that was used to determine pediatric dosing.

It may also be useful to refer to other regulatory documents that will not be included in this review but will likely be entered into DARRTS at a later date. These other documents include the complete response letter of 11/14/09, the DSI inspection letter listing the inspection-related deficiencies, and previous clinical pharmacology reviews including the following: reviews written on 1/22/09, 10/16/09, 11/12/09, and 3/25/11 (in DARRTS under IND 11,069) by Dr. Kimberly Bergman.

RECOMMENDATION

The Office of Clinical Pharmacology Division 4 has reviewed BLA 125-349 and it is acceptable from a Clinical Pharmacology perspective. The issues from previous review cycles have been adequately addressed.

REMAINING REVIEW ISSUES

1. **Consistency of the pharmacokinetic information across studies**

Introduction

Because treatment of inhalation anthrax will include antimicrobials (e.g. fluoroquinolones), the efficacy of antibiotics administered concomitantly with raxibacumab in the setting of therapeutic treatment also was evaluated in rabbits and monkeys with symptomatic anthrax disease. In these studies (reviewed for the original BLA submission), raxibacumab did not alter the substantial efficacy of levofloxacin or ciprofloxacin administered at doses providing exposures reflective of those obtained at recommended doses in humans. An additional added benefit study was requested by FDA (see Complete Response letter dated 11/14/09), in which the survival rate with antibiotic was to be more similar to that observed during the anthrax attacks in humans in 2001 (~55%) rather than the high survival rate (85-100%) observed when antibiotic was administered as soon as systemic anthrax disease was detected in the previous GLP studies. To approximate an ~55% survival rate with a full humanized dose of levofloxacin administered to rabbits with systemic anthrax infection, rabbits were treated with raxibacumab and/or levofloxacin 84 hours post spore exposure. This study achieved a survival rate with antimicrobial treatment (65%) that was consistent with the survival rate of ~55% observed in human subjects in 2001 and suggests that raxibacumab, administered even late in the course of the disease when over half of the animals have already succumbed, confers a survival benefit over antimicrobial alone. For further details regarding this study, refer to the reviews of the current submission (BLA 125-349) by the Medical Officer (Dr. Yuliya Yasinskaya) and Statistician (Dr. Lan Zeng).

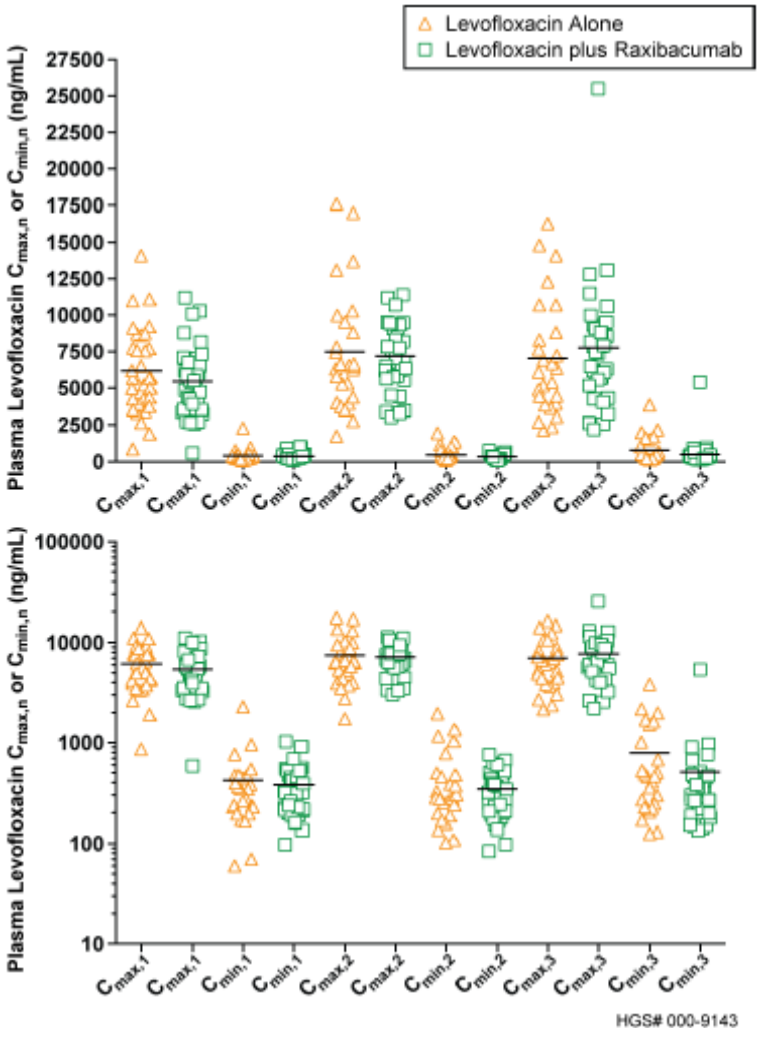
In this added benefit study in rabbits, pharmacokinetic sampling was performed for both levofloxacin and raxibacumab. The consistency of these data with previous studies was assessed. In addition, pharmacokinetic results for levofloxacin in rabbits were compared

to exposures observed in humans following the FDA-approved dosing regimen for inhalational anthrax.

Levofloxacin PK

There were no measureable plasma levofloxacin concentrations in any of the predose specimens. In both treatment groups the 2 hour and 23.5 hour collection times appear to have been switched for certain animals following either the 1st or 2nd levofloxacin dose. The mean and individual observed plasma levofloxacin concentration-time results for the levofloxacin only and levofloxacin plus raxibacumab groups are illustrated in Figure 1.

Figure 1: Plasma levofloxacin concentration-time profiles in rabbits administered levofloxacin doses alone or in combination with raxibacumab



The $C_{max,n}$ and $C_{min,n}$ results for the levofloxacin alone and raxibacumab/levofloxacin treatment groups are summarized in Table 2. There was some accumulation of plasma levofloxacin concentrations in both dose groups, with $C_{min,n}$ for the 2nd and 3rd doses ranging from 344 to 796 ng/mL. Comparisons of $C_{max,n}$ and $C_{min,n}$ across days within a

dose group generally show a pattern of increasing values across days. Unpaired t-tests of $C_{max,n}$ and $C_{min,n}$ show no differences that could be attributed to altered levofloxacin PK after raxibacumab administration.

Table 2: $C_{max,n}$ and $C_{min,n}$ in rabbits administered levofloxacin alone or raxibacumab and levofloxacin

	Levofloxacin (50 mg/kg x 3) + Placebo		Levofloxacin (50 mg/kg x 3) + Raxibacumab (40 mg/kg)		
	n	Mean ± SD	n	Mean ± SD	P-Value[1]
$C_{max,1}$ (ng/mL)	32	6179 ± 2921	36	5454 ± 2326	0.2661
$C_{min,1}$ (ng/mL)	25	424 ± 439	32	379 ± 214	0.6433
$C_{max,2}$ (ng/mL)	26	7487 ± 4105	33	7205 ± 2347	0.7561
$C_{min,2}$ (ng/mL)	25	471 ± 455	33	344 ± 178	0.1972
$C_{max,3}$ (ng/mL)	26	7042 ± 3962	34	7772 ± 4187	0.4928
$C_{min,3}$ (ng/mL)	25	796 ± 890	33	512 ± 904	0.2380

Abbreviations: $C_{max,n}$, maximum plasma levofloxacin concentration after the n^{th} dose, defined as the concentration measured 2 hours after the dose; $C_{min,n}$, minimum plasma levofloxacin concentration after the n^{th} dose, defined as the concentration measured just prior to the subsequent dose, or at 24 hours after the 3rd dose.

[1] From an unpaired t-test.

Sources: Appendix 10, Appendix 11, and Appendix 12 of HGS AB50409.INF.0.046 (Appendix K of the Battelle study report).

The levofloxacin PK results in this study were compared with the exposures (peaks [$C_{max,n}$] and troughs [$C_{min,n}$] following multiple oral doses) for humans administered the recommended levofloxacin doses, 500 or 750 mg. The reported $C_{max,n}$ and $C_{min,n}$ in humans are summarized in Table 3, along with the exposures for rabbits given the 50 mg/kg levofloxacin doses in this study. In the current study, the 50 mg/kg dose provided exposures intermediate to the human exposures at 500 and 750 mg. Overall, the 50 mg/kg levofloxacin dose in rabbits provided similar exposure to that for humans administered 500 or 750 mg of levofloxacin.

Table 3: Levofloxacin exposures for rabbits in the current study compared with exposures for humans administered 500 or 750 mg levofloxacin doses

Levofloxacin Dosing	$C_{max,n}$ (µg/mL)	$C_{min,n}$ (µg/mL)
Rabbit, based on PK results from current study[1]		
1st Dose	6.2	0.4
2nd Dose	7.5	0.5
3rd Dose	7.0	0.8
Human, based on information provided in Levaquin® product labeling		
500 mg	5.7	0.5
750 mg	8.6	1.1

Abbreviations: $C_{max,n}$, maximum plasma levofloxacin concentration after the n^{th} dose, defined as the concentration measured 2 hour after the dose; $C_{min,n}$, minimum plasma levofloxacin concentration after the n^{th} dose, defined as the concentration measured just prior to the subsequent dose, or at 24 hours after the 3rd dose.

[1] Mean values for levofloxacin treatment group.

Sources: Appendix 10 of HGS AB50409.INF.0.046 (Appendix K of the Battelle study report) and Levaquin® package insert, 2011.

Raxibacumab PK

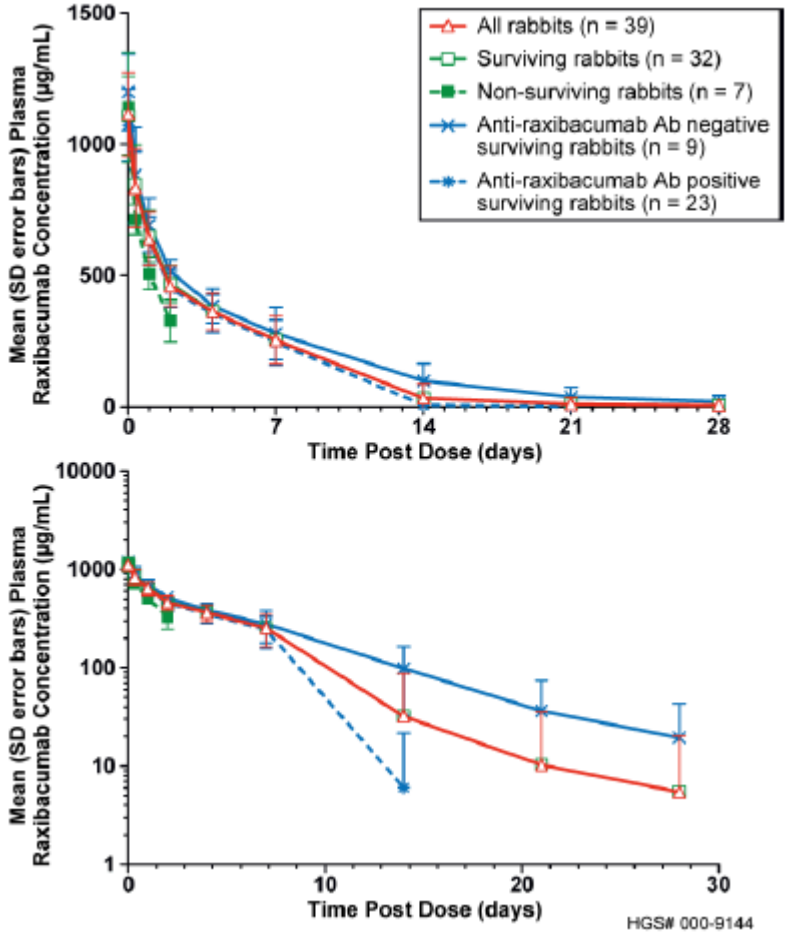
No measureable raxibacumab concentrations were detected in any of the specimens from the rabbits in the levofloxacin alone group and no measureable plasma raxibacumab concentrations were detected in the predose specimens from rabbits in the levofloxacin plus raxibacumab group.

Three of the 39 raxibacumab-treated rabbits (L29351, L34804, and L35261) were positive for anti-raxibacumab antibodies (Ab) prior to spore challenge and treatment at Study Day 0. Each of these rabbits was also positive for anti-raxibacumab antibodies at 28 days post treatment. The remaining 20 rabbits that were positive for anti-raxibacumab Ab in the raxibacumab-treated group were negative at Study Day 0 and positive at 28 days post treatment. The overall incidence of anti-raxibacumab Ab in the raxibacumab-treated group was 23/32 (72%), excluding the non-survivors for whom immunogenicity outcome is unknown. Inspection of the plasma raxibacumab concentration-time profiles for the rabbits that were positive for anti-raxibacumab Ab indicated that plasma raxibacumab concentrations became non-detectable between 14 to 21 days post dose, whereas for anti-raxibacumab Ab negative rabbits, plasma raxibacumab concentrations generally remained measureable throughout the 28 day postdose period.

The observed plasma raxibacumab concentration-time profile for the raxibacumab/levofloxacin group is illustrated in Figure 2 and includes the profiles for subgroups of surviving and non-surviving rabbits, as well as anti-raxibacumab positive and negative rabbits. It should be noted that the apparent increase in raxibacumab clearance in the later portion of the overall profile is associated with the presence of anti-raxibacumab antibodies in many of the individual rabbits at those times, as described previously. Although mean plasma raxibacumab concentrations for rabbits that died

were lower than those for surviving rabbits, there was substantial overlap of the SD error bars between the 2 subgroups.

Figure 2: Plasma raxibacumab concentration-time profiles in rabbits administered levofloxacin in combination with raxibacumab



The plasma raxibacumab concentrations best fit a 2-compartment open model with 1st order elimination from the central compartment. Body weight was found to be a significant covariate for clearance (CL). Other factors assessed (sex, age, size of spore challenge, duration of spore challenge, survival time, survival status, TBAC, bacteremia outcome at each collection time, and immunogenicity outcome) were not significant covariates accounting for inter-individual differences in PK. The lack of impact of immunogenicity as a covariate is likely related to the observation that in anti-raxibacumab Ab positive animals, plasma raxibacumab concentrations became undetectable between 14 to 21 days post dose; that is, those animals did not contribute data over the entire duration of the elimination phase of the profile.

The population PK model was evaluated using a visual predictive check. The prediction intervals were generated by running the final model in simulation mode, and generating 200 replicate simulations for each subject. The majority of the observed plasma raxibacumab concentrations are within the 90% prediction interval, suggesting that the

model describes the data well. The parameter estimates for the PK model are summarized in Table 4.

Table 4: Raxibacumab PK parameters in rabbits administered raxibacumab and levofloxacin

Primary Parameters	Mean[1]	CV%[1]
V_1 (mL)	112.17 (1.9%)	10.8 (26.6%)
CL (mL/day)	28.12 (5.5%)	27.5 (31.0%)
Effect of weight (WT) on CL (mL/day)[2]	CL x $(WT/3.12)^{1.743}$ (42.1%)	
At 2.75 kg	22.56	
At 3.00 kg	26.26	
At 3.12 kg	28.12	
At 3.25 kg	30.19	
At 3.50 kg	34.35	
At 3.55 kg	35.21	
V_2 (mL)	60.22 (7.5%)	20.6 (87.8%)
CLD_2 (mL/day)	73.62 (10.6%)	27.6 (61.9%)
Residual Variability, CV(%)	6.2% (51.7%) CV% for proportional error component 45.5 µg/mL (41.2%) SD for additive error component	
Secondary Parameters	**Mean ± SD[3]**	
C_{max} (µg/mL)	1114 ± 138	
$AUC_{0-\infty}$ (µg·day/mL)	4590 ± 1106	
$t_{1/2,\alpha}$ (days)	0.36 ± 0.05	
$t_{1/2,\beta}$ (days)	4.67 ± 1.10	
MRT (days)	6.40 ± 1.63	
V_{ss} (mL/kg)	56.00 ± 5.74	

Abbreviations: CV%, coefficient of variation; V_1, volume of distribution for the central compartment; CL, clearance; V_2, volume of distribution for the peripheral compartment; CLD_2, intercompartmental clearance; SD, standard deviation; C_{max}, maximum plasma drug concentration; $AUC_{0-\infty}$, area under the plasma drug concentration-time curve from time 0 to infinite time; $t_{1/2,\alpha}$, elimination half-life for the 1st phase; $t_{1/2,\beta}$, elimination half-life for the 2nd (terminal) phase; MRT, mean residence time; V_{ss}, volume of distribution at steady-state.

1 Values in parentheses represent the relative standard error (SE) of the estimate.

2 WT was normalized to the median body weight for the raxibacumab/levofloxacin group (3.12 kg).

3 Based on the post hoc estimates for the individual rabbits.

Source: Appendix 17 and Appendix 20 of HGS AB50409.INF.0.046 (Appendix K of the Battelle study report)

Following IV raxibacumab administration, V_1 at 112 mL (36 mL/kg for a 3.12 kg animal), is similar to the plasma volume (44 mL/kg). The steady-state volume of distribution (V_{ss}) is 56% greater than V_a at 56 mL/kg. These results suggest that although distribution of raxibacumab may initially be restricted to the plasma volume, raxibacumab doses subsequently distribute to tissues.

The disappearance of raxibacumab from plasma is multiphasic, with a mean initial phase elimination half-life ($t_{1/2\alpha}$) of 0.4 days and terminal phase elimination half-life ($t_{1/2\beta}$) of 4.7 days. The clearance (CL) of raxibacumab was 28.12 mL/day (9.01 mL/day/kg for a 3.12 kg rabbit). CL was affected by weight, and across the weight range for the raxibacumab-treated rabbits ranged from 22.56 mL/day (8.20 mL/day/kg) for a 2.75 kg rabbit to 35.21 mL/day (9.92 mL/day/kg) for a 3.55 kg rabbit. Across the weight range studied, CL is much less than the glomerular filtration rate (4493 mL/day/kg), indicating that, as expected, there is virtually no renal clearance of this monoclonal antibody.

Inter-individual variability in raxibacumab PK was low, with a coefficient of variation (CV%) of 28% or less for the primary PK parameters. Overall, these results indicate that there is minimal variability in raxibacumab disposition, once the impact of body weight on CL is taken into account, even in rabbits exhibiting symptoms of inhalation anthrax with treatment delayed until 84 hours post challenge.

Raxibacumab Concentration in Combination with Levofloxacin in Previous Studies

In previous studies, raxibacumab PK in rabbits were determined for a single IV bolus 20 or 40 mg/kg dose administered alone and a single IV bolus 40 mg/kg doses administered in combination with QD x 3 50 mg/kg intragastric (IG) levofloxacin doses, respectively. In those studies, the treatments were administered after detection of PA toxemia following a 200 x LD_{50} *B. anthracis* spore challenge, at about 26 hours post challenge, which contrasts with the much later treatment time of 84 hours post challenge in the current study. Despite the later treatment time in the current study, raxibacumab PK results for the current study are consistent with those for raxibacumab administered alone or with levofloxacin in the prior studies (see Table 5). Overall, the good agreement in raxibacumab PK among these 3 studies indicates that raxibacumab PK are not affected by co-administration of levofloxacin, nor are raxibacumab PK affected by disease severity at the time of treatment.

Table 5: Selected raxibacumab PK parameters for 40 mg/kg raxibacumab doses from current and previous studies

	Study 682-G005758 (AB50409.INF.0.036) Raxibacumab alone	Study 781-923701G (AB50409.INF.0.043) Raxi/levo combination	Study 1141-CG920871 (AB50409.INF.0.046) Raxi/levo combination
C_{max} (µg/mL)	909	928	1114
$AUC_{0-\infty}$ (µg·day/mL)	3412	4361	4590
CL (µg/mL)	35	32	28
$t_{1/2,\alpha}$ (days)	0.2	0.1	0.4
$t_{1/2,\beta}$ (days)	3.8	4.5	4.7
V_1 (mL)	132	150	112
V_2 (mL)	57	55	60

Abbreviations: V_1, volume of distribution for the central compartment; V_2, volume of distribution for the peripheral compartment ; CL, clearance; C_{max}, maximum plasma drug concentration; $AUC_{0-\infty}$, area under the plasma drug concentration-time curve from time 0 to infinite time; $t_{1/2,\alpha}$, elimination half-life for the 1st phase; $t_{1/2,\beta}$, elimination half-life for the 2nd (terminal) phase.

Sources: Table 9-3 in this report; AB50409.INF.0.036 and AB50409.INF.0.043

Reviewer Assessment

The raxibacumab pharmacokinetic parameters obtained in the added benefit study were consistent with previous studies.

2. Assessment of CSF penetration data

Introduction

In anthrax spore-challenged rabbits and monkeys assessed during the original review cycle, the animals that died or were euthanized moribund had findings consistent with anthrax disease by gross necropsy or microscopic examination and the gross and histopathology was qualitatively the same as previously reported for rabbits and monkeys, succumbing to inhalation anthrax. The incidence and severity of histopathology findings was the same or greater in the placebo treatment groups for all tissues, ***except brain***, in which the incidence and/or severity of bacteria, hemorrhage and inflammation was greater in the raxibacumab-treated groups.

In the Complete Response Letter to the raxibacumab BLA, FDA recommended that HGS conduct a study to further evaluate the effect of raxibacumab on the CNS in an animal model of inhalational anthrax and examine animals that survive and animals that die of anthrax. In response, HGS conducted a randomized, blinded, placebo-controlled study of raxibacumab in New Zealand White rabbits with systemic inhalational anthrax, examining both survivors and non-survivors, with particular focus on brain histopathology and CNS findings.

The results of this study, described in this report, confirm the findings in the earlier experience with raxibacumab-treated animals with inhalational anthrax; the increased survival rate (45.8% vs. 0% for placebo, $p < 0.0001$) and the lack of clinical sequalae in survivors were replicated. As in the previous study, an increased incidence and severity of lesions was observed in placebo-treated animals for all organs except the brain. Moreover, the raxibacumab-treated animals with the most severe lesions were those that died later in their course of disease. The brains of raxibacumab-treated non-survivors had a greater incidence and severity of lesions than the placebo-treated animals, but the raxibacumab-treated survivors did not exhibit brain pathology or the presence of bacteria or PA in the brain or cerebrospinal fluid (CSF) at sacrifice. Immunohistochemical staining of brain tissues indicated that raxibacumab was found in the brain of the raxibacumab-treated non-surviving rabbits, most often in or near lesions, and coincidently with rabbit IgG. As immunoglobulins cannot cross an intact blood brain barrier to any substantial extent, the coincident finding of raxibacumab and rabbit IgG associated with brain lesions suggest that raxibacumab accesses the brain after anthrax infection has altered the permeability of the blood brain barrier, rather than preceding or exacerbating anthrax infection in the brain. The similarity in staining patterns of endogenous rabbit IgG and raxibacumab supports that the presence of raxibacumab in the brain reflects the leakage of immunoglobulin across a damaged blood brain barrier and is not specific to raxibacumab. Consequently, it is proposed that the increased incidence of brain involvement in anthrax-infected animals that die is not evidence of a safety finding for raxibacumab, but is more likely a consequence of the site of raxibacumab benefit being in the well-vascularized visceral organs; while raxibacumab does not prevent the development of brain lesions in those animals that eventually die despite raxibacumab treatment, as raxibacumab can only access the CNS once the blood brain barrier is compromised. . For further details regarding this study, refer to the reviews of the current

submission (BLA 125-349) by the Medical Officer (Dr. Yuliya Yasinskaya), Pharm/Tox Reviewer (Dr. Terry Miller), and Statistician (Dr. Lan Zeng).

Plasma PK

The mean observed plasma raxibacumab concentration-time profile for the 40 mg/kg dose group is illustrated in Figure 3. For comparison, the mean profiles for 40 mg/kg IV raxibacumab and 40 mg/kg IV raxibacumab plus levofloxacin administered to anthrax spore challenged rabbits in previous studies are also provided in Figure 3. The plasma raxibacumab concentrations during the first 3 days post dose in the current study tended to be higher than those obtained in the prior studies, while those at ~6 days post dose were similar, with overlapping SD error bars and plasma raxibacumab concentrations in the terminal phase are similar to those observed in prior studies.

The mean observed plasma raxibacumab concentration-time profiles for rabbits that died and those that survived in the raxibacumab treatment group are illustrated in Figure 4. The plasma raxibacumab concentrations for rabbits that died and the surviving rabbits were similar, with overlapping SD error bars, indicating that survival cannot be attributed to differences in raxibacumab exposure.

The mean observed plasma raxibacumab concentration-time profiles for rabbits in the raxibacumab treatment group, subdivided by immunogenicity status, are illustrated in Figure 5. The plasma raxibacumab concentrations are similar among the subgroups during the first 6 days after raxibacumab administration. Thereafter, raxibacumab concentrations were very low or not detected in the rabbits that were positive for anti-raxibacumab Ab negative. Although the majority of surviving rabbits were anti-raxibacumab Ab positive (8 rabbits versus 3 who were negative), the decreased raxibacumab exposure did not appear to affect survival.

Figure 3: Plasma raxibacumab concentration-time profiles in rabbits administered a single IV bolus 40 mg/kg raxibacumab dose

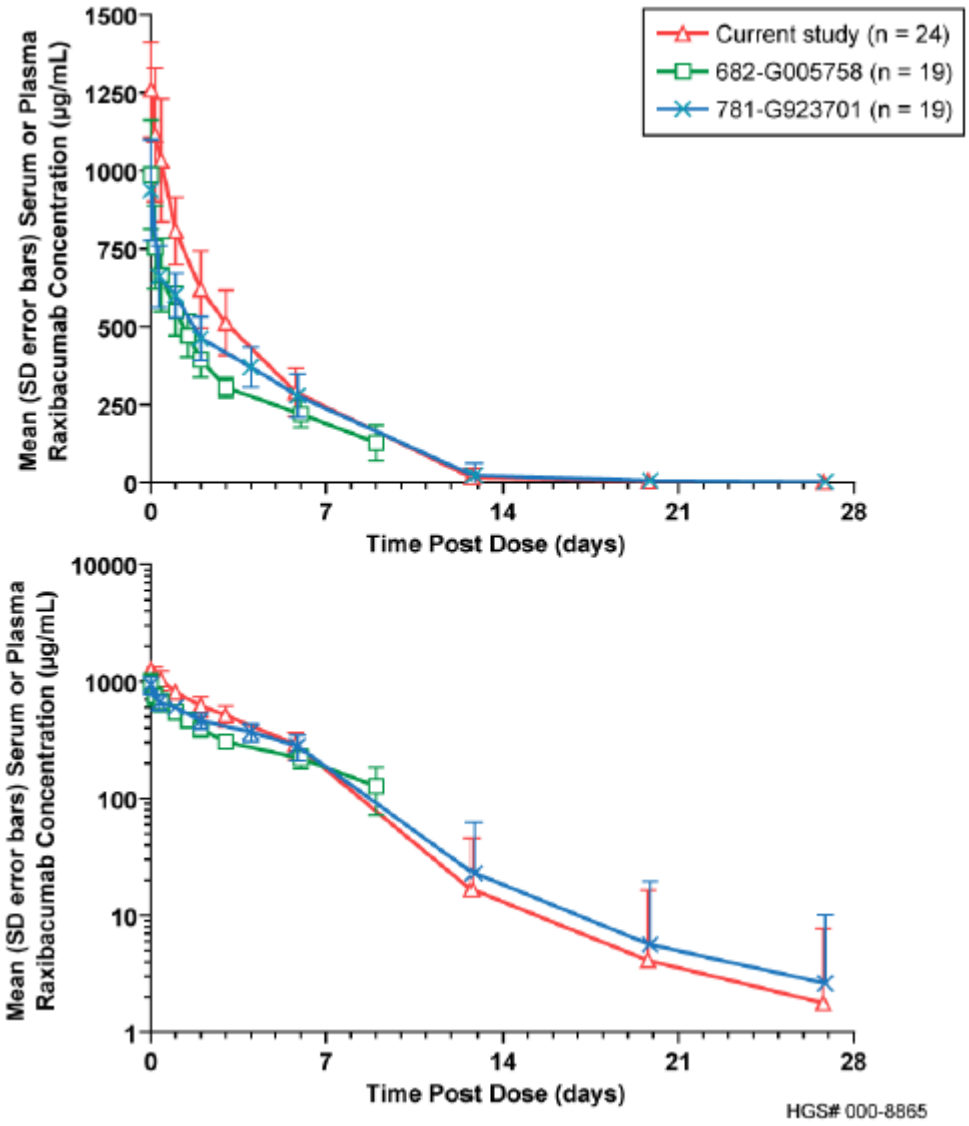

Current study (n = 24)
682-G005758 (n = 19)
781-G923701 (n = 19)
Mean (SD error bars) Serum or Plasma Raxibacumab Concentration (µg/mL)
1500
1250
1000
750
500
250
0
0
7
14
21
28
Time Post Dose (days)
Mean (SD error bars) Serum or Plasma Raxibacumab Concentration (µg/mL)
10000
1000
100
10
1
0
7
14
21
28
Time Post Dose (days)
HGS# 000-8865

Figure 4: Plasma raxibacumab concentration-time profiles in rabbits that died and those that survived

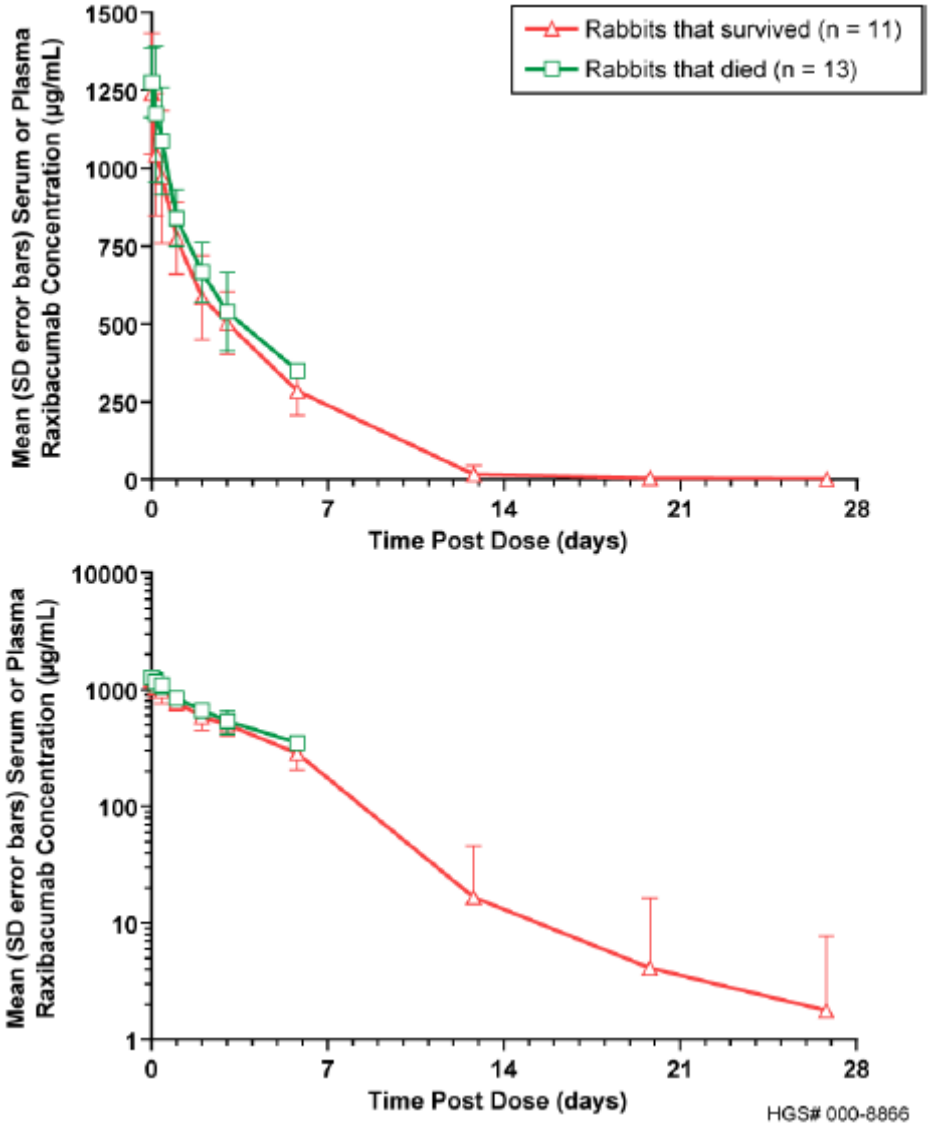
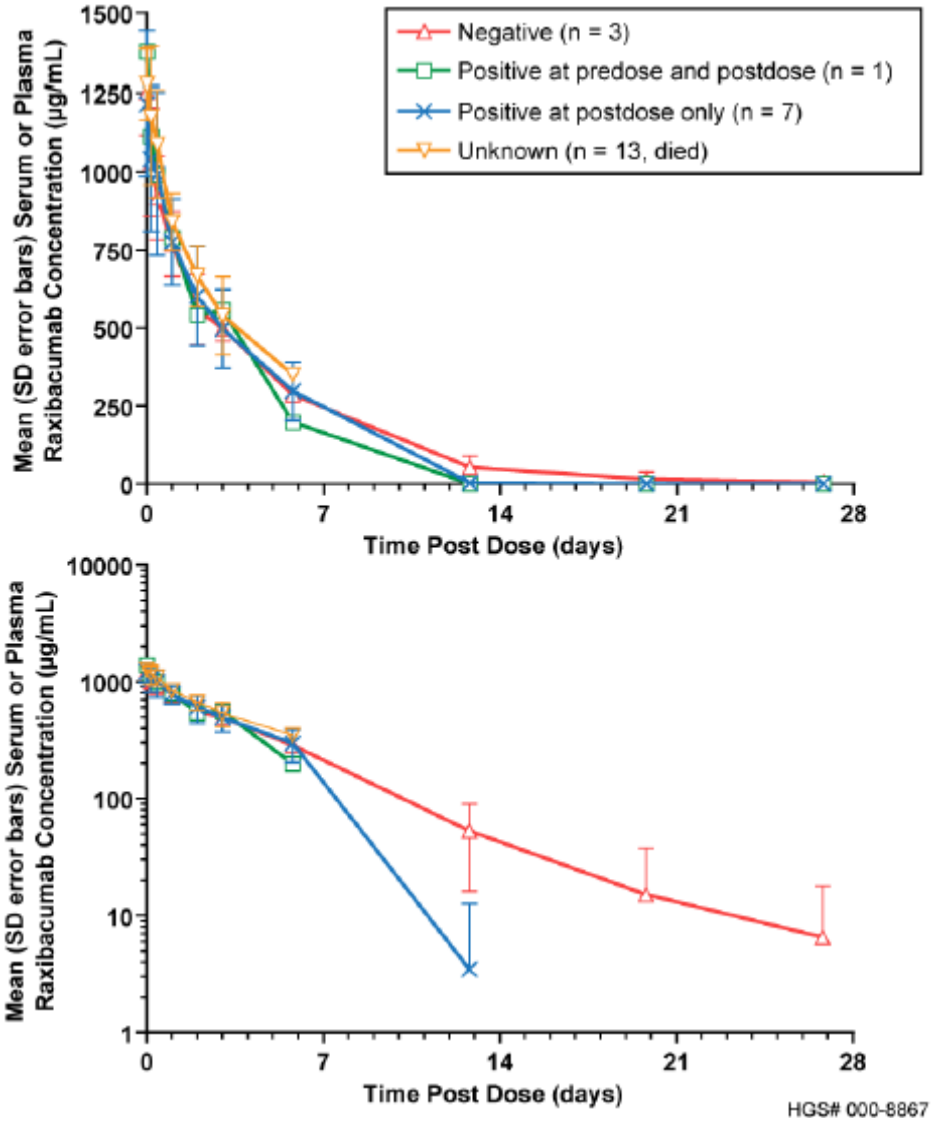


Figure 5: Plasma raxibacumab concentration-time profiles in rabbits by immunogenicity status



Raxibacumab in CSF

No animals in the placebo group had measureable raxibacumab in the CSF samples that were collected. The CSF raxibacumab concentration results and CSF to plasma raxibacumab concentration ratio results are summarized in Table 6. Of the 13 treated rabbits that died, 4 had CSF raxibacumab concentrations measured (due to inability to collect sample from all animals) and the results were variable, ranging from < 1 µg/mL to 545 µg/mL. These CSF concentrations represent from 0.2% to 88% of the concurrent plasma raxibacumab concentration. The value of 545 µg/mL was measured in a sample with blood in the pellet, suggesting that there was an error in collecting this CSF sample. Rabbit L35559 died early (Day 2 after spore challenge) and had no gross or microscopic findings in the brain. When the high CSF raxibacumab concentration for rabbit L35559 is excluded, the other 3 rabbits that died had CSF raxibacumab levels that were <2% of the concurrent plasma raxibacumab concentration. For comparison, CSF raxibacumab concentrations for the 11 rabbits that survived ranged from not detected to 0.331 µg/mL.

For the 1 surviving rabbit for whom the CSF to plasma raxibacumab concentration ratio could be determined, the CSF level was <2% of the concurrent plasma raxibacumab concentration. Overall, there was little distribution of raxibacumab into CSF.

Table 6: CSF raxibacumab concentrations and CSF:plasma concentration ratios

Survival Outcome	Rabbit No.	CSF Raxibacumab Concentration (µg/mL)	CSF:Plasma Raxibacumab Concentration Ratio
Died	L35542	1.145	0.002
	L35559	545.060	0.884
	L35562	7.480	0.018
	L35568	0.107	0.007
	N	4	4
	Mean	138.448	0.228
	SD	271.094	0.438
	Range	0.107 to 545.060	0.002 to 0.884
Survived	L35530	0.000	-[1]
	L35531	0.000	-[1]
	L35532	0.020	-[1]
	L35534	0.000	-[1]
	L35541	0.000	-[1]
	L35546	0.000	-[1]
	L35547	0.000	-[1]
	L35550	0.000	-[1]
	L35551	0.000	-[1]
	L35566	0.015	-[1]
	L35581	0.331	0.017
	N	11	1
	Mean	0.033	0.017
	SD	0.099	-
	Range	0.000 to 0.331	-

[1] Could not be calculated, due to absence or lack of measurable plasma raxibacumab concentration.

The pharmacokinetics of raxibacumab were consistent with those observed in the pivotal rabbit study with no meaningful difference in plasma raxibacumab concentrations between rabbits that survived and those that died. Raxibacumab was present in the CSF of some animals, including both survivors and non-survivors, although the concentrations were quite variable and generally occurred at much lower concentrations than the concurrent plasma raxibacumab concentration.

Reviewer Assessment

If rabbit L35559 is excluded, then the penetration of raxibacumab into the CSF is less than 2% in all of the other rabbits, whether they survived or not. The applicant states that the CSF sample for rabbit L35559 was likely contaminated with blood, which would understandably lead to an increased concentration of raxibacumab. Since raxibacumab is a large molecular weight biologic product, the penetration across an intact blood brain barrier would be expected to be low. This is consistent with the findings of the CSF penetration study if rabbit L35559 is excluded. The penetration is likely higher in rabbits that succumbed to anthrax disease as a results of a degraded blood brain barrier due to more severe progression of anthrax.

3. Acceptability of the reanalysis of ciprofloxacin concentrations

Background

As stated previously, the FDA requested the applicant revise analytical procedures and re-analyze pharmacokinetic samples obtained from human studies to address DSI inspection findings from the first review cycle. The results of the raxibacumab reanalysis have been previously reviewed and deemed acceptable. In the current resubmission, the results from the ciprofloxacin reassay were reviewed.

Study HGS1021-C1064 was an open-label study to evaluate the effect of raxibacumab on ciprofloxacin PK as well as the safety and PK of raxibacumab in combination with ciprofloxacin in healthy adult male and female subjects. Three treatment groups were evaluated. In Group 1, subjects received 15 oral (PO) 500 mg ciprofloxacin doses, given 12 hours apart (q12h). Just prior to the 11th ciprofloxacin dose on Day 5, the subjects were administered a single 40 mg/kg intravenous infusion raxibacumab dose. For Group 2, subjects were administered a single 40 mg/kg IV infusion raxibacumab dose only on Day 0. In Group 3, subjects received a 400 mg IV ciprofloxacin dose, immediately followed by a 40 mg/kg IV infusion raxibacumab dose; a second 400 mg IV ciprofloxacin dose was administered 12 hours after the first dose, and 500 mg PO ciprofloxacin doses were administered every 12 hours after the 2nd IV ciprofloxacin dose, for a total of 15 doses. Subjects were randomized in a 1:1 ratio to Group 1 or Group 2. Enrollment in Groups 1 and 2 (at least 28 evaluable subjects per group) was completed, and then an additional 28 evaluable subjects were enrolled into Group 3. In Group 1 and Group 3, specimens for plasma ciprofloxacin concentration measurement were collected at selected time points. For Group 1, plasma ciprofloxacin concentration time profiles were obtained following the first dose (Day 0, prior to raxibacumab dosing) as well as following the 13th PO dose (Day 7, after raxibacumab dosing).

Serum ciprofloxacin concentrations and ciprofloxacin PK were reported previously in the clinical pharmacokinetics report for HGS1021-C1064.PK. Based on regulatory feedback, it was decided to perform modified assays of all specimens from this study.
The new method chosen was HPLC with MS/MS detection which was conducted by (b) (4). The Sponsor then prepared a report which compared ciprofloxacin concentrations and parameters that were obtained from the original method (conducted at (b) (4)) to the reassay method conducted at (b) (4)

Results

Reassays of the duplicate samples from the study were performed using a validated method at a different laboratory to assess the validity of the original assay. The documented difference between the 2 assay methods was: internal standard for original assay and reassay were enrofloxacin and ciprofloxacin-d8, respectively. The lower limit of quantitation for both the original assay and the reassay was 10 ng/mL of ciprofloxacin in 100% plasma. Pharmacokinetic analysis of the plasma drug concentration results was performed in the same manner for both the original results and the reassay results.

Comparison of the original and reassay plasma ciprofloxacin concentrations revealed some unexpected group-dependent differences between the two sets of results. For concentrations stratified by nominal time and dose number in Group 1, the reassay to original assay (R/O) geometric mean ratios ranged from 106.61 to 116.72 with all 90% CIs within the equivalence range. In addition, the two assays were equivalent for all PK parameters for both Doses 9 and 11 in Group 1 (see Table 7). Since the original conclusion that raxibacumab did not affect the PK of ciprofloxacin was based on the results obtained for Group 1, the PK equivalence between reassay and the original assay for Group 1 fully supports that original conclusion.

Table 7: Summary of ciprofloxacin PK parameters, based on original and reassay results for Group 1

	Original (O)		Reassay (R)		Equivalence Test
Parameter	**Mean ± SD**	**Geometric Mean (CV%)**	**Mean ± SD**	**Geometric Mean (CV%)**	**R/O Geometric Mean Ratio% (90% CI)**
Dose 9, PO 500 mg ciprofloxacin without raxibacumab (N = 30)					
$C_{max,ss}$ (ng/mL)	1436 ± 519	1349 (38%)	1535 ± 512	1459 (33%)	108.19 (102.34, 114.38)
$t_{max,ss}$ (h)	1.75 (0.50 to 4.00)[1]		1.50 (0.50 to 4.00)[1], p = 0.625[2]		NA
AUC_{0-6h} (ng·h/mL)	6398 ± 2239	6045 (35%)	7032 ± 2316	6717 (31%)	111.11 (106.35, 116.07)
$AUC_{0-\tau}$ (ng·h/mL)	7708 ± 2692	7283 (35%)	8459 ± 2720	8094 (30%)	111.14 (106.33, 116.18)
$t_{1/2,z}$ (h)	4.50 ± 1.86	4.26 (32%)	4.53 ± 0.93	4.44 (20%)	104.30 (96.97, 112.18)
CL/F (L/h)	72.5 ± 24.1	68.7 (35%)	64.3 ± 17.5	61.8 (30%)	89.97 (86.08, 94.05)
V_z/F (L)	486 ± 328	422 (54%)	426 ± 164	396 (41%)	93.84 (85.36, 103.16)
$C_{min,ss}$ (ng/mL)	225 ± 89	209 (41%)	258 ± 91	244 (35%)	116.91 (110.04, 124.21)
Accumulation Index	1.20 ± 0.19	1.19 (13%)	1.19 ± 0.09	1.19 (7%)	100.30 (97.26, 103.44)
Fluctuation (%)	191 ± 50	185 (28%)	183 ± 45	177 (27%)	95.93 (90.33, 101.88)
Dose 11, PO 500 mg ciprofloxacin immediately after 40 mg/kg raxibacumab IV infusion (N = 30)					
$C_{max,ss}$ (ng/mL)	1419 ± 599	1310 (42%)	1475 ± 538	1385 (38%)	105.68 (99.43, 112.33)
$t_{max,ss}$ (h)	2.00 (0.58 to 4.12)[1]		1.52 (1.00 to 6.00)[1], p = 0.791[2]		NA
AUC_{0-6h} (ng·h/mL)	6694 ± 2270	6343 (34%)	7227 ± 2085	6949 (29%)	109.56 (104.98, 114.33)
$AUC_{0-\tau}$ (ng·h/mL)	8183 ± 2643	7787 (33%)	8808 ± 2380	8518 (27%)	109.38 (104.64, 114.33)
$t_{1/2,z}$ (h)	4.48 ± 1.28	4.33 (27%)	4.64 ± 1.09	4.53 (22%)	104.75 (96.28, 113.96)
CL/F (L/h)	67.5 ± 21.8	64.2 (33%)	60.6 ± 15.4	58.7 (27%)	91.42 (87.46, 95.57)
V_z/F (L)	426 ± 150	401 (37%)	412 ± 167	384 (39%)	95.77 (87.17, 105.21)

	Original (O)		Reassay (R)		Equivalence Test
Parameter	**Mean ± SD**	**Geometric Mean (CV%)**	**Mean ± SD**	**Geometric Mean (CV%)**	**R/O Geometric Mean Ratio% (90% CI)**
$C_{min,ss}$ (ng/mL) (N = 29)[3]	255 ± 88	238 (42%)	280 ± 73	270 (30%)	113.19 (105.48, 121.47)
Accumulation Index	1.19 ± 0.12	1.19 (10%)	1.20 ± 0.11	1.20 (8%)	101.11 (97.87, 104.44)
Fluctuation (%)	168 ± 37	164 (24%)	160 ± 41	154 (29%)	94.37 (88.34, 100.81)

Abbreviations: $C_{max,ss}$, maximum plasma drug concentration during a steady-state dosing interval; $t_{max,ss}$, time of occurrence for $C_{max,ss}$; AUC_{0-6h}, area under the plasma drug concentration-time curve from 0 to 8 h post dose; $AUC_{0-\tau}$, area under the plasma drug concentration-time curve during a steady-state dosing interval; $t_{1/2,z}$, elimination half-life for the terminal phase; CL/F, apparent clearance for oral dosing; V_z/F, apparent volume of distribution in the terminal phase for oral dosing; $C_{min,ss}$, minimum plasma drug concentration during a steady-state dosing interval; CI, Confidence Interval; NA, not applicable; CV% coefficient of variation peroentage.

[1] Median and range are presented.

[2] Wilcoxon signed rank test.

[3] Subject US003-000061 has Cmin = 0, therefore excluded from equivalence test.

Source: Summary Table 7-3 and Summary Table 7-4; Appendix 5 and Appendix 6.

For concentrations stratified by nominal time and dose number in Group 3, the R/O geometric mean ratios ranged from 117.99% to 140.17%. All 90% CIs fell outside of the equivalence range, with the reassay results higher than the original results at all collection times. Consequently, non-equivalence was found for some PK parameters between the two assays (see Table 8). Examination and comparison of the bioanalytical reports for the original assays and the reassay did not reveal any explanation for the discrepancies noted. The reason for these discrepancies remains unclear.

Table 8: Summary of ciprofloxacin PK parameters, based on original and reassay results for Group 3

Parameter	Original (O)		Reassay (R)		Equivalence Test
	Mean ± SD	Geometric Mean (CV%)	Mean ± SD	Geometric Mean (CV%)	R/O Geometric Mean Ratio% (90% CI)
Dose 1, IV 400 mg ciprofloxacin with raxibacumab (n = 28)					
C_{max} (ng/mL)	1854 ± 402	1811 (22%)	2446 ± 596	2372 (26%)	130.98 (124.63, 137.66)
t_{max} (h)	1.33 (1.23 to 1.60)[1]		1.33 (1.23 to 1.60)[1], p = NA [2]		NA
$AUC_{0-\infty}$ (ng·h/mL)	8622 ± 1816	8446 (21%)	10704 ± 2090	10503 (20%)	124.36 (119.43, 129.49)
$t_{1/2,z}$ (h)	4.02 ± 0.77	3.96 (18%)	4.16 ± 0.53	4.13 (13%)	104.25 (99.12, 109.64)
MRT (h)	5.75 ± 0.89	5.69 (15%)	5.75 ± 0.69	5.71 (12%)	100.37 (97.48, 103.35)
CL (L/h)	48.3 ± 10.0	47.4 (21%)	38.8 ± 7.9	38.1 (20%)	80.41 (77.22, 83.73)
V_{ss} (L)	278 ± 71	269 (26%)	224 ± 60	217 (25%)	80.71 (77.30, 84.28)
V_z (L)	281 ± 81	271 (29%)	233 ± 58	227 (24%)	83.83 (79.53, 88.36)
Dose 15, PO 500 mg ciprofloxacin (n = 28)					
$C_{max,ss}$ (ng/mL)	1195 ± 566	1087 (46%)	1421 ± 566	1321 (40%)	121.52 (116.13, 127.16)
$t_{max,ss}$ (h)	1.00 (0.5 to 6.00)[1]		1.00 (0.5 to 4.00)[1], p = 0.204[2]		NA
AUC_{0-8h} (ng·h/mL)	5494 ± 2643	4987 (46%)	6548 ± 2573	6104 (39%)	122.42 (116.97, 128.11)
AUC_{0-t} (ng·h/mL)	6599 ± 3208	5980 (46%)	7880 ± 3109	7345 (39%)	122.82 (117.09, 128.83)
F (%)	60 ± 21	57 (34%)	58 ± 16	56 (28%)	98.76 (93.70, 104.10)
$t_{1/2,z}$ (h)	4.45 ± 1.04	4.34 (24%)	4.56 ± 0.71	4.51 (16%)	104.02 (98.58, 109.77)
CL/F (L/h)	91.2 ± 36.9	83.6 (46%)	72.8 ± 26.6	68.1 (39%)	81.42 (77.62, 85.40)
V_z/F (L)	599 ± 292	523 (62%)	488 ± 208	443 (50%)	84.70 (78.70, 91.15)

Parameter	Original (O)		Reassay (R)		Equivalence Test
	Mean ± SD	Geometric Mean (CV%)	Mean ± SD	Geometric Mean (CV%)	R/O Geometric Mean Ratio% (90% CI)
$C_{min,ss}$ (ng/mL)	193 ± 102	174 (48%)	243 ± 111	222 (44%)	128.02 (121.17, 135.26)
Accumulation Index	1.19 ± 0.09	1.19 (8%)	1.20 ± 0.06	1.19 (5%)	100.74 (98.89, 102.62)
Fluctuation (%)	188 ± 56	181 (27%)	182 ± 45	177 (24%)	97.98 (93.74, 102.42)

Abbreviations: C_{max}, maximum plasma drug concentration for a single dose; $C_{max,ss}$, maximum plasma drug concentration during a steady-state dosing interval; t_{max}, time of occurrence for C_{max}; $t_{max,ss}$, time of occurrence for $C_{max,ss}$; AUC_{0-8h}, area under the plasma drug concentration-time curve from 0 to 8 h post dose; $AUC_{0-\infty}$, area under the plasma drug concentration-time curve from time zero to infinite time for a single dose; AUC_{0-t}, area under the plasma drug concentration-time curve during a steady-state dosing interval; F, bioavailable fraction for oral dosing; $t_{1/2,z}$, elimination half-life for the terminal phase; MRT, mean residence time; CL, clearance; CL/F, apparent clearance for oral dosing; V_{ss}, volume of distribution at steady-state; V_z, volume of distribution in the terminal phase; V_z/F, apparent volume of distribution in the terminal phase for oral dosing; ; $C_{min,ss}$, minimum plasma drug concentration during a steady-state dosing interval; CI, Confidence Interval; NA, not applicable; CV% coefficient of variation percentage.

[1] Median and range are presented.

[2] Wilcoxon signed rank test. No result available for the IV dose since values for each subject were identical between the 2 assays.

Source: Summary Table 7-5 and Summary Table 7-6; Appendix 5 and Appendix 6

Reviewer Assessment

The pre-specified equivalence criteria were met for the comparison of the original assay and the reassay with respect to both individual mean concentrations and the derived pharmacokinetic parameters for ciprofloxacin for Group 1. However, the mean ciprofloxacin Group 3 concentrations were systematically higher when reassayed than they were with the original assay. These higher concentrations led to the calculation of ciprofloxacin pharmacokinetic parameters that fell outside the 80-125% no-effect boundary when compared to the parameters derived using the ciprofloxacin concentrations determined by the original assay. The reasons for this disparity are unclear.

Ciprofloxacin was dosed differently in Group 1 versus Group 3 (PO vs. IV/PO); however, the dosing route seems unlikely to play a role in a systematic error in assessed concentrations. The ciprofloxacin label reports steady state AUC and C_{max} data at steady state following 400 mg q12h IV dosing and 500 mg q12h PO dosing. Although this does not directly parallel Group 3 in the current study, it is noteworthy that the steady state PK parameters reported in the ciprofloxacin label are larger than either the original analysis or the reanalysis (see Table below).

PK Parameter	Original Assay (Group 3)	Re-assay (Group 3)	Ciprofloxacin label[1]
$AUC_{0\text{-}tau}$ (ng*h/mL)	6599	7880	13700
C_{max} (ng/mL)	1195	1421	2970

[1]: Results are reported are for steady state following the administration of 500 mg ciprofloxacin q12h PO.

There is a larger numerical difference between the pharmacokinetic data contained in the ciprofloxacin label for a similar but not identical dosing scheme than there is between the original assay and the reassay for the Group 3 ciprofloxacin PK data.

Even though the pre-specified equivalence criteria was not met for the Group 3 ciprofloxacin concentrations, the Reviewer agrees with the applicant's position that the differences between the assays is not clinically relevant. The difference between the two analytical methods for the Group 3 concentrations and parameters is not of sufficient magnitude as to require additional studies or qualifying language.

APPENDIX 1
Individual Study Reports

Study 2100-577: Validation of a Method for the Determination of Ciprofloxacin in Human Plasma by HPLC with MS/MS Detection

Date of Report: 25 June 2005

OBJECTIVE:
To validate a method for the determination of ciprofloxacin in human plasma with sodium heparin anticoagulant by HPLC with MS/MS detection.

ASSAY METHOD:
Ciprofloxacin and the internal standard (ISTD) ciprofloxacin-d8 were extracted from human plasma by protein precipitation and analyzed using liquid chromatography (LC) with tandem mass spectrometric detection (MS/MS). A sample volume of 0.05 mL was used. The lower limit of quantitation (LLOQ) for ciprofloxacin in human plasma was 10.0 ng/mL, with linearity demonstrable to 5000 ng/mL (upper limit of quantitation, ULOQ). Results were calculated using peak area ratios. Calibration curves for ciprofloxacin in human plasma ranged from 10 to 5000 ng/mL and were generated using a weighted ($1/x^2$) linear least-squares regression. The concentration of ciprofloxacin in the QC samples, as measured by their peak area ratios, was determined from the calibration curves.

ASSAY PERFORMANCE:
Selectivity
Aliquots of blank human plasma from six different lots were tested for endogenous interferences. In all cases, the ciprofloxacin and ciprofloxacin-d8 regions were free from significant interference (<20.0% of the mean utilized LLOQ or < 5.0% of interval standard response).

Aliquots of these lots were spiked with 400 ng/mL of ciprofloxacin, including internal standard. The mean concentration of the spiked samples had a percent Relative Standard Deviation (%RSD) of ≤ 15.0% and an accuracy in the range of 85 to 115% of the theoretical. It was therefore concluded that the method demonstrated acceptable selectivity.

To evaluate matrix effect, extracts of blank matrix from the same lots were spiked with 400 ng/mL of ciprofloxacin, including internal standard. In addition, three replicates of a pure standard solution containing ciprofloxacin and internal standard at the same concentration as the spiked extract were injected. The mean sample peak area was expressed as a percentage of the mean pure standard peak area. The percentage matrix effect for ciprofloxacin and internal standard were calculated for the mean of all matrix samples. The matrix effect did not significantly impact assay performance.

Linearity

Results were calculated using peak area ratios. Calibration curves for ciprofloxacin in human plasma ranged from 10.0 to 5000 ng/mL and were generated using a weighted ($1/x^2$) linear least squares regression (see Table 1).

Precision

In this study, both intra- and inter-assay results demonstrated a %RSD for calibration standards (inter-assay) and QC samples (intra- and inter-assay) to be ≤ 15%. It was therefore concluded that the method demonstrated acceptable precision (see Table 1 and 2).

Accuracy

The accuracy of the method was determined by comparing the means of the measured concentrations of the calibration standards (inter-assay) and QC samples (intra- and inter-assay) with their theoretical concentrations. The accuracy results of this study demonstrated calculated mean values in the range of 85.0 to 115.0% of theoretical. It was therefore concluded that the method demonstrated acceptable accuracy (see Tables 1 and 2).

Sensitivity

The LLOQ for ciprofloxacin in human plasma was set at 10.0 ng/mL. A method with acceptable sensitivity requires the RSD ≤ 20.0% and an accuracy in the range of 80.0 to 120.0% of theoretical. In this study, the LLOQ results met this criteria, and it was therefore concluded that the method demonstrated acceptable sensitivity.

Table 1: Calibration curve data for ciprofloxacin in human plasma

	Theoretical Concentration (ng/mL)							
Batch	10.0	20.0	50.0	200	1000	2500	4000	5000
001	10.1	20.2	46.1	195	1050	2550	4060	5010
002	10.2	19.4	49.2	205	1020	2500	4010	4920
004	10.1	19.9	48.0	194	1020	2640	3870	5090
n	3	3	3	3	3	3	3	3
Mean	10.1	19.8	47.8	198	1030	2560	3980	5010
SD	0.0577	0.404	1.56	6.08	17.3	70.9	98.5	85.0
RSD (%)	0.6	2.0	3.3	3.1	1.7	2.8	2.5	1.7
Accuracy (%)	101.0	99.0	95.6	99.0	103.0	102.4	99.5	100.2

Table 2: Quality control sample data for ciprofloxacin in human plasma

	Theoretical Concentration (ng/mL)					
Batch	30.0	DEV (%)	400	DEV (%)	3800	DEV (%)
001	29.7	-1.0	415	3.8	4070	7.1
	30.3	1.0	407	1.8	3960	4.2
	29.3	-2.3	416	4.0	3970	4.5
	29.4	-2.0	414	3.5	4050	6.6
	31.9	6.3	416	4.0	4020	5.8
	31.4	4.7	414	3.5	3970	4.5
n	6		6		6	
Within-Batch Mean	30.3		414		4010	
SD	1.09		3.39		46.8	
RSD (%)	3.6		0.8		1.2	
Accuracy (%)	101.0		103.5		105.5	
002	31.7	5.7	390	-2.5	3940	3.7
	30.4	1.3	415	3.8	4030	6.1
	32.3	7.7	410	2.5	3980	4.7
	30.7	2.3	430	7.5	4050	6.6
	30.0	0.0	405	1.3	4130	8.7
	28.1	-6.3	390	-2.5	4050	6.6
n	6		6		6	
Within-Batch Mean	30.5		407		4030	
SD	1.47		15.4		65.4	
RSD (%)	4.8		3.8		1.6	
Accuracy (%)	101.7		101.8		106.1	
004	29.1	-3.0	423	5.8	3960	4.2
	30.2	0.7	425	6.3	3910	2.9
	30.4	1.3	417	4.3	4080	7.4
	30.7	2.3	416	4.0	4010	5.5
	30.3	1.0	417	4.3	4080	7.4
	31.4	4.7	416	4.0	4120	8.4
n	6		6		6	
Within-Batch Mean	30.4		419		4030	
SD	0.750		3.95		80.9	
RSD (%)	2.5		0.9		2.0	
Accuracy (%)	101.3		104.8		106.1	
n	18		18		18	
Overall Mean	30.4		413		4020	
SD	1.07		10.2		63	
RSD (%)	3.5		2.5		1.6	
Accuracy (%)	101.3		103.3		105.8	

APPLICANT'S CONCLUSION:

A quantitative procedure for the determination of ciprofloxacin in human plasma, over the concentration range of 10 to 5000 ng/mL, has been successfully validated for use at (b) (4)

REVIEWER ASSESSMENT:

The Reviewer agrees that the criteria for a successful assay validation were met.

Study Title: Determination of Ciprofloxacin in Human Plasma Samples from HGS1021-C1064 by HPLC with MS/MS Detection

Date of Report: June 11, 2010

OBJECTIVE:
To determine the concentration of ciprofloxacin in human plasma samples by HPLC with MS/MS detection, for Human Genome Sciences, Inc. Study Reference HGS-1021-C1064 entitled: "An Open-Label Study to Evaluate the Pharmacokinetics and Safety of Raxibacumab (Human Monoclonal Antibody to *B. Anthracis* Protective Antigen) Administered in Combination with Ciprofloxacin in Healthy Subjects."

STUDY SYNOPSIS:
Clinical Study HGS-1021-C1064 was an open-label study to evaluate the effect of raxibacumab on ciprofloxacin PK as well as the safety and PK of raxibacumab in combination with ciprofloxacin in healthy adult male and female subjects. Three treatment groups were evaluated. In Group 1, subjects received 15 oral (PO) 500 mg ciprofloxacin doses, given 12 hours apart (q12h). Just prior to the 11^{th} ciprofloxacin dose on Day 5, the subjects were administered a single 40 mg/kg intravenous (IV) infusion raxibacumab dose. For Group 2, subjects were administered a single 40 mg/kg IV infusion of raxibacumab only on Day 0. In Group 3, subjects received a 400 mg IV ciprofloxacin dose, immediately followed by a 40 mg/kg IV infusion raxibacumab dose; a second 400 mg IV ciprofloxacin dose was administered 12 hours after the 2^{nd} IV ciprofloxacin dose, for a total of 13 doses. Subjects were randomized in a 1:1 ratio to Group 1 or Group 2. Enrollment in Groups 1 and 2 (at least 28 evaluable subjects per group) was completed, and then an additional 28 evaluable subjects were enrolled in to Group 3. A minimum of 3 evaluable female subjects were to be enrolled into each group.

Serum ciprofloxacin concentrations and ciprofloxacin PK were reported previously in the clinical pharmacokinetics report for HGS1021-C1064.PK. Based on regulatory feedback, it was decided to perform modified assays of all specimens from this study. This report documents the modified assay results for the measurement of ciprofloxacin concentrations in serum.

BIOANALYTICAL METHODS:
Refer to methods summarized in the review for Study 2100-577.

RESULTS:
A summary of the analysis batches for this study is presented in Table 1. Samples were analyzed in a total of 29 analytical batches (there were no failed batches, run 1 was an analyst qualification batch, and runs 12 and 13 were aborted due to interference peaks). Results were calculated using peak area ratios of analyte to internal standard. Calibration curves for ciprofloxacin in human plasma ranged from 10.0 to 5000 ng/mL and were generated using a weighted ($1/x^2$) linear least-squares regression. The calibration curve parameters are presented in Table 2. Back-calculated concentrations for all calibration

curve points are presented in Table 3. The results for the calibration standards were evaluated and it was concluded that the method performed acceptably for this study.

Table 1: Study Summary Table

Run ID	Assay Date	Analyte Name	Regression Status	Regression Type	Comment
1	12-Mar-2010	Ciprofloxacin	Accepted	Linear	OK[1]
2	15-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
3	17-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
4	17-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
5	17-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
6	17-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
7	18-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
8	18-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
9	18-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
10	18-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
11	19-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
12	NA	Ciprofloxacin	Accepted	Linear	Aborted due to interference peak
13	NA	Ciprofloxacin	Accepted	Linear	Aborted due to interference peak
14	19-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
15	19-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
16	20-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
17	20-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
18	20-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
19	20-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
20	21-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
21	21-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
22	21-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
23	21-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
24	22-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
25	22-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
26	22-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
27	22-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
28	23-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
29	25-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
30	25-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
31	25-Mar-2010	Ciprofloxacin	Accepted	Linear	OK
32	28-Mar-2010	Ciprofloxacin	Accepted	Linear	OK

[1]Batch was an analyst qualification

Table 2: Calibration Curve Parameters for Ciprofloxacin in Human Plasma

Run Date	Curve Number	Slope	Intercept	R-Squared	Regression Footnote(s)
15-Mar-2010	2	0.005702	-0.001568	0.9998	1
17-Mar-2010	3	0.005709	0.002554	0.9990	1
17-Mar-2010	4	0.005624	0.010606	0.9986	1
17-Mar-2010	5	0.005596	0.004799	0.9994	1
17-Mar-2010	6	0.005866	0.002937	0.9983	1
18-Mar-2010	7	0.005752	0.011733	0.9983	1
18-Mar-2010	8	0.005757	0.007874	0.9976	1
18-Mar-2010	9	0.005723	-0.005117	0.9990	1
18-Mar-2010	10	0.005719	0.003395	0.9981	1
19-Mar-2010	11	0.005707	0.001304	0.9984	1
19-Mar-2010	14	0.005470	0.003427	0.9973	1
19-Mar-2010	15	0.005413	0.000453	0.9993	1
20-Mar-2010	16	0.005518	0.004273	0.9997	1
20-Mar-2010	17	0.005495	-0.004740	0.9994	1
20-Mar-2010	18	0.005746	-0.000203	0.9995	1
20-Mar-2010	19	0.005614	-0.006186	0.9984	1
21-Mar-2010	20	0.005540	0.001627	0.9988	1
21-Mar-2010	21	0.005813	-0.002077	0.9949	1
21-Mar-2010	22	0.005521	-0.003790	0.9996	1
21-Mar-2010	23	0.005736	0.006898	0.9993	1
22-Mar-2010	24	0.005851	0.002656	0.9985	1
22-Mar-2010	25	0.005872	-0.007375	0.9991	1
22-Mar-2010	26	0.005763	0.004062	0.9968	1
22-Mar-2010	27	0.005736	-0.005569	0.9978	1
23-Mar-2010	28	0.005690	0.001829	0.9998	1
25-Mar-2010	29	0.005737	-0.000696	0.9980	1
25-Mar-2010	30	0.005569	0.002712	0.9979	1
25-Mar-2010	31	0.005635	-0.001819	0.9963	1
28-Mar-2010	32	0.005749	-0.000564	0.9981	1

Regression Footnote(s):
1) Resp. = Slope * Conc. + Intercept

Table 3: Calibration Curve Data for Ciprofloxacin in Human Plasma

Assay Date	Analytical Run Number	10.0 (ng/mL)	20.0 (ng/mL)	50.0 (ng/mL)	200 (ng/mL)	1000 (ng/mL)	2500 (ng/mL)	4000 (ng/mL)	5000 (ng/mL)
15-Mar-2010	2	9.93	20.2	50.6	200	1010	2510	3990	4860
17-Mar-2010	3	9.96	20.1	49.9	205	1030	2460	4050	4720
17-Mar-2010	4	10.2	19.0	50.0	209	991	2540	4080	4770
17-Mar-2010	5	9.97	20.2	49.1	204	1010	2540	4020	4770
17-Mar-2010	6	9.87	20.1	52.1	210	1010	2440	3960	4670
18-Mar-2010	7	9.94	20.6	46.7	210	1020	2450	4050	4860
18-Mar-2010	8	10.4	18.2	50.3	197	1030	2520	4170	4860
18-Mar-2010	9	9.79	20.9	49.3	202	1000	2520	4060	4760
18-Mar-2010	10	9.73	21.2	48.8	211	981	2490	4030	4760
19-Mar-2010	11	10.2	19.8	46.7	202	1060	2510	4050	4880
19-Mar-2010	14	9.59	21.8	48.7	204	997	2510	4060	4700
19-Mar-2010	15	10.0	20.2	49.0	199	1010	2610	4000	4820
20-Mar-2010	16	9.97	20.0	50.7	202	1010	2540	3930	4850
20-Mar-2010	17	9.97	20.2	49.4	202	989	2400	4060	5150
20-Mar-2010	18	9.84	20.5	50.9	202	982	2490	4060	4860
20-Mar-2010	19	10.3	18.6	50.6	209	1010	2490	3900	4990
21-Mar-2010	20	10.1	19.5	49.1	200	1060	2500	*6290	4830
21-Mar-2010	21	9.25	22.8	51.5	197	985	2490	3910	4810
21-Mar-2010	22	9.83	20.8	49.7	196	1010	2500	3990	4980
21-Mar-2010	23	10.0	20.2	47.7	201	1020	2540	4050	4870
22-Mar-2010	24	9.68	21.4	49.2	205	968	2460	4070	4930
22-Mar-2010	25	9.82	20.7	50.2	199	995	2530	4100	4750
22-Mar-2010	26	10.4	18.2	51.3	201	1040	2480	4210	4690
22-Mar-2010	27	9.81	20.4	52.1	201	950	2540	4210	4660
23-Mar-2010	28	10.0	20.0	*82.9	199	1030	2510	3970	4920
25-Mar-2010	29	10.2	19.1	**No Value	216	1010	2450	3920	4880
25-Mar-2010	30	10.4	18.2	49.8	206	1020	2500	4070	4900
25-Mar-2010	31	10.5	17.7	50.3	207	1020	2530	4100	4800
28-Mar-2010	32	10.4	18.3	50.4	207	1000	2520	3900	5090
Mean		10.0	20.0	49.8	204	1010	2500	4030	4840
S.D.		0.281	1.19	1.36	4.82	24.2	40.5	83.3	116
RSD (%)		2.8	6.0	2.7	2.4	2.4	1.6	2.1	2.4
%Bias		0.0	0.0	-0.4	2.0	1.0	0.0	0.8	-3.2
n		29	29	27	29	29	29	28	29

* Failed Acceptance Criteria
** No value calculated due to lack of peak in the injection

QC sample results are presented in Table 4. The results for the QC samples were evaluated and it was concluded that the method performed acceptably for this study. No interfering peaks were found in the areas of interest that were determined to significantly impact the data. Batch runs 12 and 13 were aborted due to interference peaks. The interference peaks in runs 12 and 13 were unique to those runs and do not represent the selectivity observed throughout the remainder of the runs. Data for samples that were reassayed are presented in Table 5. The reasons for reassay and the reported results are included in the table.

Ninety-one samples were chosen and reanalyzed to confirm original ciprofloxacin results and test the repeatability of the method. The original and the reassay values of these samples are shown in Table 6. Results of the incurred sample reproducibility analysis met acceptance criteria, and 79% of samples were confirmed within 20.0% of each other.

Table 4: Quality Control Sample Data for Ciprofloxacin in Human Plasma

Run Date	Curve Number	30.0 ng/mL Dilution=1	400 ng/mL Dilution=1	3800 ng/mL Dilution=1
15-Mar-2010	2	28.7	424	3930
		30.6	411	3930
17-Mar-2010	3	31.4	401	3890
		30.8	396	3850
17-Mar-2010	4	29.9	412	3900
		29.6	413	3960
17-Mar-2010	5	30.3	407	3900
		31.0	416	3960
17-Mar-2010	6	31.3	409	3770
		30.8	386	3800
18-Mar-2010	7	27.6	401	4030
		28.0	411	3880
18-Mar-2010	8	30.9	417	3950
		27.0	406	3940
18-Mar-2010	9	29.9	422	3900
		33.6	415	4030
18-Mar-2010	10	28.9	393	4080
		33.7	401	3900
19-Mar-2010	11	30.2	400	3850
		29.0	414	3950
19-Mar-2010	14	31.1	413	4020
		30.2	424	4220
19-Mar-2010	15	33.9	416	4000
		31.3	406	3950
20-Mar-2010	16	29.2	411	3940
		29.0	419	3780
20-Mar-2010	17	34.5	424	3910
		31.4	408	3860
20-Mar-2010	18	31.6	396	3710
		30.6	390	3790
20-Mar-2010	19	31.4	392	3900
		27.6	395	3870
21-Mar-2010	20	32.8	443	4130
		29.8	419	3960
21-Mar-2010	21	29.1	402	3720
		29.9	402	3650
21-Mar-2010	22	31.4	415	3910
		31.7	418	3820
21-Mar-2010	23	27.6	396	3870
		27.5	386	3840
22-Mar-2010	24	29.4	397	3780
		30.4	400	4010
22-Mar-2010	25	34.0	402	3800
		31.4	404	3680
22-Mar-2010	26	30.1	403	3790
		30.1	398	3890
22-Mar-2010	27	31.1	423	3780
		29.6	386	3850
23-Mar-2010	28	31.7	395	3780
		31.2	399	3760
25-Mar-2010	29	30.5	413	3950
		29.4	408	3850

Run Date	Curve Number	30.0 ng/mL Dilution=1	400 ng/mL Dilution=1	3800 ng/mL Dilution=1
25-Mar-2010	30	29.5	409	3930
		29.2	398	3980
25-Mar-2010	31	30.4	407	3890
		30.3	398	3820
28-Mar-2010	32	29.9	416	3900
		30.0	398	3990
Mean		30.4	407	3890
S.D.		1.63	11.3	106
RSD (%)		5.4	2.8	2.7
Accuracy (%)		101.3	101.8	102.4
%Bias		1.3	1.8	2.4
n		58	58	58
Overall RSD (%)		3.6		

Table 5: Analytical Reassay Summary for Ciprofloxacin in Human Plasma

Subject	Treatment	Period	Time	Custom ID	Original Conc. ng/mL	Original Curve Number	Reason for Reassay	Reassay Conc. ng/mL	Reassay Curve Number	Reported Conc. ng/mL	Reason for Reported Conc.
US001-000001	1	1	Day 0 0h 0m	IB-1533731-1	48.6	2	1	41.4, 46.8	28, 28	46.8	1
US001-000001	1	1	Day 3 1h 0m	IB-1533739-1	394	2	2	383, 388	28, 28	388	1
US003-000015	1	1	Day 2 1h 0m	IB-1534382-1	1720	11	5, 3	931, 1690, 1670	30, 32, 32	1720	2
US003-000015	1	1	Day 3 1h 0m	IB-1534386-1	1170	11	4	1110, 1270	28, 28	1170	1
US003-000023	1	1	Day 4 6h 0m	IB-1534540-1	935	28	2	721, 702	29, 29	721	1
US003-000033	1	1	Day 5 1h 0m	IB-1534804-1	2890	16	2	2860, 2860	29, 29	2860	1
US003-000035	1	1	Day 6 1h 0m	IB-1534858-1	3020	17	2	2830, 2750	29, 29	2830	1
US003-000043	1	1	Day 0 0.5h 0m	IB-1535031-1	3370	19	2	2560, 2410	29, 29	2560	1
US003-000047	1	1	Day 0 4h 0m	IB-1535139-1	1210	21	2	884, 1240	29, 29	1210	1
US003-000050	1	1	Day 6 1h 0m	IB-1535226-1	874	22	2	612, 587	29, 29	612	1

Reasons For Reassay:
1). Positive Predose
2). Low Internal Standard
3). Reassay to Confirm
4). High Internal Standard
5). Incurred Sample Repeat

Reasons For Reported Conc:
1). Median of All Results, Reassayed Results Meet Acceptable Criteria
2). Original Concentration is Within Acceptance Criteria

Table 6: Incurred Sample Reproducibility Data for Ciprofloxacin in Human Plasma

Subject	Treatment	Period	Time	Custom ID	Original Conc. ng/mL	Original Curve Number	Reassay Conc. ng/mL	Reassay Curve Number	Relative % Difference
US001-000004	1	1	Day 3 0h 0m	IB-1533774-1	182	2	184	30	-1.1
US001-000004	1	1	Day 4 1h 0m	IB-1533778-1	1820	2	1250	30	37.1
US001-000005	1	1	Day 2 0h 0m	IB-1533804-1	169	3	170	30	-0.6
US001-000005	1	1	Day 7 1h 0m	IB-1533837-1	2300	3	1820	30	23.3
US001-000007	1	1	Day 2 0h 0m	IB-1533842-1	129	3	107	30	18.6
US001-000007	1	1	Day 6 1h 0m	IB-1533871-1	2180	3	1350	30	47.0
US001-000009	1	1	Day 3 0h 0m	IB-1533882-1	202	4	243	30	-18.4
US001-000009	1	1	Day 5 1h 0m	IB-1533896-1	2740	4	2760	30	-0.7
US001-000012	1	1	Day 3 0h 0m	IB-1533918-1	133	4	125	30	6.2
US001-000012	1	1	Day 6 1h 0m	IB-1533943-1	1770	4	1990	30	-11.7
US001-000015	1	1	Day 3 1h 0m	IB-1533955-1	165	5	134	30	20.7
US001-000015	1	1	Day 4 2h 0m	IB-1533960-1	1500	5	1350	30	10.5
US001-000016	1	1	Day 3 0h 0m	IB-1533990-1	89.1	5	64.0	30	32.8
US001-000016	1	1	Day 5 1.5h 0m	IB-1534005-1	1190	5	779	30	41.7
US001-000017	1	1	Day 3 0h 0m	IB-1534026-1	174	6	146	30	17.5
US001-000017	1	1	Day 6 1h 0m	IB-1534051-1	2080	6	1660	30	22.5
US001-000020	1	1	Day 2 0h 0m	IB-1534058-1	179	6	142	30	23.1
US001-000020	1	1	Day 6 1h 0m	IB-1534087-1	2270	6	1960	30	14.7
US002-000002	1	1	Day 4 1h 0m	IB-1534138-1	1790	7	2340	30	-26.6
US002-000002	1	1	Day 4 1h 0m	IB-1534145-1	171	7	230	30	-29.4
US002-000007	1	1	Day 5 1h 0m	IB-1534227-1	3070	8	3060	30	0.3
US002-000007	1	1	Day 7 12h 0m	IB-1534234-1	206	8	172	30	18.0
US002-000009	1	1	Day 4 12h 0m	IB-1534252-1	212	9	192	30	9.9
US002-000009	1	1	Day 6 1h 0m	IB-1534267-1	2200	9	1730	30	23.9
US003-000006	1	1	Day 2 1h 0m	IB-1534275-1	3100	9	3490	30	-11.8
US003-000006	1	1	Day 4 12h 0m	IB-1534288-1	202	9	134	30	40.5
US003-000007	1	1	Day 4 12h 0m	IB-1534324-1	197	10	152	30	25.8
US003-000007	1	1	Day 6 1h 0m	IB-1534337-1	1400	10	1440	30	-2.8
US003-000015	1	1	Day 5 12h 0m	IB-1534407-1	228	11	190	30	18.2
US003-000017	1	1	Day 2 1h 0m	IB-1534421-1	1540	11	1320	30	15.4
US003-000017	1	1	Day 5 1h 0m	IB-1534438-1	89.9	11	71.6	30	22.7
US003-000021	1	1	Day 5 2h 0m	IB-1534512-1	1360	27	1200	30	12.5
US003-000021	1	1	Day 6 0h 0m	IB-1534520-1	226	27	246	30	-8.5
US003-000023	1	1	Day 2 0h 0m	IB-1534528-1	165	28	157	30	5.0
US003-000023	1	1	Day 5 1h 0m	IB-1534546-1	1950	28	2230	30	-13.4
US003-000027	1	1	Day 6 0h 0m	IB-1534627-1	169	14	173	30	-2.3
US003-000027	1	1	Day 7 1h 0m	IB-1534630-1	2390	14	2130	30	11.5
US003-000029	1	1	Day 3 0h 0m	IB-1534675-1	192	15	126	30	41.5
US003-000029	1	1	Day 4 1h 0m	IB-1534686-1	2130	15	2000	30	6.3
US003-000030	1	1	Day 3 1h 0m	IB-1534712-1	199	15	222	30	-10.9
US003-000030	1	1	Day 4 2h 0m	IB-1534717-1	947	15	766	30	21.1
US003-000032	1	1	Day 0 0.5h 0m	IB-1534767-1	2600	16	2560	30	1.6
US003-000032	1	1	Day 6 1h 0m	IB-1534782-1	195	16	156	30	22.2
US003-000033	1	1	Day 0 12h 0m	IB-1534800-1	209	16	131	30	45.9
US003-000033	1	1	Day 6 1h 0m	IB-1534808-1	3010	16	2800	30	7.2
US003-000034	1	1	Day 0 0.5h 0m	IB-1534819-1	1760	17	1750	30	0.6
US003-000034	1	1	Day 6 1h 0m	IB-1534834-1	131	17	186	30	-34.7
US003-000035	1	1	Day 0 12h 0m	IB-1534852-1	178	17	171	30	4.0
US003-000035	1	1	Day 5 1h 0m	IB-1534854-1	3010	17	3010	30	0.0
US003-000037	1	1	Day 0 12h 0m	IB-1534883-1	184	17	183	30	0.5
US003-000037	1	1	Day 5 1h 0m	IB-1534887-1	1820	17	1770	30	2.8

Subject	Treatment	Period	Time	Custom ID	Original Conc. ng/mL	Original Curve Number	Reassay Conc. ng/mL	Reassay Curve Number	Relative % Difference
US003-000038	1	1	Day 6 1h 0m	IB-1534917-1	2190	18	2160	31	1.4
US003-000038	1	1	Day 7 12h 0m	IB-1534926-1	110	18	116	31	-5.3
US003-000039	1	1	Day 6 1h 0m	IB-1534942-1	2080	18	2280	31	-9.2
US003-000039	1	1	Day 7 12h 0m	IB-1534951-1	134	18	137	31	-2.2
US003-000040	1	1	Day 0 0.5h 0m	IB-1534953-1	3270	18	3370	31	-3.0
US003-000040	1	1	Day 7 12h 0m	IB-1534977-1	184	18	185	31	-0.5
US003-000041	1	1	Day 0 0.5h 0m	IB-1534979-1	3140	19	3290	31	-4.7
US003-000041	1	1	Day 7 12h 0m	IB-1535003-1	190	19	202	31	-6.1
US003-000042	1	1	Day 0 0.5h 0m	IB-1535005-1	2550	19	2650	31	-3.8
US003-000042	1	1	Day 0 12h 0m	IB-1535012-1	217	19	232	31	-6.7
US003-000043	1	1	Day 0 12h 0m	IB-1535038-1	186	19	188	31	-1.1
US003-000043	1	1	Day 5 1h 0m	IB-1535042-1	1850	19	1800	31	2.7
US003-000044	1	1	Day 0 12h 0m	IB-1535064-1	201	20	196	31	2.5
US003-000044	1	1	Day 5 1h 0m	IB-1535068-1	3930	20	4020	31	-2.3
US003-000046	1	1	Day 0 0.5h 0m	IB-1535109-1	3300	20	3270	31	0.9
US003-000046	1	1	Day 7 12h 0m	IB-1535133-1	194	20	218	31	-11.7
US003-000047	1	1	Day 0 0.5h 0m	IB-1535135-1	3260	21	3400	31	-4.2
US003-000047	1	1	Day 0 12h 0m	IB-1535142-1	218	21	221	31	-1.4
US003-000048	1	1	Day 0 0.5h 0m	IB-1535161-1	3200	21	3400	31	-6.1
US003-000048	1	1	Day 7 12h 0m	IB-1535185-1	121	21	127	31	-4.8
US003-000050	1	1	Day 5 1h 0m	IB-1535224-1	2270	22	2300	31	-1.3
US003-000050	1	1	Day 7 12h 0m	IB-1535237-1	93.9	22	94.1	31	-0.2
US003-000051	1	1	Day 0 0.5h 0m	IB-1535239-1	2820	22	2670	31	5.5
US003-000051	1	1	Day 0 12h 0m	IB-1535246-1	147	22	163	31	-10.3
US003-000052	1	1	Day 0 0.5h 0m	IB-1535265-1	1900	22	1910	31	-0.5
US003-000052	1	1	Day 7 12h 0m	IB-1535289-1	191	22	196	31	-2.6
US003-000053	1	1	Day 0 12h 0m	IB-1535298-1	228	23	238	31	-4.3
US003-000053	1	1	Day 6 1h 0m	IB-1535306-1	3430	23	3600	31	-4.8
US003-000054	1	1	Day 0 0.5h 0m	IB-1535317-1	1840	23	1930	31	-4.8
US003-000054	1	1	Day 7 12h 0m	IB-1535341-1	192	23	190	31	1.0
US003-000055	1	1	Day 0 12h 0m	IB-1535350-1	191	23	188	31	1.6
US003-000055	1	1	Day 6 1h 0m	IB-1535358-1	1980	23	2150	31	-8.2
US003-000056	1	1	Day 0 0.5h 0m	IB-1535369-1	1910	24	1940	31	-1.6
US003-000056	1	1	Day 7 12h 0m	IB-1535393-1	185	24	206	31	-10.7
US003-000057	1	1	Day 0 0.5h 0m	IB-1535395-1	1490	24	1550	31	-3.9
US003-000057	1	1	Day 7 12h 0m	IB-1535419-1	160	24	171	31	-6.6
US003-000059	1	1	Day 0 12h 0m	IB-1535453-1	158	25	174	31	-9.6
US003-000059	1	1	Day 6 1h 0m	IB-1535461-1	2350	25	2360	31	-0.4
US003-000060	1	1	Day 2 1h 0m	IB-1535473-1	2280	25	2260	31	0.9
US003-000060	1	1	Day 4 12h 0m	IB-1535488-1	158	25	169	31	-6.7

Relative % Difference= ((Original-Reassay)/(Original + Reassay))*200

79% of the repeat results and original results are within 20.0% of each other.
Note: 91 of 91 samples had results within the curve range that were able to be calculated for this statistic.

APPLICANT'S CONCLUSION:

Sample results were accepted if calibration curve and QC data from the reported batches indicated that the method met the acceptance criteria for those batches. Reported data met the following criteria, as defined in [redacted] (b) (4) SOPs:

- Batches were considered acceptable if no more than one-fourth of the calibration standards were excluded, and a minimum of six non-zero back-calculated concentrations for calibration standards were within the range of 85.0% to 115.9% of theoretical [80% to 120.0% at the lower limit of quantitation (LLOQ)].
- Batches were considered acceptable if at least one-half of the undiluted QC samples at each concentration and two-thirds of all undiluted QC samples in the curve range were within the range of 85.0% to 115.0% of theoretical
- The incurred sample reproducibility (ISR) analysis was considered acceptable if at least two-thirds (rounded up) of the repeat results and original results were within 20.0% of each other

The calibration curve and QC sample data from the reported batches indicate that the methods met the acceptance criteria for those batches.

REVIEWER ASSESSMENT:
The Reviewer agrees with the applicant's conclusions.

Study Title: Comparison Report of Pharmacokinetic Results Generated Using Ciprofloxacin Assay Performed at [redacted] and Ciprofloxacin Reassay Performed at [redacted] for Protocol HGS1021-C1064

Date of Report: June 15, 2010

OBJECTIVES:
To compare ciprofloxacin pharmacokinetics based on assay results from [redacted] (reassay) with pharmacokinetics based on the assay results from [redacted]

STUDY DESIGN:
Study HGS1021-C1064 was an open-label study to evaluate the effect of raxibacumab on ciprofloxacin PK as well as the safety and PK of raxibacumab in combination with ciprofloxacin in healthy adult male and female subjects. Three treatment groups were evaluated. In Group 1, subjects received 15 oral (PO) 500 mg ciprofloxacin doses, given 12 hours apart (q12h). Just prior to the 11th ciprofloxacin dose on Day 5, the subjects were administered a single 40 mg/kg intravenous infusion raxibacumab dose. For Group 2, subjects were administered a single 40 mg/kg IV infusion raxibacumab dose only on Day 0. In Group 3, subjects received a 400 mg IV ciprofloxacin dose, immediately followed by a 40 mg/kg IV infusion raxibacumab dose; a second 400 mg IV ciprofloxacin dose was administered 12 hours after the first dose, and 500 mg PO ciprofloxacin doses were administered every 12 hours after the 2nd IV ciprofloxacin dose, for a total of 15 doses. Subjects were randomized in a 1:1 ratio to Group 1 or Group 2. Enrollment in Groups 1 and 2 (at least 28 evaluable subjects per group) was completed, and then an additional 28 evaluable subjects were enrolled into Group 3. A minimum of three evaluable female subjects were to be enrolled into each group. In Group 1 and Group 3, specimens for plasma ciprofloxacin concentration measurement were collected at selected time points. For Group 1, plasma ciprofloxacin concentration time profiles were obtained following the first dose (Day 0, prior to raxibacumab dosing) as well as following the 13th PO dose (Day 7, after raxibacumab dosing).

Plasma ciprofloxacin concentrations and ciprofloxacin PK were reported previously (referred to as the original results; Clinical Pharmacokinetic Report HGS1021-C1064.PK). Reassays of samples from this study were performed using a validated method at a different laboratory (referred to as the reassay results). This report documents a comparison of the original and the reassay results, PK analysis of the reassay results, and comparison of the PK based on the original assay and reassay.

Subjects Evaluated for Pharmacokinetics: A subject was considered evaluable for ciprofloxacin PK if the subject met either of the following 2 criteria: 1) received the 1st of 11 of the planned ciprofloxacin doses as well as the raxibacumab dose and had predose through at least 8 h postdose plasma ciprofloxacin concentration measured for the 9th and 11th dose (Group 1) or 2) received the first ciprofloxacin dose as well as the raxibacumab dose, and had predose through at least 8 h postdose plasma ciprofloxacin concentrations measured for the first ciprofloxacin dose (Group 3). Of the 32 subjects treated in Group

1, 30 subjects met those criteria and were considered evaluable for ciprofloxacin PK. For Group 3, all 28 subjects met those criteria and were considered evaluable for ciprofloxacin PK.

PHARMACOKINETIC/STATISTICAL ANALYSIS:

Pharmacokinetic Methods: For plasma ciprofloxacin concentration-time profiles (collected after the 9^{th} and 11^{th} doses in Group 1, as well as after the 1^{st} IV dose and the 15^{th} dose in Group 3), individual subject's profiles were analyzed using non-compartmental techniques to determine PK parameters.

Statistical Methods: Descriptive statistics were used to summarize plasma ciprofloxacin concentration-time results and PK parameters by dose number and by treatment group, for the original assay and reassay. For a nominal sample collection time of a dose of the treatment group, equivalence of plasma ciprofloxacin concentrations between the original and reassay was tested using a 2-sided 90% confidence interval (CI). By dose number and by treatment group, equivalence of each PK parameter between the original and reassay was also evaluated using a two-sided 90% CI. For all equivalence tests, the criterion is the same: the 2 assays were considered equivalent if the 90% CI fell within the 80-125% range. The exception was time of occurrence for the peak plasma ciprofloxacin concentration (t_{max} or $t_{max,ss}$) for which the Wilcoxon signed rank test was used. A significance level of $\alpha = 0.05$ was used for all statistical comparisons.

RESULTS:

No data from the evaluable subjects were excluded from the analyses. Subjects with full PK profiles for the 4 doses (Doses 9 and 11 of Group 1 and Doses 1 and 15 of Group 3) were used for PK parameter estimation by non-compartmental analysis. This is consistent with the original analyses.

One of the 60 subjects administered ciprofloxacin in this study (Subject No. US001-000001) had a measureable plasma ciprofloxacin concentration prior to administration of the 1^{st} ciprofloxacin dose at 39 ng/mL by the original assay and 47 ng/mL by the reassay. That predose concentration was less than 5.0% of the 1070 (original assay) or 1310 (reassay) ng/mL concentration at 1 hour postdose (presumed peak) measured on Day 2, and was less than 15% of the 405 (original assay) or 333 (reassay) ng/mL predose concentration measured prior to the 9^{th} ciprofloxacin dose. Therefore, this subject's postdose concentrations were not adjusted for the predose measurement.

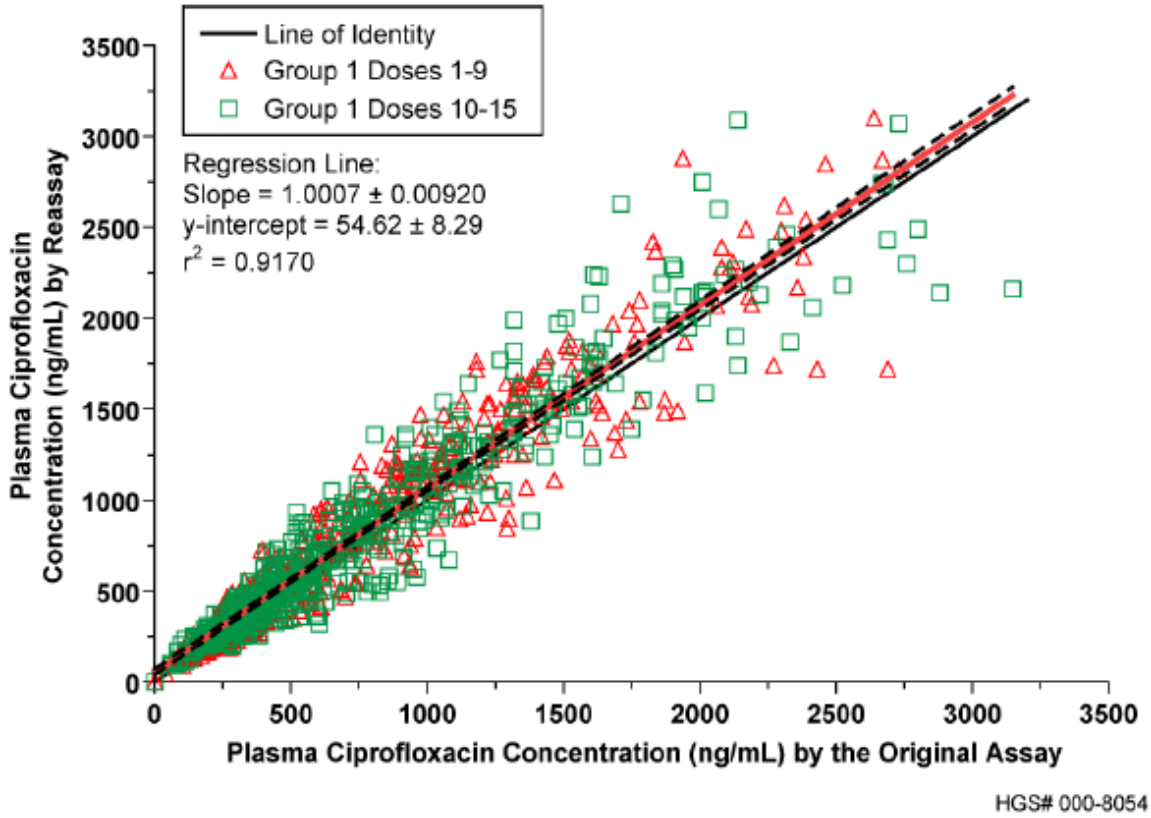
Since the terminal half-life of ciprofloxacin was about 4.5 hours, steady state was thought to be reached at Dose 5 on Day 2 for both Group 1 and Group 3. Any peak (trough) ciprofloxacin concentrations for Doses 5-15 were considered to be steady-state maximum (minimum) ciprofloxacin concentrations, $C_{max,ss}$ ($C_{min,ss}$) for the treatment groups. The time for $C_{max,ss}$ is $t_{max,ss}$.

A comparison of the plasma ciprofloxacin concentrations for the reassay vs. those from the original assays is provided in Figure 1 for Group 1 and Figure 2 for Group 3. Figure 1 shows the following linear relationship between the reassay results (C_R) and the

original assay results (C_O) of Group 1 with r^2 of 0.9170 as shown by the fitted line in red (with dashed lines representing 90% CI):

$$C_R = 1.0007 \times C_O + 54.62$$

Figure 1: Reassay vs. original assay plasma ciprofloxacin concentrations for treatment Group 1



Relative to the original assay, the reassay resulted in approximately the same mean ciprofloxacin concentrations for doses of Treatment Group 1. Figure 2 shows the following linear relationship between the reassay results (C_R) and the original assay results (C_O) of Group 3 with an r^2 of 0.9106 as shown by the fitted line in red (with the dashed lines representing 90% CI):

$$C_R = 1.204 \times C_O + 33.83$$

Relative to the original assay, the reassay resulted in about 20% higher mean concentrations for doses of Treatment Group 3.

Table 1 and Table 2 provide a summary of the comparison of the reassay and original assay results by nominal sample collection time for Group 1 and Group 3, respectively. For doses in Group 1, the mean ratios of the geometric means for the reassay:original assay concentrations results ranged from 106.61% to 116.72% with all 90% CIs within the equivalence range of 80-125%. The mean with standard deviation (SD) error bars plasma ciprofloxacin concentration-time profiles based on the original assay and reassay

results are illustrated in Figure 3 and Figure 4 for Groups 1 and 3, respectively. Figure 3 shows that profiles for the original assay results and reassay results for Group 1 are very similar with SD bars that overlapped at all nominal time points. However, Figure 4 shows that for Group 3 the profiles for the original assay are lower than those for the reassay results at all time points. Overall, the reassay was equivalent to the original assay for concentrations at all time points for Group 1, but not for the concentrations in Group 3.

Figure 2: Reassay vs. original assay plasma ciprofloxacin concentrations for treatment Group 3

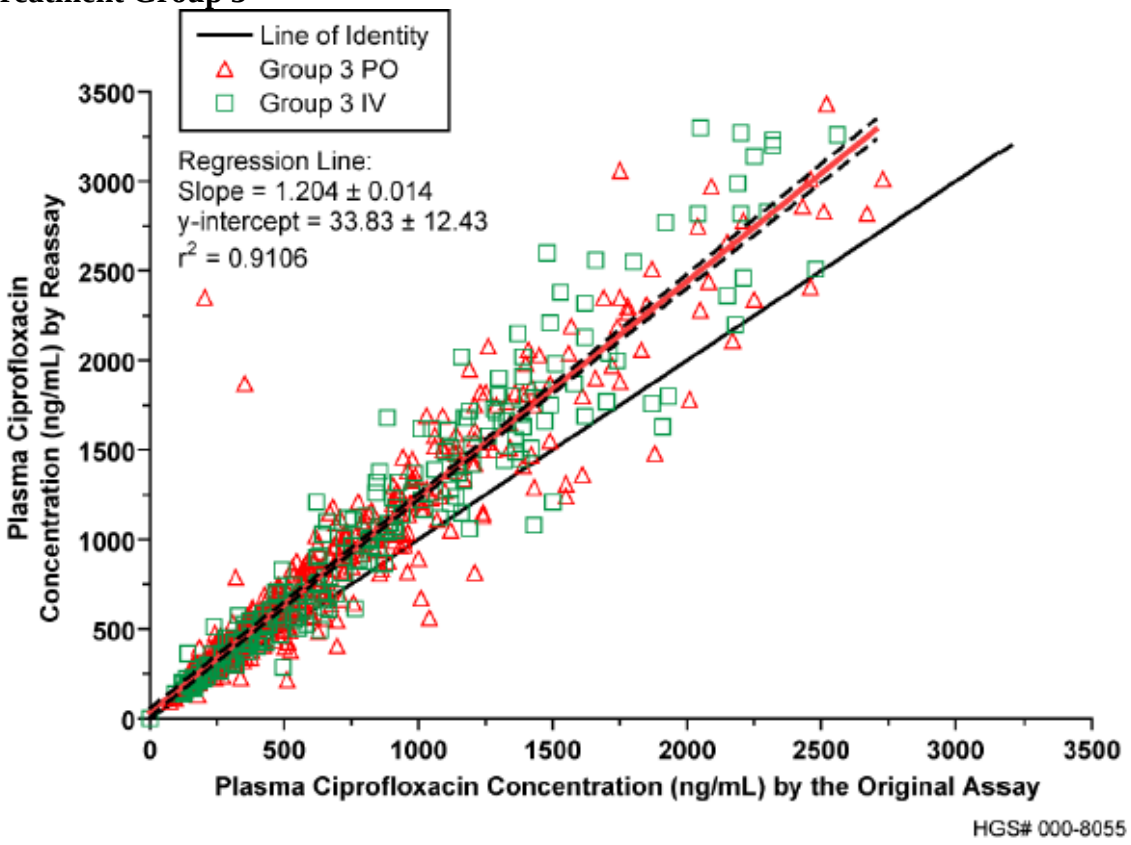


Table 1: Summary of original and reassay plasma ciprofloxacin concentrations (mean ± SD) by collection time (Group 1)

Nominal Sample Collection Time	Original Assay (O) (ng/mL)	Reassay (R) (ng/mL)	R/O Geometric Mean Ratio% (90% CI)[1]
Dose 5, Predose (n = 30)	367 ± 138	400 ± 175	106.61 (99.77, 113.93)
Dose 5, 1 h Postdose (n = 31)[2]	1063 ± 485	1188 ± 550	111.28 (106.49, 116.28)
Dose 6, Predose (n = 31)[2]	239 ± 91	266 ± 99	112.44 (105.89, 119.40)
Dose 6, 1 h Postdose (n = 31)[2]	1054 ± 728	1165 ± 826	108.79 (102.47, 115.49)
Dose 7, Predose (n = 31)[2]	416 ± 148	453 ± 173	107.88 (102.70, 113.31)
Dose 7, 1 h Postdose (n = 30)	923 ± 421	1015 ± 446	109.91 (103.98, 116.17)
Dose 8, Predose (n = 30)	228 ± 85	247 ± 100	107.26 (100.42, 114.56)
Dose 8, 1 h Postdose (n = 30)	1161 ± 791	1176 ± 723	106.68 (99.40, 114.49)
Dose 9, Predose (n = 30)	384 ± 145	417 ± 174	106.62 (99.34, 114.43)
Dose 9, 0.5 h Postdose (n = 30)	733 ± 540	781 ± 550	108.80 (102.38, 115.63)
Dose 9, 1 h Postdose (n = 30)	1068 ± 597	1179 ± 615	114.03 (107.07, 121.44)
Dose 9, 1.5 h Postdose (n = 30)	1159 ± 489	1277 ± 495	112.83 (106.21, 119.85)
Dose 9, 2 h Postdose (n = 30)	1182 ± 427	1328 ± 433	114.43 (108.64, 120.54)
Dose 9, 4 h Postdose (n = 30)	890 ± 401	971 ± 428	109.83 (103.61, 116.42)
Dose 9, 6 h Postdose (n = 30)	600 ± 235	659 ± 235	111.86 (104.93, 119.26)
Dose 9, 8 h Postdose (n = 30)	446 ± 192	472 ± 158	109.57 (101.41, 118.39)
Dose 9, 12 h Postdose (n = 30)	237 ± 112	265 ± 96	116.20 (108.96, 123.93)

Nominal Sample Collection Time	Original Assay (O) (ng/mL)	Reassay (R) (ng/mL)	R/O Geometric Mean Ratio% (90% CI)[1]
Dose 10, 1 h Postdose (n = 30)	725 ± 626	728 ± 583	107.34 (100.62, 114.52)
Dose 11, Predose (n = 30)	527 ± 172	563 ± 185	107.38 (99.90, 115.42)
Dose 11, 0.5 h Postdose (n = 29)	679 ± 592	705 ± 490	108.23 (102.50, 114.28)
Dose 11, 1 h Postdose (n = 30)	1013 ± 608	1090 ± 601	110.21 (104.22, 116.55)
Dose 11, 1.5 h Postdose (n = 30)	1106 ± 558	1229 ± 597	112.04 (104.19, 120.48)
Dose 11, 2 h Postdose (n = 30)	1234 ± 594	1307 ± 540	109.14 (103.37, 115.24)
Dose 11, 4 h Postdose (n = 30)	968 ± 308	1028 ± 298	107.15 (101.49, 113.13)
Dose 11, 6 h Postdose (n = 30)	653 ± 227	731 ± 199	114.17 (107.93, 120.76)
Dose 11, 8 h Postdose (n = 30)	498 ± 178	530 ± 157	108.86 (101.14, 117.17)
Dose 11, 12 h Postdose (n = 29)	275 ± 106	290 ± 72	110.28 (100.56, 120.94)
Dose 12, 1 h Postdose (n = 30)	973 ± 647	1038 ± 689	108.91 (101.63, 116.70)
Dose 13, Predose (n = 30)	437 ± 194	471 ± 201	109.05 (101.83, 116.78)
Dose 13, 1 h Postdose (n = 30)	1078 ± 604	1201 ± 753	108.82 (101.10, 117.12)
Dose 14, Predose (n = 30)	262 ± 90	303 ± 93	116.72 (110.05, 123.79)
Dose 14, 1 h Postdose (n = 30)	1107 ± 822	1246 ± 865	114.80 (109.13, 120.76)
Dose 15, Predose (n = 30)	378 ± 159	399 ± 149	108.44 (100.74, 116.72)
Dose 15, 1 h Postdose (n = 30)	1118 ± 661	1200 ± 638	109.67 (101.97, 117.96)
Dose 15, 12 h Postdose (n = 30)	273 ± 124	293 ± 112	110.02 (102.84, 117.70)

[1] Geometric mean ratio% and 90% confidence interval of the reassay values relative to the original assay value. Samples with no measurable concentration reported for either assay were excluded from the equivalence tests.

[2] Include results for US003-000036 who is not PK evaluable.

Table 2: Summary of the original and reassay plasma ciprofloxacin concentrations (mean ± SD) by collection (Group 3)

Nominal Sample Collection Time	Original Assay [O] (ng/mL)	Reassay [R] (ng/mL)	R/O Geometric Mean Ratio% (90% CI)[1]
Dose 1, 0.5 h Postdose (n = 28)	1854 ± 402	2446 ± 596	130.98 (124.63, 137.66)
Dose 1, 1 h Postdose (n = 28)	1341 ± 370	1665 ± 406	125.04 (119.30, 131.05)
Dose 1, 1.5 h Postdose (n = 27)	1081 ± 295	1362 ± 309	127.04 (119.68, 134.85)
Dose 1, 2 h Postdose (n = 28)	989 ± 237	1204 ± 279	121.66 (115.82, 127.79)
Dose 1, 4 h Postdose (n = 28)	665 ± 171	808 ± 192	121.67 (113.78, 130.09)
Dose 1, 6 h Postdose (n = 28)	450 ± 110	542 ± 93	122.20 (114.95, 129.91)
Dose 1, 8 h Postdose (n = 28)	330 ± 104	380 ± 81	117.99 (108.58, 128.21)
Dose 1, 12 h Postdose (n = 28)	169 ± 43	210 ± 47	124.91 (119.72, 130.33)
Dose 11, Predose (n = 28)	371 ± 128	462 ± 167	124.89 (117.10, 133.20)
Dose 11, 1 h Postdose (n = 28)	922 ± 563	1168 ± 670	129.11 (121.12, 137.63)
Dose 12, Predose (n = 28)	315 ± 113	397 ± 117	127.77 (119.89, 136.17)
Dose 12, 1 h Postdose (n = 28)	1222 ± 666	1579 ± 894	128.64 (122.44, 135.15)
Dose 13, Predose (n = 28)	412 ± 156	518 ± 180	127.17 (120.57, 134.12)
Dose 13, 1 h Postdose (n = 28)	776 ± 556	943 ± 640	125.95 (118.56, 133.80)
Dose 14, Predose (n = 28)	230 ± 89	293 ± 89	130.26 (122.03, 139.05)
Dose 14, 1 h Postdose (n = 28)	1161 ± 715	1616 ± 940	140.17 (121.30, 161.98)
Dose 15, Predose (n = 28)	381 ± 124	473 ± 126	126.36 (120.34, 132.69)
Dose 15, 0.5 h Postdose (n = 28)	744 ± 392	914 ± 426	125.13 (119.22, 131.34)
Dose 15, 1 h Postdose (n = 28)	882 ± 370	1172 ± 442	134.59 (121.89, 148.62)
Dose 15, 1.5 h Postdose (n = 28)	936 ± 369	1142 ± 446	122.48 (116.12, 129.19)
Dose 15, 2 h Postdose (n = 28)	948 ± 534	1130 ± 551	121.52 (113.98, 129.57)
Dose 15, 4 h Postdose (n = 28)	777 ± 517	906 ± 498	121.27 (114.70, 128.22)
Dose 15, 6 h Postdose (n = 28)	528 ± 333	601 ± 281	120.42 (111.13, 130.49)

Nominal Sample Collection Time	Original Assay [O] (ng/mL)	Reassay [R] (ng/mL)	R/O Geometric Mean Ratio% (90% CI)[1]
Dose 15, 8 h Postdose (n = 28)	382 ± 228	452 ± 210	123.80 (115.82, 132.32)
Dose 15, 12 h Postdose (n = 28)	193 ± 99	236 ± 94	126.65 (117.08, 137.01)

[1] Geometric mean ratio% and 90% confidence interval of the reassay values relative to the original assay value. Samples with no measurable concentration reported for either assay were excluded from the equivalence tests.

Figure 3: Mean (±SD error bars) plasma ciprofloxacin concentrations of Dose 9 and Dose 11 of treatment Group 1

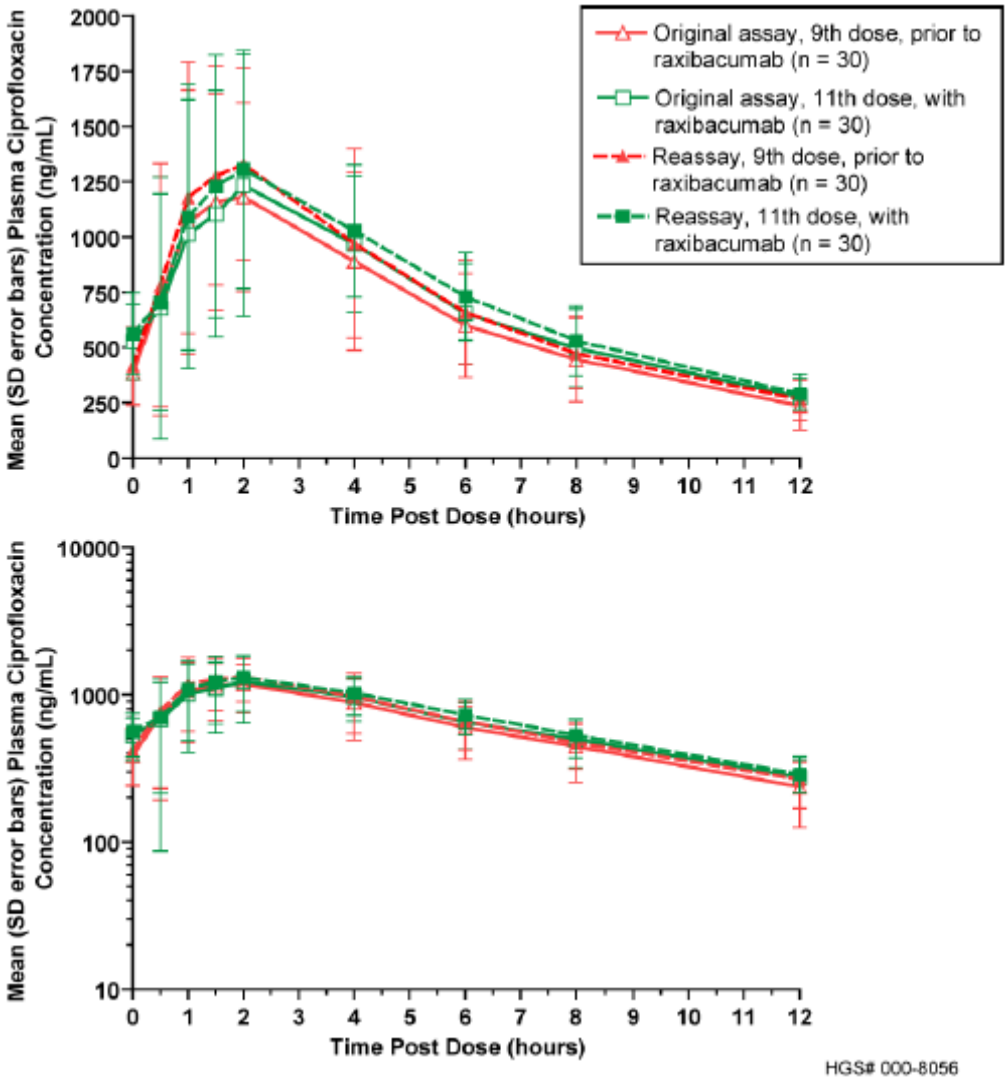
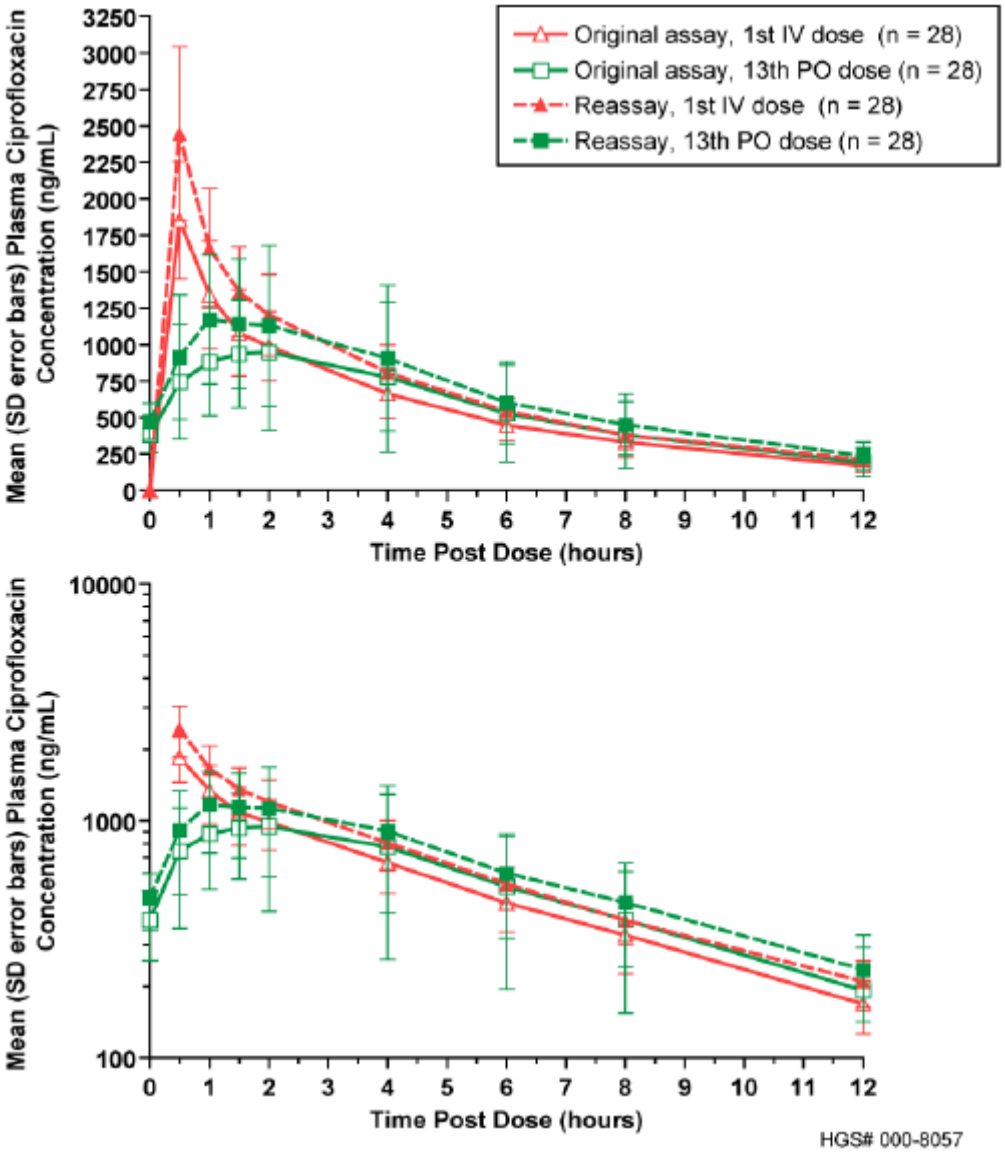


Figure 4: Mean (±SD error bars) plasma ciprofloxacin concentrations of Dose 1 and Dose 15 of treatment Group 3



Key mean (±SD) non-compartmental PK results are summarized in Table 3 and Table 4 or Group 1 and 3, respectively. Equivalence tests or Wilcoxon signed rank tests of these PK parameters for the reassay results versus those for the original assay results are also summarized in those tables.

For Group 1, equivalence between the original assay and reassay was shown for all PK parameters for both Doses 9 and 11 since the 90% CI fell within the 80 to 125% range, and there was no statistically significant difference in $t_{max,ss}$ between the 2 assays for Dose 9 (p=0.625) and Dose 11 (p=0.791). For Dose 1 of Group 3, t_{max} estimates were the same for each subject by the 2 assays; equivalence between the 2 assays was shown for $t_{1/2z}$ and MRT but not for C_{max}, $AUC_{0\text{-}inf}$, CL, V_{ss}, V_z with 90% CI falling slightly outside of the 80-125% range for those parameters. For Dose 15 of Group 3, no statistically significant

difference was found for $t_{max,ss}$ (p=0.204) between the 2 assays; equivalence between the 2 assays could be declared for $t_{1/2z}$, accumulation index, fluctuation and bioavailability but could not be declared for $C_{max,ss}$, $AUC_{0\text{-}tau}$, $AUC_{0\text{-}8}$, CL/F, $C_{min,ss}$, or V_z/F.

Overall, ciprofloxacin PK was equivalent for all doses of Treatment Group 1, but equivalence was not demonstrated for all parameters for Treatment Group 3 between the original assay and reassay. The results observed in Treatment Group 3 do not suggest any clinically meaningful differences shown between the 2 assays.

Table 3: Summary of ciprofloxacin PK parameters, based on original and reassay results for Group 1

	Original (O)		Reassay (R)		Equivalence Test
Parameter	**Mean ± SD**	**Geometric Mean (CV%)**	**Mean ± SD**	**Geometric Mean (CV%)**	**R/O Geometric Mean Ratio% (90% CI)**
Dose 9, PO 500 mg ciprofloxacin without raxibacumab (N = 30)					
$C_{max,ss}$ (ng/mL)	1436 ± 519	1349 (38%)	1535 ± 512	1459 (33%)	108.19 (102.34, 114.38)
$t_{max,ss}$ (h)	1.75 (0.50 to 4.00)[1]		1.50 (0.50 to 4.00)[1], p = 0.625[2]		NA
$AUC_{0\text{-}6h}$ (ng·h/mL)	6398 ± 2239	6045 (35%)	7032 ± 2316	6717 (31%)	111.11 (106.35, 116.07)
$AUC_{0\text{-}t}$ (ng·h/mL)	7708 ± 2692	7283 (35%)	8459 ± 2720	8094 (30%)	111.14 (106.33, 116.18)
$t_{1/2,z}$ (h)	4.50 ± 1.86	4.26 (32%)	4.53 ± 0.93	4.44 (20%)	104.30 (96.97, 112.18)
CL/F (L/h)	72.5 ± 24.1	68.7 (35%)	64.3 ± 17.5	61.8 (30%)	89.97 (86.08, 94.05)
V_z/F (L)	486 ± 328	422 (54%)	426 ± 164	396 (41%)	93.84 (85.36, 103.16)
$C_{min,ss}$ (ng/mL)	225 ± 89	209 (41%)	258 ± 91	244 (35%)	116.91 (110.04, 124.21)
Accumulation Index	1.20 ± 0.19	1.19 (13%)	1.19 ± 0.09	1.19 (7%)	100.30 (97.26, 103.44)
Fluctuation (%)	191 ± 50	185 (28%)	183 ± 45	177 (27%)	95.93 (90.33, 101.88)
Dose 11, PO 500 mg ciprofloxacin immediately after 40 mg/kg raxibacumab IV infusion (N = 30)					
$C_{max,ss}$ (ng/mL)	1419 ± 599	1310 (42%)	1475 ± 538	1385 (38%)	105.68 (99.43, 112.33)
$t_{max,ss}$ (h)	2.00 (0.58 to 4.12)[1]		1.52 (1.00 to 6.00)[1], p = 0.791[2]		NA
$AUC_{0\text{-}6h}$ (ng·h/mL)	6694 ± 2270	6343 (34%)	7227 ± 2085	6949 (29%)	109.56 (104.98, 114.33)
$AUC_{0\text{-}t}$ (ng·h/mL)	8183 ± 2643	7787 (33%)	8808 ± 2380	8518 (27%)	109.38 (104.64, 114.33)
$t_{1/2,z}$ (h)	4.48 ± 1.28	4.33 (27%)	4.64 ± 1.09	4.53 (22%)	104.75 (96.28, 113.96)
CL/F (L/h)	67.5 ± 21.8	64.2 (33%)	60.6 ± 15.4	58.7 (27%)	91.42 (87.46, 95.57)
V_z/F (L)	426 ± 150	401 (37%)	412 ± 167	384 (39%)	95.77 (87.17, 105.21)

	Original (O)		Reassay (R)		Equivalence Test
Parameter	**Mean ± SD**	**Geometric Mean (CV%)**	**Mean ± SD**	**Geometric Mean (CV%)**	**R/O Geometric Mean Ratio% (90% CI)**
$C_{min,ss}$ (ng/mL) (N = 29)[3]	255 ± 88	238 (42%)	280 ± 73	270 (30%)	113.19 (105.48, 121.47)
Accumulation Index	1.19 ± 0.12	1.19 (10%)	1.20 ± 0.11	1.20 (8%)	101.11 (97.87, 104.44)
Fluctuation (%)	168 ± 37	164 (24%)	160 ± 41	154 (29%)	94.37 (88.34, 100.81)

Abbreviations: $C_{max,ss}$, maximum plasma drug concentration during a steady-state dosing interval; $t_{max,ss}$, time of occurrence for $C_{max,ss}$; $AUC_{0\text{-}6h}$, area under the plasma drug concentration-time curve from 0 to 8 h post dose; $AUC_{0\text{-}t}$, area under the plasma drug concentration-time curve during a steady-state dosing interval; $t_{1/2,z}$, elimination half-life for the terminal phase; CL/F, apparent clearance for oral dosing; V_z/F, apparent volume of distribution in the terminal phase for oral dosing; $C_{min,ss}$, minimum plasma drug concentration during a steady-state dosing interval; CI, Confidence Interval; NA, not applicable; CV% coefficient of variation percentage.

[1] Median and range are presented.

[2] Wilcoxon signed rank test.

[3] Subject US003-000061 has Cmin = 0, therefore excluded from equivalence test.

Source: Summary Table 7-3 and Summary Table 7-4; Appendix 5 and Appendix 6.

Table 4: Summary of ciprofloxacin PK parameters, based on original and reassay results for Group 3

Parameter	Original (O)		Reassay (R)		Equivalence Test
	Mean ± SD	Geometric Mean (CV%)	Mean ± SD	Geometric Mean (CV%)	R/O Geometric Mean Ratio% (90% CI)
Dose 1, IV 400 mg ciprofloxacin with raxibacumab (n = 28)					
C_{max} (ng/mL)	1854 ± 402	1811 (22%)	2446 ± 596	2372 (26%)	130.98 (124.63, 137.66)
t_{max} (h)	1.33 (1.23 to 1.60)[1]		1.33 (1.23 to 1.60)[1], p = NA [2]		NA
$AUC_{0-\infty}$ (ng·h/mL)	8622 ± 1816	8446 (21%)	10704 ± 2090	10503 (20%)	124.36 (119.43, 129.49)
$t_{1/2,z}$ (h)	4.02 ± 0.77	3.96 (18%)	4.16 ± 0.53	4.13 (13%)	104.25 (99.12, 109.64)
MRT (h)	5.75 ± 0.89	5.69 (15%)	5.75 ± 0.69	5.71 (12%)	100.37 (97.48, 103.35)
CL (L/h)	48.3 ± 10.0	47.4 (21%)	38.8 ± 7.9	38.1 (20%)	80.41 (77.22, 83.73)
V_{ss} (L)	278 ± 71	269 (26%)	224 ± 60	217 (25%)	80.71 (77.30, 84.28)
V_z (L)	281 ± 81	271 (29%)	233 ± 58	227 (24%)	83.83 (79.53, 88.36)
Dose 15, PO 500 mg ciprofloxacin (n = 28)					
$C_{max,ss}$ (ng/mL)	1195 ± 566	1087 (46%)	1421 ± 566	1321 (40%)	121.52 (116.13, 127.16)
$t_{max,ss}$ (h)	1.00 (0.5 to 6.00)[1]		1.00 (0.5 to 4.00)[1], p = 0.204[2]		NA
AUC_{0-8h} (ng·h/mL)	5494 ± 2643	4987 (46%)	6548 ± 2573	6104 (39%)	122.42 (116.97, 128.11)
AUC_{0-t} (ng·h/mL)	6599 ± 3208	5980 (46%)	7880 ± 3109	7345 (39%)	122.82 (117.09, 128.83)
F (%)	60 ± 21	57 (34%)	58 ± 16	56 (28%)	98.76 (93.70, 104.10)
$t_{1/2,z}$ (h)	4.45 ± 1.04	4.34 (24%)	4.56 ± 0.71	4.51 (16%)	104.02 (98.58, 109.77)
CL/F (L/h)	91.2 ± 36.9	83.6 (46%)	72.8 ± 26.6	68.1 (39%)	81.42 (77.62, 85.40)
V_z/F (L)	599 ± 292	523 (62%)	488 ± 208	443 (50%)	84.70 (78.70, 91.15)

Parameter	Original (O)		Reassay (R)		Equivalence Test
	Mean ± SD	Geometric Mean (CV%)	Mean ± SD	Geometric Mean (CV%)	R/O Geometric Mean Ratio% (90% CI)
$C_{min,ss}$ (ng/mL)	193 ± 102	174 (48%)	243 ± 111	222 (44%)	128.02 (121.17, 135.26)
Accumulation Index	1.19 ± 0.09	1.19 (8%)	1.20 ± 0.06	1.19 (5%)	100.74 (98.89, 102.62)
Fluctuation (%)	188 ± 56	181 (27%)	182 ± 45	177 (24%)	97.98 (93.74, 102.42)

Abbreviations: C_{max}, maximum plasma drug concentration for a single dose; $C_{max,ss}$, maximum plasma drug concentration during a steady-state dosing interval; t_{max}, time of occurrence for C_{max}; $t_{max,ss}$, time of occurrence for $C_{max,ss}$; AUC_{0-8h}, area under the plasma drug concentration-time curve from 0 to 8 h post dose; $AUC_{0-\infty}$, area under the plasma drug concentration-time curve from time zero to infinite time for a single dose; AUC_{0-t}, area under the plasma drug concentration-time curve during a steady-state dosing interval; F, bioavailable fraction for oral dosing; $t_{1/2,z}$, elimination half-life for the terminal phase; MRT, mean residence time; CL, clearance; CL/F, apparent clearance for oral dosing; V_{ss}, volume of distribution at steady-state; V_z, volume of distribution in the terminal phase; V_z/F, apparent volume of distribution in the terminal phase for oral dosing; ; $C_{min,ss}$, minimum plasma drug concentration during a steady-state dosing interval; CI, Confidence Interval; NA, not applicable; CV% coefficient of variation percentage.

[1] Median and range are presented.

[2] Wilcoxon signed rank test. No result available for the IV dose since values for each subject were identical between the 2 assays.

Source: Summary Table 7-5 and Summary Table 7-6; Appendix 5 and Appendix 6

DISCUSSION:

The objective of this report was to compare ciprofloxacin PK based on reassay results with the PK based on the original assay results. Subjects in this study were administered a single 40 mg/kg IV infusion raxibacumab dose, with or without co-administered ciprofloxacin (IV or PO). Reassays of the duplicate samples from the study were performed using a validated method at a different laboratory to assess the validity of the original assay. The documented difference between the 2 assay methods was: internal standard for original assay and reassay were enrofloxacin and ciprofloxacin-d8, respectively. The lower limit of quantitation for both the original assay and the reassay was 10 ng/mL of ciprofloxacin in 100% plasma. Pharmacokinetic analysis of the plasma drug concentration results was performed in the same manner for both the original results and the reassay results.

Comparison of the original and reassay plasma ciprofloxacin concentrations revealed some unexpected group-dependent differences between the two sets of results. For concentrations stratified by nominal time and dose number in Group 1, the reassay to original assay (R/O) geometric mean ratios ranged from 106.61 to 116.72 with all 90% CIs within the equivalence range. In addition, the two assays were equivalent for all PK parameters for both Doses 9 and 11 in Group 1. Since the original conclusion that raxibacumab did not affect the PK of ciprofloxacin was based on the results obtained for Group 1, the PK equivalence between reassay and the original assay for Group 1 fully supports that original conclusion.

For concentrations stratified by nominal time and dose number in Group 3, the R/O geometric mean ratios ranged from 117.99% to 140.17%. All 90% CIs fell outside of the equivalence range, with the reassay results higher than the original results at all collection times. Consequently, non-equivalence was found for some PK parameters between the two assays. Examination and comparison of the bioanalytical reports for the original assays and the reassay did not reveal any explanation for the discrepancies noted. The reason for these discrepancies remains unclear.

APPLICANT'S CONCLUSIONS:
Relative to the original assay, the reassay resulted in approximately the same mean ciprofloxacin concentration-time profiles for Group 1 doses, but the reassay results were about 20% higher than the original assay results for Group 3. Ciprofloxacin concentrations were equivalent between the 2 assays at all collection times for Group 1, but not equivalent at every collection time for Group 3. Differences were confined to Group 3.

For Group 1, equivalence between the reassay and original assay results was shown for all PK parameters for both Doses 9 and 11. There were no statistically significant differences in $t_{max,ss}$ for Dose 9 and Dose 11 between the 2 assays for Group 1. For Dose 1 in Group 3, equivalence between the reassay and original assay results was shown for $t_{1/2z}$ and MRT but not for C_{max}, $AUC_{0\text{-}inf}$, CL, V_{ss}, or V_z since the 90% CI fell slightly outside the 80-125% range for those parameters. The t_{max} estimates were the same for each subject by the 2 assays. For Dose 15 in Group 3, equivalence between the reassay and original assay results could be declared for $t_{1/2z}$, accumulation index, fluctuation and bioavailability, but could not be declared for $C_{max,ss}$, $AUC_{0\text{-}tau}$, $AUC_{0\text{-}8}$, CL/F, $C_{min,ss}$, or V_z/F since the 90% CI fell slightly outside the 80-125% range for these parameters. There was no statistically significant difference in $t_{max,ss}$ between the 2 assays.

- Between the original assay and reassay, ciprofloxacin PK was equivalent for all parameters in Group 1, but was not equivalent for all parameters in Group 3.
- There appears to be no clinically meaningful differences between the plasma ciprofloxacin concentrations generated by the 2 assays given the comparable results in Group 1 and the similar results in Group 3, where the 90% CI values of R/O geometric mean ratios were generally within the range of 110-135%.

REVIEWER ASSESSMENT:
The pre-specified equivalence criteria were met for the comparison of the original assay and the reassay with respect to both individual mean concentrations and the derived pharmacokinetic parameters for ciprofloxacin for Group 1. However, the mean ciprofloxacin Group 3 concentrations were systematically higher when reassayed than they were with the original assay. These higher concentrations led to the calculation of ciprofloxacin pharmacokinetic parameters that fell outside the 80-125% no-effect boundary when compared to the parameters derived using the ciprofloxacin concentrations determined by the original assay. The reasons for this disparity are unclear.

Ciprofloxacin was dosed differently in Group 1 versus Group 3 (PO vs. IV/PO); however, the dosing route seems unlikely to play a role in a systematic error in assessed concentrations. The ciprofloxacin label reports steady state AUC and C_{max} data at steady state following 400 mg q12h IV dosing and 500 mg q12h PO dosing. Although this does not directly parallel Group 3 in the current study, it is noteworthy that the steady state PK parameters reported in the ciprofloxacin label are larger than either the original analysis or the reanalysis (see Table below).

PK Parameter	Original Assay (Group 3)	Re-assay (Group 3)	Ciprofloxacin label[1]
$AUC_{0\text{-}tau}$ (ng*h/mL)	6599	7880	13700
C_{max} (ng/mL)	1195	1421	2970

[1]: Results are reported are for steady state following the administration of 500 mg ciprofloxacin q12h PO.

There is a larger numerical difference between the pharmacokinetic data contained in the ciprofloxacin label for a similar but not identical dosing scheme than there is between the original assay and the reassay for the Group 3 ciprofloxacin PK data.

Even though the pre-specified equivalence criteria was not met for the Group 3 ciprofloxacin concentrations, the Reviewer agrees with the applicant's argument that the differences between the assays is not clinically relevant. Therefore, the difference between the two analytical methods for the Group 3 concentrations and parameters is not of sufficient magnitude as to require additional studies or qualifying language.

APPENDIX 2
OFFICE OF CLINICAL PHARMACOLOGY:
PHARMACOMETRIC REVIEW

1 SUMMARY OF FINDINGS

1.1 Key Review Questions

The purpose of this review is to address the following key questions.

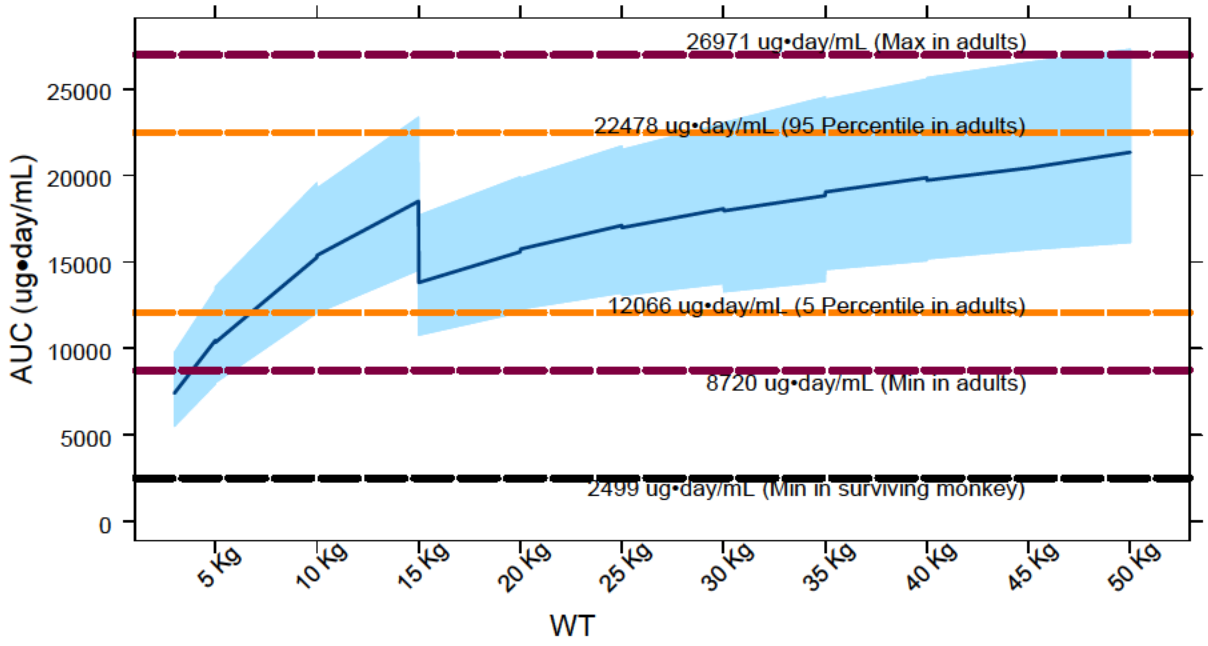
1.1.1 What is an appropriate raxibacumab dosing regimen for pediatrics (birth-16 years) to match exposures to that of adults after 40 mg/Kg IV dose?

The recommended dose for pediatric population is described in Table 1. As shown in Figure 1 and Figure 2, the proposed dosing recommendations reasonably match AUC and Cmax in healthy adults after 40 mg/Kg dose. The proposed dosing is also simple to implement in emergency settings.

Table 1. Raxibacumab dosing recommendations for pediatric patients based on body weight (WT)

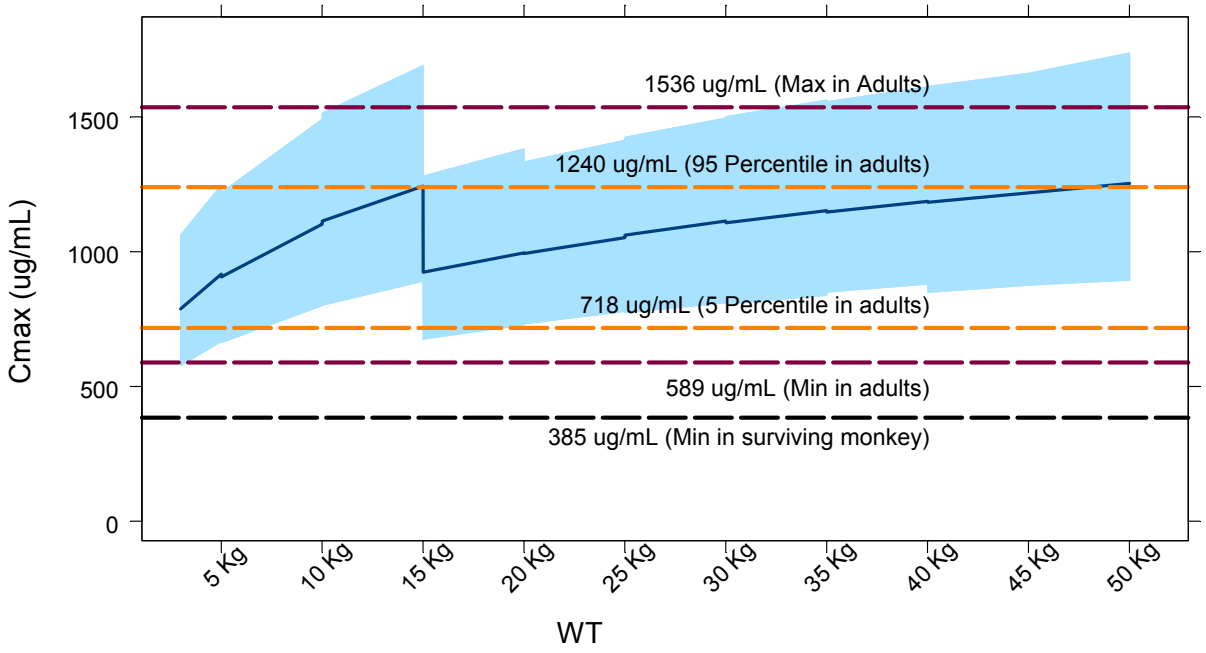
WT (Kg)	IV Dose (mg/kg)
≤15	80
>15 to ≤50	60
>50	40

Figure 1. Predicted raxibacumab exposures (AUC) at the proposed dosing recommendations in Table 1. The solid blue line and shaded area represent median and predicted 5th, and 95th percentile of raxibacumab AUC in pediatric population. The dash lines represent the range (minimum and maximum), 5th and 95th percentile AUC for adults (predicted from posthoc estimate by sponsor) at 40 mg/Kg dose[a] and minimum observed AUC in surviving monkey[b] at 20 mg/Kg.



a. From Summary Table 9, HGS1021-POP01.PK in Clinical Pharmacokinetics Report
b. From Appendix 37, HGS1021-POP01.PK in Clinical Pharmacokinetics Report

Figure 2. Predicted raxibacumab Cmax at the proposed dosing recommendations in Table 1. The solid blue line and shaded area represent predicted 5th, median and 95th percentile of raxibacumab Cmax in pediatric population. The dash lines represent the range (min and max), 5th and 95th percentile Cmax for adults (predicted from posthoc estimate by sponsor) at 40 mg/Kg dose[a] and min observed Cmax in surviving monkey[b].



a. From Summary Table 9, HGS1021-POP01.PK in Clinical Pharmacokinetics Report
b. From Appendix 37, HGS1021-POP01.PK in Clinical Pharmacokinetics Report

1.2 Recommendations

Dosing recommendation presented in Table 1 can match raxibacumab exposures in pediatrics to exposures in adults after 40 mg/Kg IV raxibacumab and it is simple to implement under emergency.

2 PERTINENT REGULATORY BACKGROUND

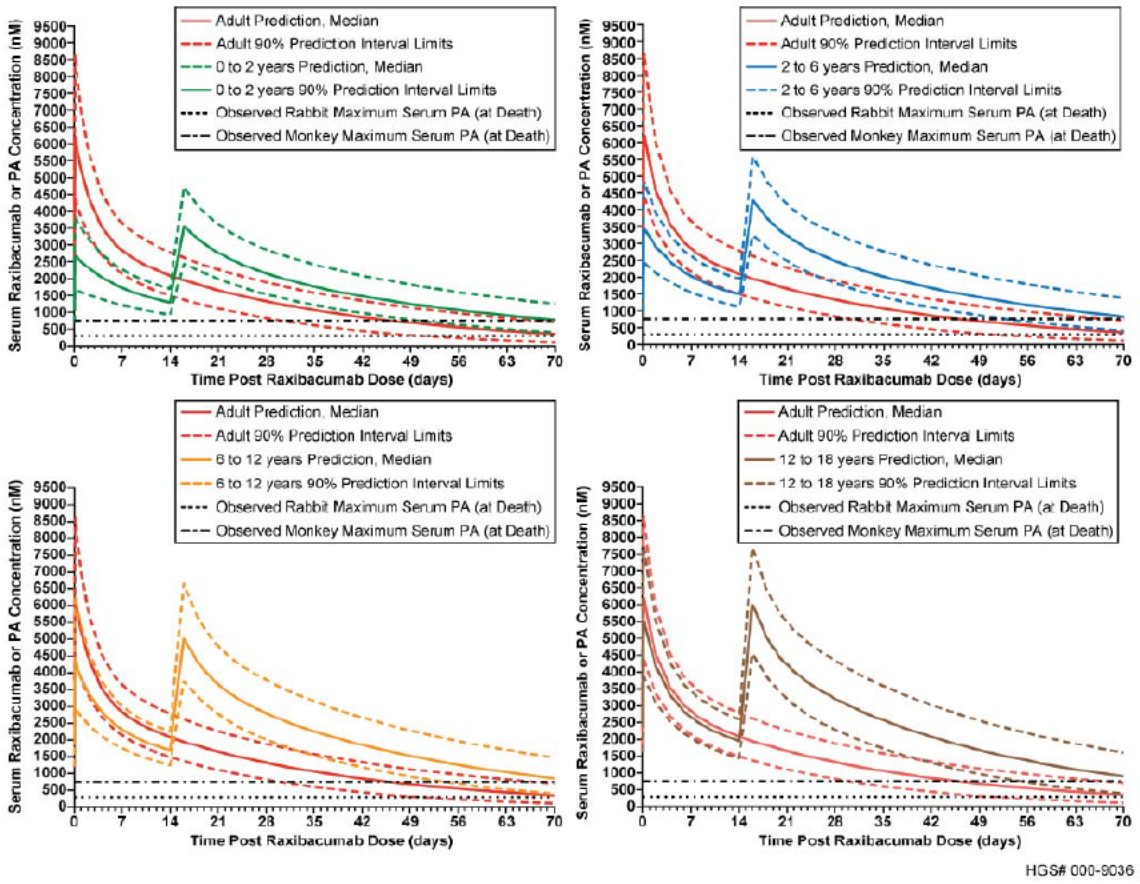
Raxibacumab is an unapproved investigational product. The Centers for Disease Control and Prevention is sponsoring the use of intravenous raxibacumab for treatment of inhalation anthrax during an emergency involving *Bacillus anthracis*. The Centers for Disease Control and Prevention (CDC) also submitted a protocol entitled "Intravenous Administration of Raxibacumab as a Therapeutic Agent for Treatment of Anthrax" on

December 22, 2008. The protocol specified that if raxibacumab administration is deemed necessary in the pediatric population, then theoretical doses for such patients need to be determined. The protocol, however, did not provide any details on how to derive such doses (see Dr. Lim's review, 1/23/09).
At the time of this review, there were no pediatric PK, efficacy and safety data available. This review aims at deriving dosing recommendations for raxibacumab IV in pediatric patients.

3 RESULTS OF SPONSOR'S ANALYSIS

The sponsor proposed a pediatric dosing recommendation in the cover letter submitted to agency (BB-IND 11069**,** Dec 14, 2011**)**. Due to the lack of raxibacumab pharmacokinetics (PK) data in children, pediatric exposures were simulated according to 3 scenarios: 1. using mg/kg scaling (allometric exponents of 1 for all PK parameters); 2. using allometric exponents determined from modeling of adult data; and, 3. using allometric exponents published in the literature (*Deng et al, Landes Bioscience, 2011, p61*). Simulations given by using method 2 and 3 suggested that the concentration in age<12 yrs may be lower than that observed in adults. In addition, for pediatric population, the duration of concentration above maximum serum PA in monkey is around 14 days, which is shorter than 28 days in adults. Therefore, based on the simulation results (Figure 3),it is concluded that an initial IV 40 mg/kg raxibacumab dose is recommended for all pediatric subjects, and in subjects under the age of 12 years, administration of a second IV 40 mg/kg raxibacumab dose 14 days after the first dose should be considered if clinical signs and symptoms suggest that PA toxemia has not resolved. 2 doses administered 14 days apart were shown to be safe in the Phase 3 study in healthy adults subjects (HGS Report HGS1021-C1063.CSR).

Figure 3. Median and 90% prediction interval serum raxibacumab concentration-time profiles for two 40 mg/kg raxibacumab IV infusion doses twice given 14 days apart, relative to the highest expected serum/plasma PA concentrations, for model using method 2



Source: Sponsor's cover letter (BB-IND 11069, Dec 14th,2011)
Reviewer's comment: The results of modeling and simulations by sponsor are acceptable. The models described the adults' PK data adequately. The simulation results suggested that 2nd after 14 days of first dose for subject<12 yrs will provide prolonged exposure. However, there are several potential limitations in sponsor's dosing proposal.1).the Cmax in younger subjects (<12 yrs) is lower than that in adults. Lower Cmax may lead to a lower survival based on results in animal studies (see pervious sponsor submission BLA 125349 and associated PM review by Dr. Kimberly Bergman, 12/10/ 2009). 2).A second dose at 14 days is a cautious approach. This may not be appropriate given that the inhalational anthrax is a life-threatening condition. The most efficacious dose should be given immediately (b) (4) .

4 REVIEWER'S ANALYSIS

This section describes methodological details used in deriving the pediatric dosing recommendations.

4.1 Introduction

FDA has made provision for approval of drugs or biologics when it is unethical to assess the efficacy in controlled human clinical trials as outlined in 21CFR601.90 through 21CFR601.95 "Evidence Needed to Demonstrate Effectiveness of New Drugs When Human Efficacy Studies Are Not Ethical or Feasible." Based on rabbit and monkey PK/PD and efficacy studies and human PK and safety studies, sponsor concluded that the 40 mg/kg raxibacumab IV dose can provide adults with the desired level of protection against lethality.

The PK and safety profile of 40 mg/Kg IV raxibacumab in healthy adults was characterized in 4 clinical studies: a Phase 1 study in healthy volunteers, a drug interaction study of raxibacumab with ciprofloxacin conducted in healthy volunteers, an immunogenicity and safety study in healthy volunteers who received a 2nd raxibacumab dose 4 months after their initial dose, and a Phase 3 study in healthy volunteers. Clinical review of IND 102964 (Dr. Eileen Navarro, Jan 2009) says" *Based on the totality of the preclinical and clinical data reviewed to date, and given that the use of raxibacumab under this CDC IND will be under the safeguard of a product with known quality, under the direct supervision of a physician and principal investigator, with full patient consent and following a clinical protocol with investigator responsibilities under the FDC Act, the review team finds it safe to proceed with the investigational use of Raxibacumab 40 mg/kg for severe systemic anthrax.* "

4.2 Objectives

The objective of this analysis is to derive a simple pediatrics dosing regimen to match adults PK (AUC and Cmax) receiving 40 mg/Kg .

4.3 Methods

4.3.1 Data Sets

Data excluded from the PopPK analysis, with brief explanation of exclusion, was listed in Appendix 5 in Clinical Pharmacokinetic Report for Protocol HGS1021-POP01 provided by sponsor. Pharmacokinetic data for this analysis are collected from 322 healthy adults in a total of 3 clinical studies as Table 2. Figure 4 illustrates distribution of age and body weight of healthy adults in the PK dataset. Of the 322 subjects, 150 (47%) were male and 172 (53%) were female; 230 (71%) were White, 52 (16%) were Black, and 15 (5%) were Asian; 272 (84%) were non-Hispanic and 50 (16%) were Hispanic; and, 301 (93%) were < 65 years of age and 21 (7%) were ≥ 65 years of age with a range of 18 to 87 years and weight range of 45 to 156 Kg.

Table 2. Clinical Data for PopPK analysis[a]

Protocol No. Study Title Number of Study Centers	Phase	Study Population	IV Dose (mg/kg)	Regimen	Number of Evaluable Subjects
HGS1021-C1063 A Randomized, Single-Blind, Placebo-Controlled Study to Evaluate the Safety and Tolerability of Raxibacumab (Human Monoclonal Antibody to *B. Anthracis* Protective Antigen) in Healthy Subjects 6 US centers	3	Healthy volunteers (54% females; 12% Hispanic, 21% non-white; ≥ 18 years of age, 9% ≥ 65 years of age)	40 40	Single dose 2 doses given 14 days apart	217[1] 23[1]
HGS1021-C1064 An Open-Label Study to Evaluate the Pharmacokinetics and Safety of Raxibacumab (Human Monoclonal Antibody to *B. anthracis* Protective Antigen) Administered in Combination with Ciprofloxacin in Healthy Subjects 3 US centers	2/3	Healthy volunteers (43 males, 45 females; 53 White, 31 Black or African American, 4 other; 18 to 60 years of age)	40[2] 40 40[3]	Single dose Single dose Single dose	30 28 28
HGS1021-C1069 An Open-Label Study to Evaluate the Immunogenicity and Safety of Raxibacumab (Human Monoclonal Antibody to *B. anthracis* Protective Antigen) Administered in Healthy Subjects 2 US centers	2/3	Healthy volunteers recruited from HGS1021-C1064 subjects (12 males, 8 females; 13 White, 7 Black or African American; 23 to 61 years of age)	40	2 doses, given ≥ 4 months after 1 prior dose (in HGS1021-C1064)	20[4]
				Total	326

[1] Number of subjects for raxibacumab-treated group.

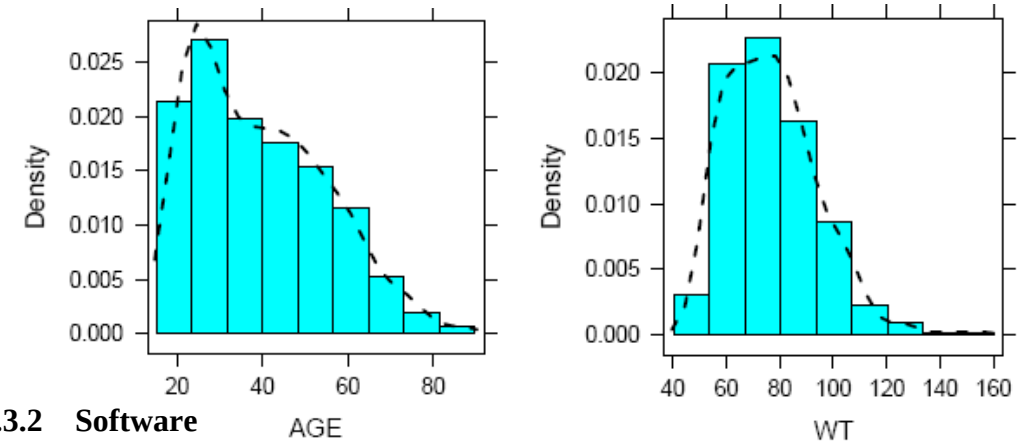
[2] Raxibacumab was administered concurrent with oral (PO) 500 mg q12 hour x 15 ciprofloxacin doses; the raxibacumab dose was administered immediately following the 11th ciprofloxacin dose.

[3] Raxibacumab was administered concurrent with IV 400 mg q12 hour x 2 ciprofloxacin doses, which were followed by PO 500 mg q12 hour x 13 ciprofloxacin doses; the raxibacumab dose was administered immediately following the 1st IV ciprofloxacin dose.

[4] Number of subjects, not included in the total.

a. From: Sponsor's Clinical Pharmacokinetic Report

Figure 4. Distribution of Age and Body weight of healthy adults



4.3.2 Software

NONMEM version VI was used for pharmacokinetic modeling and simulation. Splus version 6.2 and EXCEL were used for data formatting, plotting and simulation.

4.3.3 Models

4.3.3.1 Pharmacokinetic modeling of raxibacumab data from healthy adults at 40 mg/Kg

Raxibacumab is a humanized monoclonal antibody which is primarily metabolized through and barely excreted as unchanged drug in the urine (see pervious sponsor submission BLA 125349 and associated PM review by Dr. Kimberly Bergman, 12/10/ 2009). Therefore, the elimination of raxibacumab will not dependent on the renal or liver function. Based on the PK analysis conducted by sponsor and previous review, two compartment PK model with first order elimination in central compartment after IV administration adequately described pharmacokinetics of raxibacumab in healthy adults. PopPK analysis of raxibacumab in adult subjects has already demonstrated a clear relationship between clearance and body weight.

Similar relationships have been observed for other monoclonal antibodies (e.g., basiliximab, canakinumab, infliximab, certolizumab). Pediatric dosing has previously been derived using pharmacometrics approach in the absence of pediatric PK data, such as peramivir and pralidoxime.

We used an empirical model with estimated power exponent to describe the effect of body weight on PK (see parameterization in Table 3). The relationships between PK parameters (CL, Vd, Q and V2) and body weight and age were explored with different covariate models. Inter-individual variability for all PK parameters was included as exponent and mixed residual models (proportional and additive residuals) were used after attempting various residual structures.

Table 3. Parameter estimates from the final PK model in healthy adults

Parameter	Equation	Estimate (%RSE)
Clearance	$TVCL = Q1 \bullet \left(Body\ weight\ in\ Kg \Big/ 70 \right)^{Q5}$	$Q1 = 172\,(1.3)\ \text{mL} \bullet \text{day}^{-1} \bullet 70\text{Kg}^{-0.796}$ $Q5 = 0.796\,(7.7)$
Central compartment distribution volume	$TVV1 = Q2 \bullet \left(Body\ weight\ in\ Kg \Big/ 70 \right)^{Q6}$	$Q2 = 3020\,(1.0)\ \text{mL} \bullet 70\text{Kg}^{-0.752}$ $Q6 = 0.752\,(6.0)$
Intercompartmental clearance	$TVQ = Q3$	$Q3 = 452\,(7.7)\ \text{mL} \bullet \text{day}^{-1}$
Peripheral compartment distribution volume	$TVV2 = Q4$	$Q4 = 2210\ (2.5)\ \text{mL}$

4.3.4 Simulations to derive dosing recommendations in pediatric patients

The estimates for fixed effects and random effects from the modeling results of adults PK for raxibacumab were used as input for simulations. The body weight for subjects varied from 5 to 100 Kg by 5 Kg. Each group by body weight has 1000 subjects with the last observation time at 100 days to approximate AUC_{∞}. In most adults PK studies, the IV dose was infused for about 0.1 day (2.2-2.5 hr). Assuming that 0.1 day (2.4 hr) is reasonable for delivery of raxibacumab in pediatric population, the Cmax was evaluated to assess the duration of infusion for 0.1 day.

Several dosing scenarios were simulated as Table 4. In the base case scenario, the IV dose is 40 mg/Kg for all subjects in the simulations. In scenario I and II, the dose was adjusted for subjects with different body weight.

Table 4. Dosing Scenarios Simulated

Scenarios	Dosing Regimen	
Base case	40 mg/kg for all subjects	
Scenario I	**WT (Kg)**	**Dose (mg/kg)**
	≤5	100
	>5 to ≤10	80
	>10 to ≤15	70
	>15 to ≤35	60
	>35 to ≤50	50
Scenario II	**WT (Kg)**	**Dose (mg/kg)**
	≤15	80
	>15 to ≤50	60

4.4 Results

4.4.1 Pop PK model

The relationship between PK parameters and covariates was first explored based on the analysis of base model without any covariates. As shown in Figure 5 there is a clear relationship between post hoc CL, V1 and body weight, but Q and V2 is not dependent on body weight. The relationship between PK parameter and other covariates, such as age, was not significant.

Figure 5. Relationship between raxibacumab post hoc PK parameters and body weight from base model (model without any covariates)

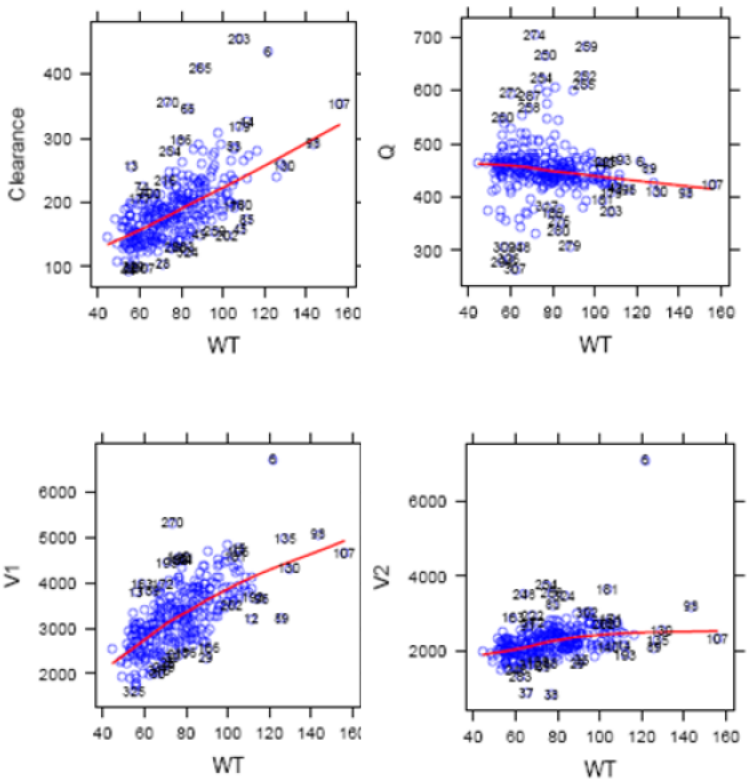
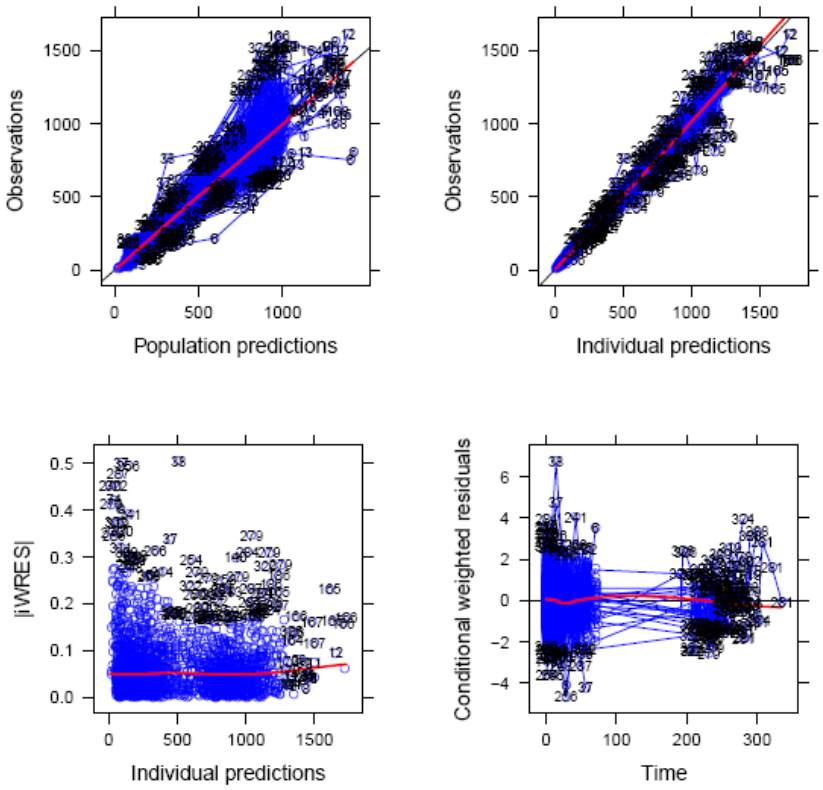


Table 5. Estimates of Inter-individual variability and residuals from the models with different covariates structure. Model A: Sponsor's model (See sponsor's Clinical Pharmacokinetic Report);Model B: Base model without any covariates; Model C: Model with effect of body weight on all PK parameters; Model D: Model with effect of body weight on CL, V1, and V2; Model E: Model with effect of body weight on CL and V1-Final Model

	Model A	Model B	Model C	Model D	Model E
CL (CV%)	20.0	26.5	20.0	19.9	19.9
V1 (CV%)	16.0	23.1	16.6	16.7	16.6
Q (CV%)	33.5	31.1	33.2	33.3	31.6
V2 (CV%)	21.2	26.7	21.4	21.9	26.4
Proportional residual (CV%)	9.5	9.7	9.6	9.6	9.6
Additive residual (ug/mL)	9.4	9.1	9.4	9.4	9.4

Final model (Model E) with body weight as covariate in CL and V1 was selected to simulate the PK profiles of pediatric population, although the inter-individual variability demonstrated a slight increase for V2 (peripheral volume) after the body weight was removed as covariate for V2. The diagnostic plots for the final model suggested it can describe raxibacumab pharmacokinetics in healthy adults adequately as shown in Figure 6. Further analysis was conducted to ensure that V2 and Q is not dependent on body weight. The PK parameterization of final model is listed in Table 3. The exponent estimate for CL and V1 were 0.796 (Q5) and 0.752 (Q6), respectively.

Figure 6. Diagnostic plots for the final PK model for healthy adults

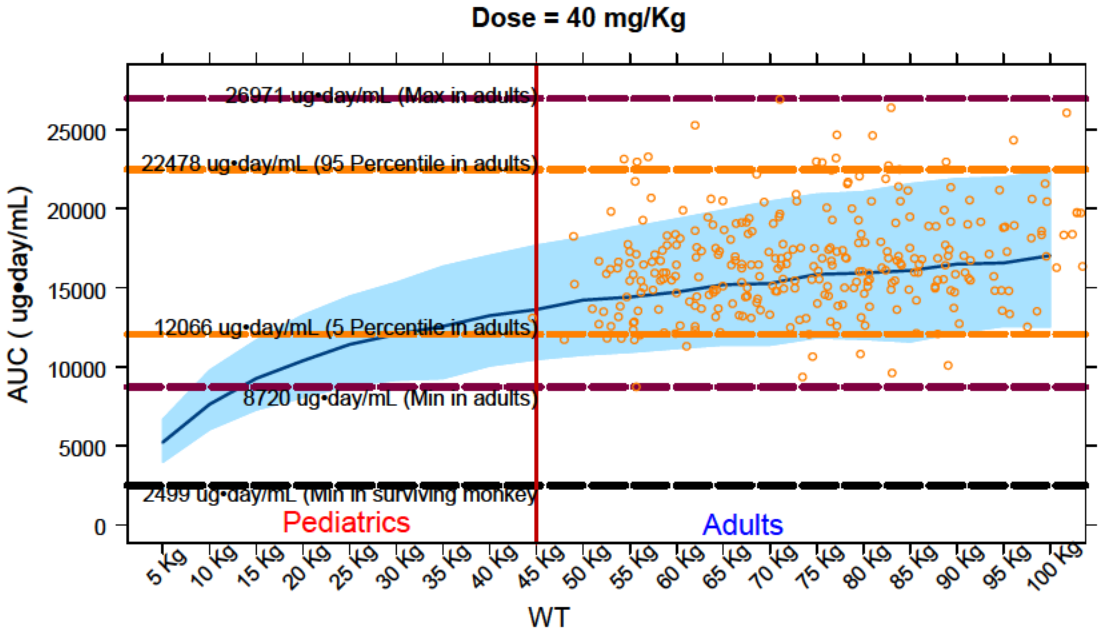


4.4.2 Simulations

Simulations were performed for a population with body weight range from 5-100 Kg at dose 40 mg/Kg (Base case in Table 4). The predicted AUC and Cmax were compared with those observed in healthy adults with body weight > 45 Kg as shown in Figure 7 and Figure 8. The 90% prediction interval of AUC and Cmax for adults was comparable with those seen in adults at 40 mg/Kg. However, 40 mg/Kg dose will not be able to provide similar exposure for population with body weight less than 45 Kg. To match the exposure level in pediatric population with that in adults, we explored several dose regimens for pediatric population with lighter body weight (Scenarios I and II in Table 4).
As shown in Figure 9 and Figure 10, the simulated exposure profiles in scenario I (Table 4) match well with the exposure of adults at 40 mg/Kg raxibacumab. In comparison with Scenario II, the dosing regimen in scenario I is more difficult to implement with a fine adjustment of dose based on body weight. Scenario II is relatively simple to implement in emergency and matches the exposure with adults at 40 mg/Kg raxibacumab. Therefore, dose regimen in scenario II (Table 4) is recommended.

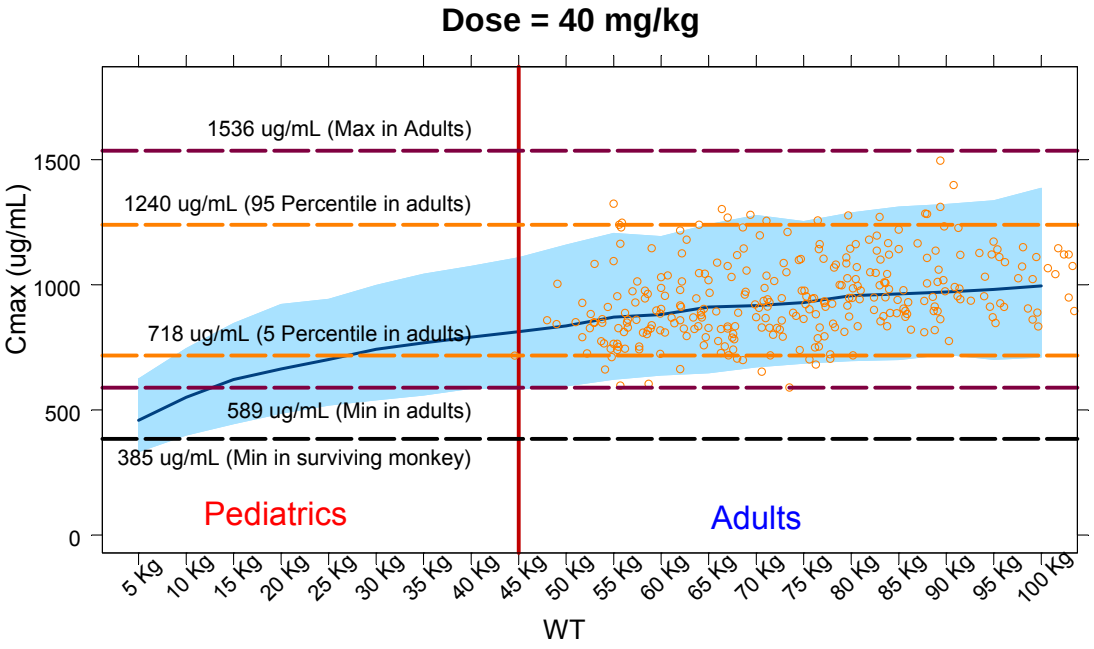
Figure 7. Predicted raxibacumab AUC at 40 mg/Kg IV dose (base case). The solid blue line and shaded area represent predicted 5th, median and 95th percentile of raxibacumab AUC in pediatric population. The dash lines represent the range (minimum and maximum), 5th and 95th percentile AUC for adults (predicted from

posthoc estimate by sponsor) at 40 mg/Kg dose[a] and minimum observed AUC in surviving monkey[b] at 20 mg/Kg.



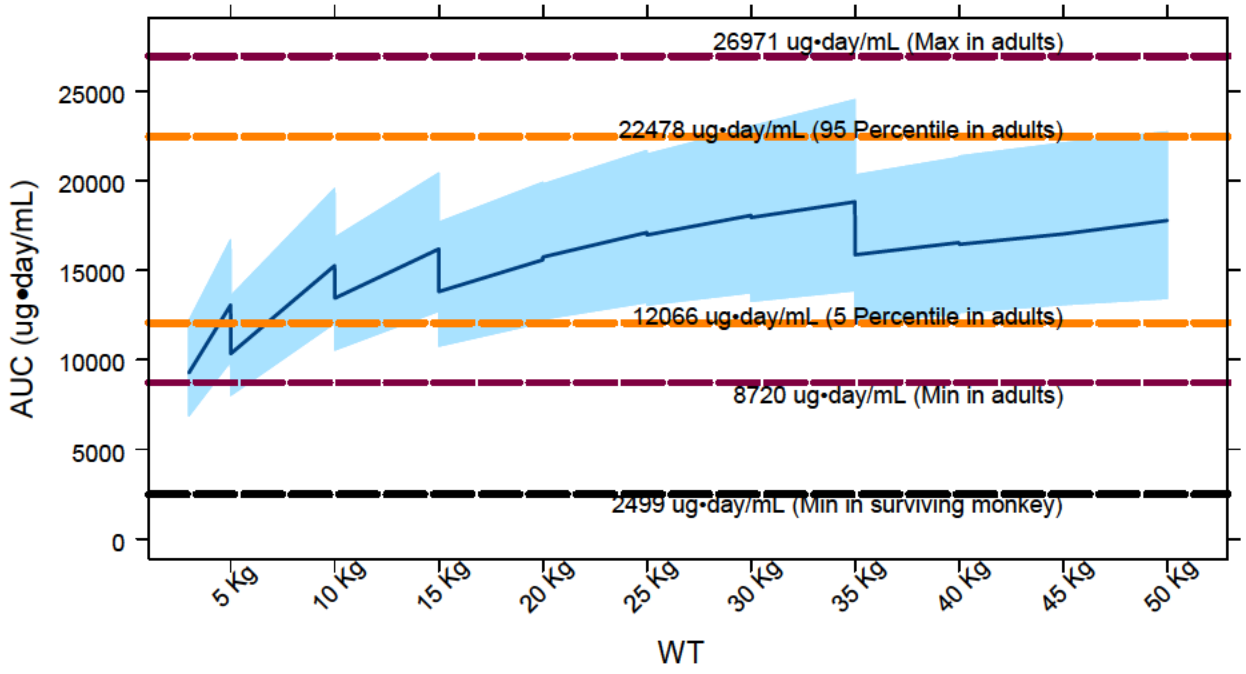
a. From Summary Table 9, HGS1021-POP01.PK in Clinical Pharmacokinetics Report
b. From Appendix 37, HGS1021-POP01.PK in Clinical Pharmacokinetics Report

Figure 8. **Predicted raxibacumab Cmax at 40 mg/Kg IV dose (base case). The solid blue line and shaded area represent predicted 5th, median and 95th percentile of raxibacumab Cmax in pediatric population. The dash lines represent the range (minimum and maximum), 5th and 95th percentile AUC for adults (predicted from posthoc estimate by sponsor) at 40 mg/Kg dose[a] and minimum observed AUC in surviving monkey[b] at 20 mg/Kg.**



a. From Summary Table 9, HGS1021-POP01.PK in Clinical Pharmacokinetics Report
b. From Appendix 37, HGS1021-POP01.PK in Clinical Pharmacokinetics Report

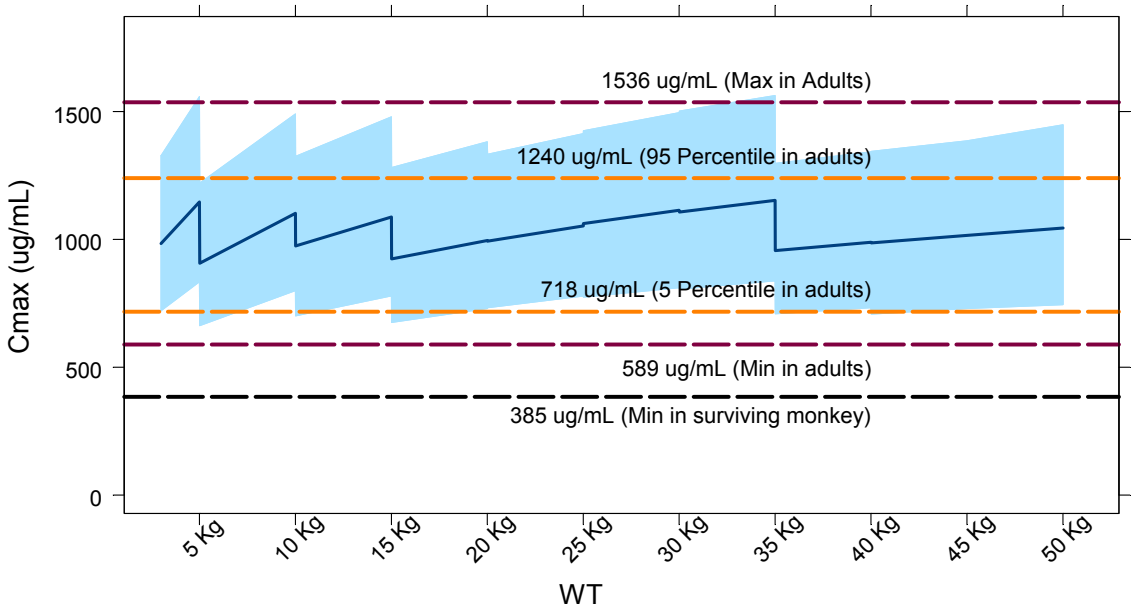
Figure 9. Predicted raxibacumab AUC at dose proposed in scenario I. The solid blue line and shaded area represent median and predicted 5th, and 95th percentile of raxibacumab AUC in pediatric population. The dash lines represent the range (minimum and maximum), 5th and 95th percentile AUC for adults (predicted from posthoc estimate by sponsor) at 40 mg/Kg dose[a] and minimum observed AUC in surviving monkey[b] at 20 mg/Kg.



a. From Summary Table 9, HGS1021-POP01.PK in Clinical Pharmacokinetics Report
b. From Appendix 37, HGS1021-POP01.PK in Clinical Pharmacokinetics Report

Figure 10. Predicted raxibacumab Cmax at doses proposed in scenario I. The solid blue line and shaded area represent median and predicted 5th, and 95th percentile of raxibacumab AUC in pediatric population. The dash lines represent the range (minimum and maximum), 5th and 95th percentile AUC for adults (predicted from

posthoc estimate by sponsor) at 40 mg/Kg dose[a] and minimum observed AUC in surviving monkey[b] at 20 mg/Kg.



a. From Summary Table 9, HGS1021-POP01.PK in Clinical Pharmacokinetics Report
b. From Appendix 37, HGS1021-POP01.PK in Clinical Pharmacokinetics Report

5 LISTING OF ANALYSES CODES AND OUTPUT FILES

File Name	Description	Location in \\cdsnas\pharmacometrics\
Readme.txt	A detailed description for the content in corresponding folder	…\Reviews\Ongoing PM Reviews \Raxibacumab_IND102964_JYU\PPK_Analyses\Modeling\ …\Reviews\Ongoing PM Reviews \Raxibacumab_IND102964_JYU\PPK_Analyses\Simulations\

This is a representation of an electronic record that was signed electronically and this page is the manifestation of the electronic signature.

/s/

RYAN P OWEN
11/21/2012

KIMBERLY L BERGMAN
11/21/2012

CLINICAL PHARMACOLOGY FILING FORM/CHECKLIST FOR NDA/BLA SUBMISSIONS

Office of Clinical Pharmacology

New Drug Application Filing and Review Form

NDA/BLA Number	**BLA 125349**	**Brand Name**	**TBD**
OCP Division (I, II, III, IV, V)	**DCP4**	**Generic Name**	**Raxibacumab**
Medical Division	**DAIP**	**Drug Class**	**Monoclonal antibody**
OCP Reviewer	**Ryan Owen, PhD**	**Indication(s)**	**Treatment of inhalational anthrax**
OCP Team Leader	**Kimberly Bergman, PharmD**	**Dosage Form**	**Intravenous solution**
Pharmacometrics Reviewer	**NA**	**Dosing Regimen**	**40 mg/kg single dose**
Date of Submission	**June 15, 2012**	**Route of Administration**	**Intravenous**
Estimated Due Date of OCP Review	**November 21, 2012**	**Sponsor**	**Human Genome Sciences**
Medical Division Due Date	**November 30, 2012**	**Priority Classification**	**Resubmission**
PDUFA Due Date	**December 15, 2012**	**AC Meeting (if applicable)**	**November 2, 2012**

Clinical Pharmacology and Biopharmaceutics Information

	"X" if included at filing	Number of studies submitted	Number of studies reviewed	Critical Comments If any
STUDY TYPE				
Table of Contents present and sufficient to locate reports, tables, data, etc.	**X**			
Tabular Listing of All Human Studies	**X**			
HPK Summary	**X**			
Labeling	**X**			
Reference Bioanalytical and Analytical Methods				
I. Clinical Pharmacology				
Mass balance:	**NA**			
Isozyme characterization:	**NA**			
Blood/plasma ratio:	**NA**			
Plasma protein binding:	**NA**			
Pharmacokinetics (e.g., Phase I) -				
HEALTHY VOLUNTEERS -				
single dose:	**X**	**2**	**2**	Reviewed under original submission dated May 13, 2009
multiple dose:	**X**	**1**	**1**	Reviewed under original submission dated May 13, 2009
PATIENTS -	**NA**			BLA submitted under FDA's Animal Rule; indication unable to be studied in humans
single dose:				
multiple dose:				
Dose proportionality -				
fasting / non-fasting single dose:				
fasting / non-fasting multiple dose:				
Drug-drug interaction studies -				
In-vivo effects on primary drug:				
In-vivo effects of primary drug:				
In-vitro:				
Subpopulation studies -				
ethnicity:				
gender:				
pediatrics:				

CLINICAL PHARMACOLOGY FILING FORM/CHECKLIST FOR NDA/BLA SUBMISSIONS

geriatrics:				
renal impairment:				
hepatic impairment:				
PD -	**NA**			BLA submitted under FDA's Animal Rule; indication unable to be studied in humans; protective antigen kinetics in pivotal animal species were modeled (reviewed during original review cycle)
Phase 2:				
Phase 3:				
PK/PD -	**NA**			BLA submitted under FDA's Animal Rule; indication unable to be studied in humans; CNS toxicity findings in pivotal animal species were subject of PK/PD anlyses (reviewed during original review cycle)
Phase 1 and/or 2, proof of concept:				
Phase 3 clinical trial:				
Population Analyses -	**NA**			BLA submitted under FDA's Animal Rule; indication unable to be studied in humans
Data rich:				
Data sparse:				
II. Biopharmaceutics	**NA**			Intravenous product
Absolute bioavailability				
Relative bioavailability -				
solution as reference:				
alternate formulation as reference:				
Bioequivalence studies -				
traditional design; single / multi dose:				
replicate design; single / multi dose:				
Food-drug interaction studies				
Bio-waiver request based on BCS				
BCS class				
Dissolution study to evaluate alcohol induced dose-dumping				
III. Other CPB Studies				
Genotype/phenotype studies	**NA**			
Chronopharmacokinetics	**NA**			
Pediatric development plan	**NA**			BLA submitted under FDA's Animal Rule; indication unable to be studied in humans; pediatric dosing recommendations based on Mab priors and M/S conducted by FDA
Literature References	**NA**			
TOTAL NUMBER OF STUDIES	***3***			***Reviewed under original submission dated May 13, 2009; the clinical pharmacology review of this submission will focus on outstanding bioanalytical validation analyses to address OSI findings during the first review cycle.***

CLINICAL PHARMACOLOGY FILING FORM/CHECKLIST FOR NDA/BLA SUBMISSIONS

On **initial** review of the NDA/BLA application for filing:

	Content Parameter	Yes	No	N/A	Comment
Criteria for Refusal to File (RTF)					
1	Has the applicant submitted bioequivalence data comparing to-be-marketed product(s) and those used in the pivotal clinical trials?			X	TBM formulation and manufacturing process studied in pivotal animal models and in healthy subjects.
2	Has the applicant provided metabolism and drug-drug interaction information?	X			
3	Has the sponsor submitted bioavailability data satisfying the CFR requirements?	X			
4	Did the sponsor submit data to allow the evaluation of the validity of the analytical assay?	X			
5	Has a rationale for dose selection been submitted?	X			
6	Is the clinical pharmacology and biopharmaceutics section of the NDA organized, indexed and paginated in a manner to allow substantive review to begin?	X			
7	Is the clinical pharmacology and biopharmaceutics section of the NDA legible so that a substantive review can begin?	X			
8	Is the electronic submission searchable, does it have appropriate hyperlinks and do the hyperlinks work?	X			
Criteria for Assessing Quality of an NDA (Preliminary Assessment of Quality)					
	Data				
9	Are the data sets, as requested during pre-submission discussions, submitted in the appropriate format (e.g., CDISC)?	X			
10	If applicable, are the pharmacogenomic data sets submitted in the appropriate format?			X	
	Studies and Analyses				
11	Is the appropriate pharmacokinetic information submitted?	X			
12	Has the applicant made an appropriate attempt to determine reasonable dose individualization strategies for this product (i.e., appropriately designed and analyzed dose-ranging or pivotal studies)?	X			Weight-based dosing is proposed for this monoclonal antibody.
13	Are the appropriate exposure-response (for desired and undesired effects) analyses conducted and submitted as described in the Exposure-Response guidance?			X	
14	Is there an adequate attempt by the applicant to use exposure-response relationships in order to assess the need for dose adjustments for intrinsic/extrinsic factors that might affect the pharmacokinetic or pharmacodynamics?			X	

CLINICAL PHARMACOLOGY FILING FORM/CHECKLIST FOR NDA/BLA SUBMISSIONS

15	Are the pediatric exclusivity studies adequately designed to demonstrate effectiveness, if the drug is indeed effective?			X	
16	Did the applicant submit all the pediatric exclusivity data, as described in the WR?			X	
17	Is there adequate information on the pharmacokinetics and exposure-response in the clinical pharmacology section of the label?	X			
	General				
18	Are the clinical pharmacology and biopharmaceutics studies of appropriate design and breadth of investigation to meet basic requirements for approvability of this product?	X			
19	Was the translation (of study reports or other study information) from another language needed and provided in this submission?			X	

IS THE CLINICAL PHARMACOLOGY SECTION OF THE APPLICATION FILEABLE?

Yes, the resubmission is fileable from a clinical pharmacology perspective.

Note: The clinical pharmacology information supporting approval of raxibacumab for treatment of inhalational anthrax under the FDA's Animal Rule was reviewed under the original BLA submission dated May 13, 2009. The clinical pharmacology review of this current resubmission will focus on outstanding bioanalytical validation analyses to address OSI findings identified during the first review cycle.

If the NDA/BLA is not fileable from the clinical pharmacology perspective, state the reasons and provide comments to be sent to the Applicant.

Not applicable.

Please identify and list any potential review issues to be forwarded to the Applicant for the 74-day letter.

None.

Reviewing Clinical Pharmacologist Date

Team Leader/Supervisor Date

This is a representation of an electronic record that was signed electronically and this page is the manifestation of the electronic signature.

/s/

RYAN P OWEN
07/26/2012

KIMBERLY L BERGMAN
07/26/2012

OFFICE OF CLINICAL PHARMACOLOGY ADDENDUM

BLA:	125349
Submission Date(s):	14MAY2009
Brand Name:	Not Applicable
Generic Name:	Raxibacumab
Primary Reviewer:	Kimberly L. Bergman, Pharm.D.
Team Leader:	Philip M. Colangelo, Pharm.D., Ph.D.
OCP Division:	DCP4
OND Division:	DSPTP
Applicant:	Human Genome Sciences, Inc.
Relevant IND(s):	BB-IND 11069
Submission Type; Code:	Original BLA submitted under 21 CFR 601 Subpart H
Formulation; Strength(s):	Raxibacumab injection for intravenous use; 50 mg/mL in 50 mL sterile, single-use vials containing 35.1 mL of liquid formulation per vial
Indication	(b) (4)

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1. EXECUTIVE SUMMARY

Raxibacumab (HGS1021, PA mAb, or ABthrax™) is a fully human monoclonal antibody that specifically binds the Protective Antigen (PA) of *Bacillus anthracis*, thereby preventing its association with the anthrax toxin receptor on host cells. The IND for raxibacumab (BB-IND 11069) was initiated in June 2003, and raxibacumab was granted Fast Track Status and Orphan Drug Designation (03-17550) for the treatment of inhalation anthrax on 15 August 2003 and 13 November 2003, respectively. On 30 January 2009, the Agency reviewed raxibacumab efficacy, safety, and product quality data and determined the data were sufficient to support the use of a single 40 mg/kg IV raxibacumab dose for the treatment of inhalational anthrax under a CDC-held IND, and currently, raxibacumab is being stored in the Strategic National Stockpile (SNS) for use under this CDC-held IND. The current BLA for raxibacumab (submitted 14 May 2009) seeks approval under the "Animal Efficacy Rule" (21 CFR 601, Subpart H, "Approval of Biological Products when Human Efficacy Studies are not Ethical or Feasible"). The indication currently being sought for raxibacumab is therapeutic treatment of inhalation anthrax. (b) (4) (b) (4) The proposed dosage and route of administration of raxibacumab is a single dose of 40 mg/kg administered as an intravenous (IV) infusion over approximately 2 hours. Raxibacumab is proposed to be administered alone or in combination with antibiotics.

To allow selection of an effective dose in humans for therapeutic treatment of anthrax, the applicant has submitted clinical pharmacology data for raxibacumab in humans and in the two pivotal animal species, rabbits and cynomolgus monkeys (Studies 682-G005758 and 724-G005829, respectively). The pharmacokinetics (PK) and safety of raxibacumab administered as the product proposed for licensure has been evaluated in a Phase 1 antibiotic interaction study with ciprofloxacin (HGS1021-C1064), a repeat dose immunogenicity study (HGS1021-C1069) and a Phase 2/3 safety study (HGS1021-C1063).

On October 20, 2009, the Office of Clinical Pharmacology primary reviewer was made aware of significant findings from an audit of analytical sites used for studies submitted under BLA 125349. In September 2009, Division of Scientific Investigations (DSI) inspections for the analytical portions of human studies HGS1021-C1063 and HGS1021-C1064 were conducted. During these inspections, multiple issues were identified and FDA Forms-483 were issued to the analytical sites responsible for analysis of raxibacumab and ciprofloxacin concentrations. Inspection findings during the audit conducted by DSI for clinical studies HGS1021-C1063 and HGS1021-C1064 raised concerns regarding the reliability of the pharmacokinetic data submitted in the application and utilized for selection of an effective dose in humans for therapeutic treatment of anthrax.

1.1. Recommendation

The pharmacokinetic data for raxibacumab and ciprofloxacin in this BLA should be considered unreliable until the specific issues identified during the audit of analytical sites are satisfactorily addressed by the applicant.

1.2. Summary of Important Clinical Pharmacology Findings

Raxibacumab is a recombinant, fully human, IgG1λ monoclonal antibody that binds the Protective Antigen (PA) of *Bacillus anthracis* with high affinity and inhibits its biological activity. To allow selection of an effective dose in humans for therapeutic treatment of anthrax, the applicant has submitted clinical pharmacology data for raxibacumab in humans and in the two pivotal animal species, rabbits and cynomolgus monkeys (Studies 682-G005758 and 724-G005829, respectively). The pharmacokinetics (PK) and safety of raxibacumab administered as the product proposed for licensure has been evaluated in a Phase 1 antibiotic interaction study with ciprofloxacin (HGS1021-C1064), a repeat dose immunogenicity study (HGS1021-C1069) and a Phase 2/3 safety study (HGS1021-C1063).

Upon original review of this application by the Office of Clinical Pharmacology (OCP), the clinical pharmacology information submitted by the applicant was considered adequate to support the proposed human dose of 40 mg/kg and the suitability of the validated electro-chemiluminescence (ECL) assay used to measure raxibacumab serum concentrations in rabbits, monkeys and humans. The OCP review of BLA 125349 was finalized on October 13, 2009. On October 20, 2009, the OCP primary reviewer was made aware of significant findings from an audit of analytical sites used for studies submitted under BLA 125349. In September 2009, Division of Scientific Investigations (DSI) inspections for the analytical portions of human studies HGS1021-C1063 and HGS1021-C1064 were conducted. During these inspections, multiple issues were identified and FDA Forms 483 were issued to Human Genome Sciences and (b) (4) the analytical sites where the nonclinical and clinical pharmacokinetic samples were analyzed for raxibacumab and ciprofloxacin concentrations, respectively. The detailed DSI inspection report is presented in Appendix 1. Inspection findings during the audit conducted by DSI for clinical studies HGS1021-C1063 and HGS1021-C1064 raised concerns regarding the integrity and reliability of pharmacokinetic data submitted in the application and utilized for selection of an effective dose in humans for therapeutic treatment of anthrax.

On October 20, 2009, a teleconference was held between the review division (Division of Special Pathogen and Transplant Products; DSPTP) and the applicant. The applicant submitted a follow-up correspondence dated October 29, 2009 (BLA 125349/Submission Number 017) for the purpose of addressing the inspection issues pertaining to the raxibacumab assay. This submission contained a revised assay validation report, assay qualification report, and standard operating procedure (SOP), CLI-2879, for quantitation of raxibacumab in human serum. The applicant also proposed to re-assay pharmacokinetic samples for raxibacumab that were obtained in study HGS1021-C1064, the study used for comparison to rabbits and monkeys for purposes of supporting the therapeutic dose in humans.

Upon agency review of the applicant's submission and proposal to address the DSI inspection findings, it was determined that the revised assay validation, qualification, and SOP documents did not fully address the inspection issues identified in the Form FDA-483. The following recommendations were conveyed to the applicant via facsimile on November 6, 2009. It was emphasized that these comments should be addressed prior to re-assay of samples:

1. The calibration (standard) curve should consist of a minimum of six acceptable standard points. Calibration standards should be serum-based, and the calibration curve should have calibrator samples at the upper limit of quantitation (ULOQ) and at the lower limit of quantitation (LLOQ) to cover the expected study sample concentration range. We recommend that you select appropriate dilution factors so that ULOQ and LLOQ are on the linear segment of the ECL versus concentration plot.

2. Estimation of raxibacumab concentrations in unknown samples by extrapolation of standard curves below the LLOQ or above the ULOQ is not acceptable. Instead, the standard curve should be redefined or samples with higher concentration should be diluted to be within the calibration range and re-assayed.
3. Quality control (QC) samples in duplicate at three concentrations (one near the LLOQ, one in midrange, and one close to the ULOQ) should be incorporated in each assay run. All QCs should be serum-based.
4. The acceptance of analytical runs for the back-calculated concentrations should be based on the limits of accuracy (%RE) and precision (%CV). For the calibration standards in each run, a minimum of 75% of the back-calculated values (including the ULOQ) should fall within +20%, except for LLOQ and ULOQ, where it can be +25% of the nominal value. A minimum of 66% of the back-calculated QC concentrations, and at least 50% at each concentration, should fall within +20% of the nominal concentrations. The nominal concentrations, and not indirectly determined values, should be used.
5. Incurred sample reproducibility should be evaluated during the analytical runs.
6. The evaluation of stability/recovery for at least three freeze/thaw cycles should be repeated, documenting actual dates and times of each freeze/thaw cycle.
7. Following incorporation of our aforementioned comments and recommendations into the revised procedures, the pharmacokinetic samples from human studies HGS1021-C1063, HGS1021-C1064, and HGS1021-C1069 should be re-assayed using the revised procedures. Pharmacokinetic parameters for raxibacumab from these re-analyses should be re-calculated in humans following re-assay of the samples. Depending on the outcome of these re-analyses, additional pharmacokinetic studies may be required.
8. Because of the outstanding issues with the assay of raxibacumab in human serum, we have now become concerned about the reliability of the pharmacokinetic data that was generated from the animal efficacy studies in New Zealand White rabbits and Cynomolgus macaque monkeys, which used similar bioanalytical procedures as that for the human studies. Therefore, the pharmacokinetic samples from animal studies 682-G005758, 724-G005829, 781-G923701, and 789-G923702 should be re-assayed using the revised procedures, and with incorporation of our comments and recommendations mentioned above. Pharmacokinetic parameters for raxibacumab from these re-analyses should be re-calculated in rabbits and monkeys following re-assay of the samples. Depending on the outcome of these re-analyses, additional pharmacokinetic studies may be required.

A determination of the appropriateness of the proposed human dose of raxibacumab based on a cross-species comparison of raxibacumab exposure cannot be made until the DSI issues are resolved. Thus, the pharmacokinetic data for raxibacumab and ciprofloxacin in this BLA should be considered unreliable until the specific issues identified during the audit of analytical sites are satisfactorily addressed by the applicant.

12NOV2009

Kimberly L. Bergman, Pharm.D.
Office of Clinical Pharmacology
Division of Clinical Pharmacology 4

RD/FT Initialed by: 11/12/09

Philip M. Colangelo, Pharm.D., Ph.D
Team Leader - DSPTP

cc:
Division File: BLA 125349
HFD-590 (CSO/Saville)
HFD-590 (MO/Yasinskaya, Lim)
HFD-880 (Division File, Lazor, Reynolds, Colangelo)

2. APPENDICES

2.1. Division of Scientific Investigations Memorandum

OFFICE OF CLINICAL PHARMACOLOGY REVIEW

BLA:	125349
Submission Date(s):	14MAY2009
Brand Name:	Not Applicable
Generic Name:	Raxibacumab
Primary Reviewer:	Kimberly L. Bergman, Pharm.D.
Team Leader:	Philip M. Colangelo, Pharm.D., Ph.D.
PM Team Leader:	Pravin Jadhav, Ph.D.
OCP Division:	DCP4
OND Division:	DSPTP
Applicant:	Human Genome Sciences, Inc.
Relevant IND(s):	BB-IND 11069
Submission Type; Code:	Original BLA submitted under 21 CFR 601 Subpart H
Formulation; Strength(s):	Raxibacumab injection for intravenous use; 50 mg/mL in 50 mL sterile, single-use vials containing 35.1 mL of liquid formulation per vial
Indication	(b) (4)

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1. EXECUTIVE SUMMARY

Raxibacumab (HGS1021, PA mAb, or ABthrax™) is a fully human monoclonal antibody that specifically binds the Protective Antigen (PA) of *Bacillus anthracis*, thereby preventing its association with the anthrax toxin receptor on host cells. The anthrax toxin is a protein complex consisting of enzymatic (A) and binding (B) moieties. Lethal factor (LF) and edema factor (EF) proteins constitute the A moieties while PA constitutes the B moiety. The bacterially-secreted PA protein binds target cells of the host via the anthrax toxin receptor (ATR). The PA-ATR interaction initiates a series of molecular events that manifest as clinical disease. The indication currently being sought for raxibacumab is therapeutic treatment of inhalation anthrax, (b) (4) (b) (4)

The IND for raxibacumab (BB-IND 11069) was initiated in June 2003, and raxibacumab was granted Fast Track Status and Orphan Drug Designation (03-17550) for the treatment of inhalation anthrax on 15 August 2003 and 13 November 2003, respectively. On 30 January 2009, the Agency reviewed raxibacumab efficacy, safety, and product quality data and determined the data were sufficient to support the use of a single 40 mg/kg IV raxibacumab dose for the treatment of inhalational anthrax under a CDC-held IND, and currently, raxibacumab is being stored in the Strategic National Stockpile (SNS) for use under this CDC-held IND. The current BLA for raxibacumab seeks approval under the "Animal Efficacy Rule" (21 CFR 601, Subpart H, "Approval of Biological Products when Human Efficacy Studies are not Ethical or Feasible").

In accordance with the Animal Rule and because evaluation of new treatment options for inhalational anthrax is not possible in controlled clinical trials in humans for ethical reasons, the efficacy of raxibacumab was evaluated in two animal species, specifically New Zealand white rabbits and cynomolgus monkeys, with symptomatic anthrax disease (Studies Study 682-G005758 and 724-G005829, respectively). These pivotal efficacy studies were placebo-controlled, parallel-group randomized studies powered to provide a statistically significant result if a clinically important increase in survival was observed. Raxibacumab doses evaluated in both pivotal animal model efficacy studies were 20 and 40 mg/kg single doses administered intravenously. Because the current standard therapy for inhalation anthrax is post-exposure prophylaxis (PEP) with antimicrobials, the efficacy of antibiotics administered concomitantly with raxibacumab in the setting of therapeutic treatment also was evaluated in rabbits and monkeys with symptomatic anthrax disease (Studies 781-G923701 and 789-G923702). To allow selection of an effective dose in humans for therapeutic treatment of anthrax, the applicant has submitted clinical pharmacology data for raxibacumab in humans and in the two pivotal animal species, rabbits and cynomolgus monkeys. The pharmacokinetics (PK) and safety of raxibacumab administered as the product proposed for licensure has been evaluated in a Phase 1 antibiotic interaction study with ciprofloxacin (HGS1021-C1064), a repeat dose immunogenicity study (HGS1021-C1069) and a Phase 2/3 safety study (HGS1021-C1063). Overall, the safety of raxibacumab has been evaluated in over 400 healthy human volunteers, including 326 subjects treated with the proposed dose of 40 mg/kg with product manufactured and formulated by the same process proposed for licensure.

1.1. Recommendation

Based on a cross-species comparison of raxibacumab exposure and exposure-response information for raxibacumab dose/concentration, PA concentration, and survival, the clinical pharmacology information provided by the applicant is acceptable and supports the use of the 40 mg/kg dose of raxibacumab. The dose and concentration response relationship with the endpoint of survival indicates potential to achieve higher response rates at higher doses. If future trials are conducted, higher doses (e.g. 60 or 80 mg/kg)

should be explored along with 40 mg/kg. In any future trials studying higher doses in animals, safety assessments should include an examination of CNS pathology.

1.2. Phase 4 Commitments

No phase IV commitments are recommended.

1.3. Summary of Important Clinical Pharmacology Findings

Raxibacumab is a recombinant, fully human, IgG1λ monoclonal antibody that binds the Protective Antigen (PA) of *Bacillus anthracis* with high affinity and inhibits its biological activity. The clinical pharmacology characteristics of raxibacumab have been defined in rabbits, monkeys and humans. Raxibacumab dose selection is supported by a cross-species comparison of exposure and exposure-response (E/R) relationships described in in vivo animal models of inhalation anthrax infection. These studies show raxibacumab demonstrates the following clinical pharmacology characteristics:

- Following single intravenous administration of raxibacumab 40 mg/kg in healthy, male and female human subjects, mean raxibacumab steady-state volume of distribution was greater than plasma volume, suggesting some tissue distribution. Clearance values were much smaller than the glomerular filtration rate indicating that there is virtually no renal clearance of raxibacumab. These findings were similar in monkeys and rabbits.
- Humans achieve similar to or greater exposure to raxibacumab following a single 40 mg/kg IV dose compared to rabbits and monkeys receiving the same dose. Mean raxibacumab Cmax in humans following a single 40 mg/kg IV dose was similar to or greater than mean Cmax values in monkeys and rabbits. Mean raxibacumab AUCinf in humans following a single 40 mg/kg IV dose was 2.4 and 4.6-fold that of the mean AUCinf values in monkeys and rabbits, respectively. Mean total clearance of raximacumab was substantially slower in humans as compared to rabbits by a factor of approximately 5 and in monkeys by a factor of approximately 3. Thus, half-life of raxibacumab was longer in humans compared to mean half-lives observed in monkeys and rabbits (20.6 ± 6.5 days versus 10.1 ± 2.4 days and 4.1 ± 0.85 days, respectively). Variability in Cmax was similar across species; %CV values ranged between approximately 13 to 17%. Variability in AUCinf was wider, ranging between 13 and 32% across species.
- Serum raxibacumab concentration-time profiles were similar following administration of raxibacumab alone and with both PO and IV ciprofloxacin. Overall, exposure to ciprofloxacin appears to have no consistent or meaningful impact on raxibacumab PK. Likewise, ciprofloxacin exposure is equivalent when ciprofloxacin is administered alone and with raxibacumab. Thus, there is no significant interaction between raxibacumab and ciprofloxacin.
- The exposure-response data suggest a relationship between raxibacumab dose, concentration, and survival. The dose and dosing regimen selected by the applicant, raxibacumab 40 mg/kg IV single dose, is consistent with the known relationship between dose-concentration-response.
- Dose and concentration response for the endpoint of survival indicates potential to achieve higher response rates at higher doses. If future trials are conducted, higher doses (e.g. 60 or 80 mg/kg) should be explored along with 40 mg/kg.
- Based on in vitro binding kinetics studies, the proposed dose of 40 mg/kg raxibacumab in humans would be expected to maintain levels required for virtually complete binding (99.9%) of PA for 7 days and those required for 99% binding for up to 42 days. Despite the theoretical importance of targeting virtually complete binding of PA, the duration of time raxibacumab concentrations remain above the threshold of 202 μg/mL (99.9% PA binding) does not appear to impact efficacy in inhalational anthrax models.
- Based on predicted human PK profiles, more than 95% of humans administered a 40 mg/kg IV raxibacumab dose can be expected to achieve serum raxibacumab concentrations that are

equimolar to or in excess of the highest observed serum/plasma PA concentration in any animal that died in the therapeutic intervention studies (for 48 and 28 days relative to rabbits and monkeys, respectively). In addition, modeling and simulation indicated that a single 40 mg/kg IV raxibacumab dose in humans is adequate to bind at least 99.7% of serum PA for up to 28 days after administration, in at least 95% of patients who receive raxibacumab.

- In the pivotal animal studies, animals that were treated with raxibacumab and died exhibited a higher incidence and severity of histopathologic findings in the brain versus placebo treated animals. No clear relationship between exposure and CNS pathology can be discerned based on the limited amount of data in animals that died. Overall, the raxibacumab 20 mg/kg dose group had a higher incidence and severity of CNS findings than the 40 mg/kg group in both rabbits and monkeys. Due to the small numbers of animals that exhibited specific pathology findings, no definitive conclusions about exposure-response for bacteria, hemorrhage, inflammation, or necrosis could be made.
- Raxibacumab was non-immunogenic in humans and there were no subjects with a positive anti-raxibacumab antibody response in the human studies following single or repeat (every 14 days or following $\geq$ 4 months) doses.

10/19/09

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2. QUESTION BASED REVIEW

2.1. General Attributes of the Drug

2.1.1. What are the highlights of the chemistry and physical-chemical properties of the drug substance and the formulation of the drug product?

Raxibacumab is also known as HGS1021, PA mAb, or ABthrax™. Raxibacumab is a recombinant, fully human, IgG1λ monoclonal antibody that binds PA with high affinity and inhibits its biological activity. Raxibacumab is expressed in the NS0 mouse myeloma cell line, secreted into culture media, and purified by a series of chromatography and filtration steps. Raxibacumab is a fully human IgG1 antibody comprising 2 identical light chains and 2 identical heavy chains, (b) (4) (b) (4)

Structural Formula: (b) (4)

The molecular formula represents the amino acid sequence of raxibacumab (b) (4) (b) (4)

Chemical Structure: The completely verified amino acid sequences of the heavy and light chain of raxibacumab are shown in Figure 2.1.1-1. (b) (4) (b) (4)

Figure 2.1.1-1 Amino Acid Sequences of the Light and Heavy Chain of Raxibacumab*

Heavy Chain

(b) (4)

Light Chain

(b) (4)

* (b) (4).

Source: Section 2.3.S

Chemical Name: Immunoglobulin G1, anti-(anthrax protective antigen) (human monoclonal PA heavy chain), disulfide with human monoclonal PA λ-chain, dimer

United States Adopted Name (USAN): Raxibacumab

Company Laboratory Code: HGS1021, PA mAb, or ABthrax™

Chemical Abstract Service (CAS) Registry Number: 565451-13-0

Molecular Weight: 145.9 kDa

Raxibacumab is supplied in 50 mL sterile, single-use vials containing a minimum of 35.1 mL of liquid formulation per vial. Each vial contains 50 mg/mL raxibacumab in 0.13 mg/mL citric acid, 2.8 mg/mL sodium citrate, 10 mg/mL sucrose, 18 mg/mL glycine, 0.2 mg/mL polysorbate 80, pH 6.5. Raxibacumab should be stored at 2-8°C and is stable for up to 8 hours at room temperature. The calculated raxibacumab dose (40 mg/kg) to be administered to the subject will be diluted in normal saline with a final volume of 250 mL. The rate of raxibacumab infusion should be set for 15 mL/hour for the first 20 minutes and then adjusted to 125 mL/hour for the remainder of the infusion period. With this schedule, approximately 250 mL should be infused over the course of approximately 2 hours and 15 minutes.

The qualitative and quantitative composition of the proposed raxibacumab biologic drug product is shown in Table 2.1.1-1.

Table 2.1.1-1 Composition of Raxibacumab Biologic Drug Product

Component	Concentration (mg/mL)	Amount Per Vial (mg/vial)[1]	Function	Grade
Raxibacumab	50	1700	API	HGS Specification
Citric acid (b) (4)	0.13	(b) (4)		Multicompendial[2]
Sodium citrate (b) (4)	2.8			Multicompendial[2]
Sucrose	10			Multicompendial[3]
Glycine	18			Multicompendial[2]
Polysorbate 80 (b) (4)	0.2 (b) (4)			Multicompendial[3] USP

[1] Amount listed is deliverable amount.

[2] According to supplier's definition, multicompendial grade includes full compendial testing as appropriate to USP or National Formulary (NF), European Pharmacopoeia (EP), British Pharmacopoeia (BP), and Japanese Pharmacopoeia (JP).

[3] According to supplier's definition, multicompendial grade includes full compendial testing as appropriate to USP/NF, EP, and JP.

Source: Section 2.3.P.1.2

During manufacturing process development, the M10 (b) (4) development process was originally used to manufacture biologic drug substance (BDS) in 2003 for nonclinical and Phase 1 clinical studies. Process development continued for raxibacumab production (b) (4) (b) (4) were used during further (b) (4) development of raxibacumab (M11 (b) (4) process). For the animal efficacy

and Phase 2/3 human safety studies, material from (b) (4) BDS manufacturing process (M11) was used. This process is also used for preparation of the commercial raxibacumab drug product. As a comparison of exposure across species forms the basis of extrapolation of efficacy findings from animals to humans, only the human study using investigational product manufactured by the same process (designated M11) as the product used in the pivotal animal studies will be used for purposes of comparison between species (Clinical Study HGS1021-C1064).

2.1.2. What is the proposed mechanism of drug action and therapeutic indication?

Raxibacumab is a recombinant, fully human, IgG1λ monoclonal antibody that binds PA with high affinity and inhibits its biological activity. Thus, the principal mechanism of action is to neutralize the effects of anthrax toxins.

Raxibacumab is proposed for the treatment (b) (4) (b) (4)

2.1.3. What is the proposed dosage and route of administration?

The proposed dosage and route of administration of raxibacumab is a single dose of 40 mg/kg administered as an intravenous (IV) infusion after dilution with normal saline (~250 mL) over approximately 2 hours. Raxibacumab is proposed to be administered alone or in combination with antibiotics.

Patients should be premedicated with oral diphenhydramine (25-50 mg) within one hour prior to the infusion of raxibacumab, due to the possibility of infusion-related and hypersensitivity reactions.

2.2. General Clinical Pharmacology

2.2.1. What are the design features of the clinical pharmacology and clinical studies used to support dosing or claims?

To allow selection of an effective dose in humans for therapeutic treatment of anthrax, the applicant has submitted clinical pharmacology data for raxibacumab in humans and in the two pivotal animal species, rabbits and non-human primates. The pharmacokinetics (PK) and safety of raxibacumab administered as the product proposed for licensure has been evaluated in a Phase 1 antibiotic interaction study with ciprofloxacin (HGS1021-C1064), a repeat dose immunogenicity study (HGS1021-C1069) and a Phase 2/3 safety study (HGS1021-C1063).

In accordance with the Animal Rule and because evaluation of new treatment options for inhalational anthrax is not possible in controlled clinical trials in humans for ethical reasons, the efficacy of raxibacumab was evaluated in two animal species, specifically New Zealand white rabbits and cynomolgus monkeys, with symptomatic anthrax disease (Studies Study 724-G005829 and 682-G005758). These pivotal efficacy studies were placebo-controlled, parallel-group randomized studies powered to provide a statistically significant result if a clinically important increase in survival was observed. Raxibacumab doses evaluated in both pivotal animal model efficacy studies were 20 and 40 mg/kg single doses administered intravenously. Because the current standard therapy for inhalation anthrax is post-exposure prophylaxis (PEP) with antimicrobials, the efficacy of antibiotics administered concomitantly with raxibacumab in the setting of therapeutic treatment also was evaluated in rabbits and monkeys with symptomatic anthrax disease (Studies 781-G923701 and 789-G923702).

2.2.2. What is the basis for selecting the response endpoints (i.e., clinical or surrogate endpoints) or biomarkers (collectively called pharmacodynamics [PD]) and how are they measured in clinical pharmacology and clinical studies?

The primary endpoint of the animal efficacy studies is survival, which is the desired benefit in humans. Survival time has also been evaluated as a secondary endpoint.

2.2.3. Are the active moieties in the biological fluid appropriately identified and measured to assess pharmacokinetic parameters?

The active moiety raxibacumab was appropriately identified and measured in serum from rabbits, monkeys and humans by a validated electrochemiluminescence assay.

2.2.4. Exposure-Response

2.2.4.1. What are the characteristics for exposure-response relationships (dose-response, concentration-response) for efficacy?

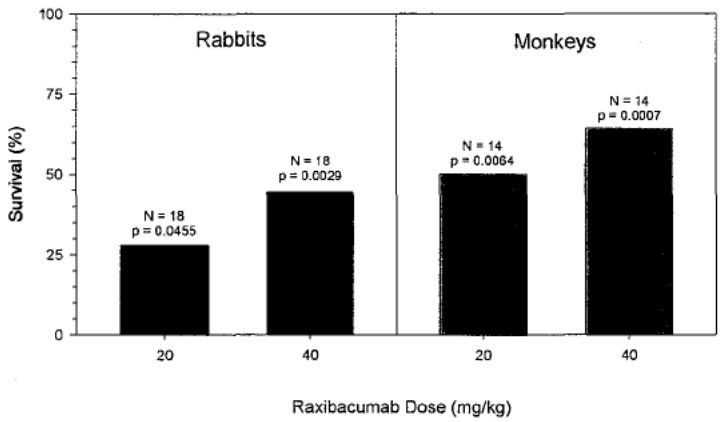
The exposure-response relationship for raxibacumab has been evaluated in animal models of inhalation anthrax infection in the following studies:

- Study 724-G005829: an animal efficacy study evaluating raxibacumab efficacy as therapeutic treatment against inhalation anthrax in the monkey model; this study included an evaluation of the PK of raxibacumab and the kinetics of *Bacillus anthracis* PA following a single IV raxibacumab dose in monkeys with inhalation anthrax (Pivotal Monkey Efficacy Study).
- Study 682-G005758: an animal efficacy study evaluating raxibacumab efficacy as therapeutic treatment against inhalation anthrax in the rabbit model; this study included an evaluation of the PK of raxibacumab and the kinetics of *Bacillus anthracis* protective antigen (PA) following a single IV raxibacumab dose in rabbits with inhalation anthrax (Pivotal Rabbit Efficacy Study).

Exposure-Survival Relationship

Survival rates by raxibacumab dose in the pivotal animal efficacy studies are summarized in Figure 2.2.4.1-1 and survival curves are presented in Figure 2.2.4.1-2. Survival rates in both studies exhibited a dose-response.

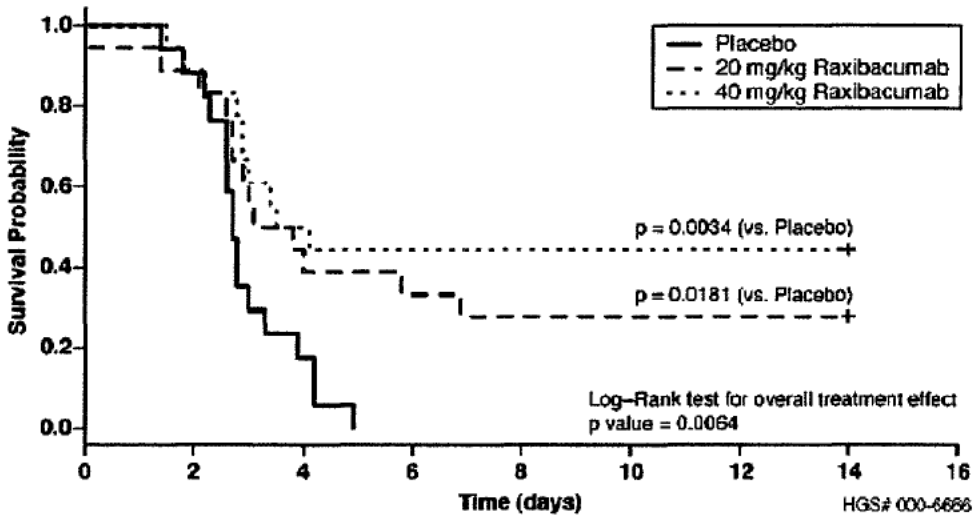
Figure 2.2.4.1-1 Percent Survival in Rabbits and Monkeys Treated with Raxibacumab for Anthrax



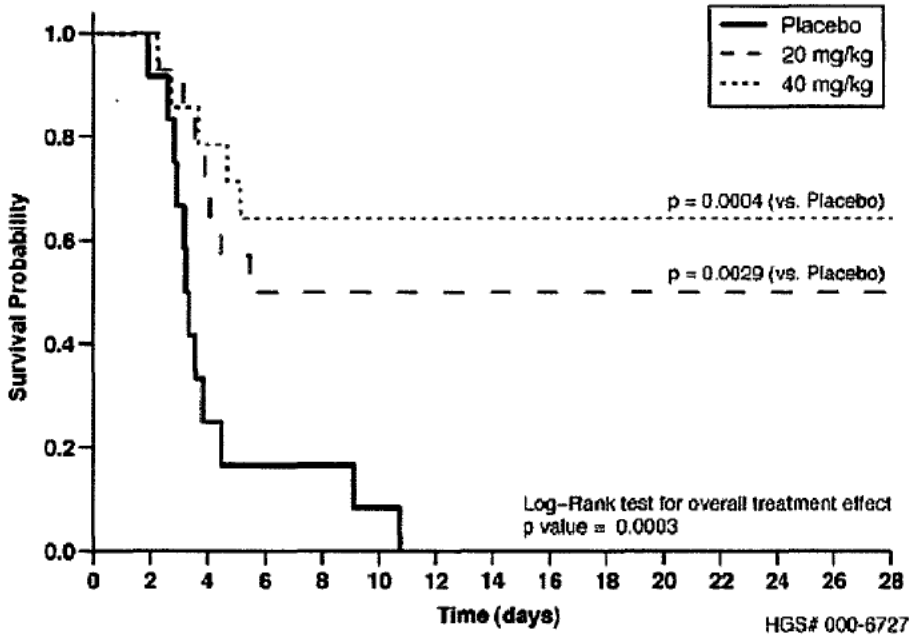
P-values from Fisher's exact test of the intent to treat population.

Figure 2.2.4.1-2 Survival Curves for Pivotal Animal Studies 724-G005829 and 682-G005758

Rabbits:



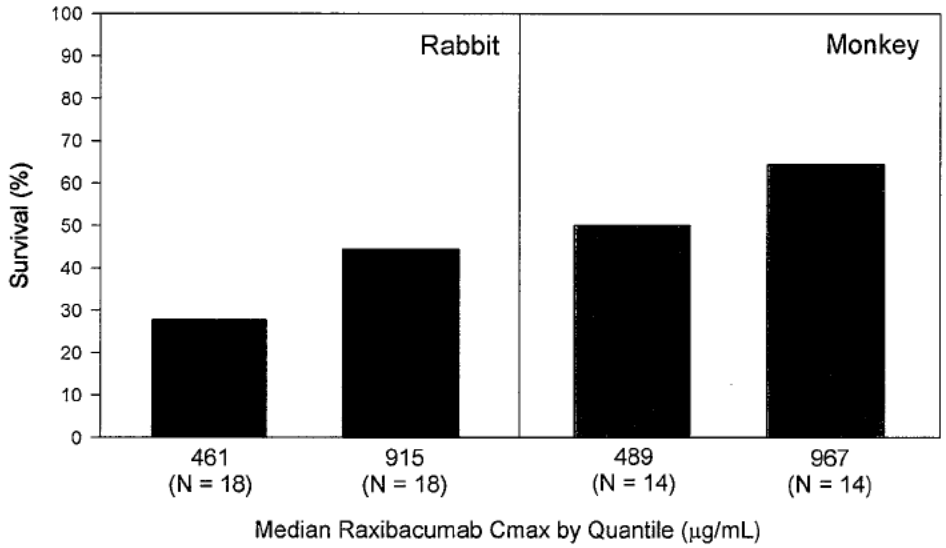
Monkeys:



P-values from log rank test.

Mirroring the dose-response seen with 20 and 40 mg/kg doses of raxibacumab, a concentration-response is seen between the probability of survival and quantiles of raxibacumab Cmax, as presented in Figure 2.2.4.1-3. The data suggest a relationship between raxibacumab dose, concentration, and survival.

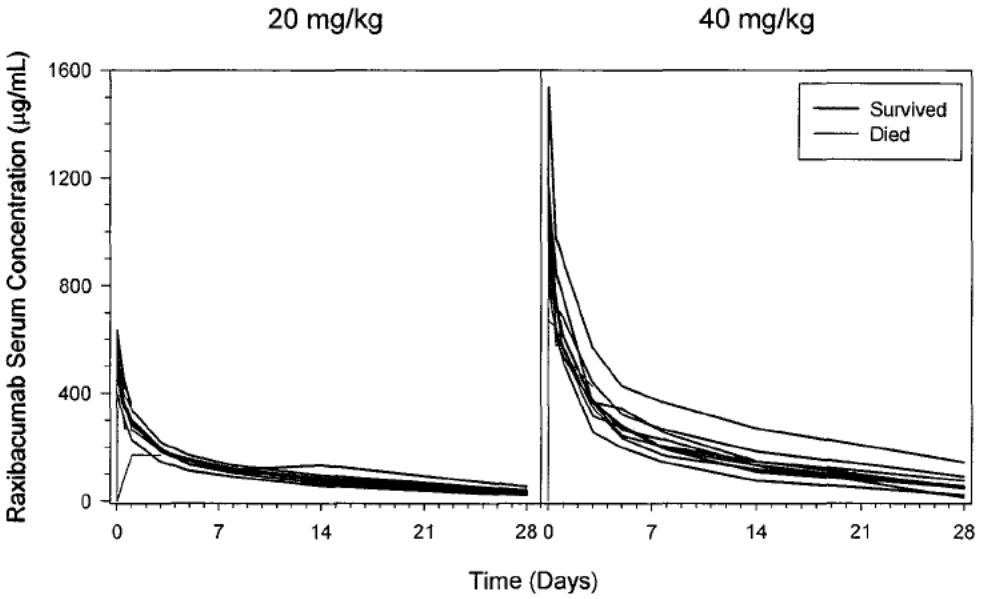
Figure 2.2.4.1-3 Comparison of Probabilities of Survival by Raxibacumab Cmax Quantile



A comparison of raxibacumab serum concentration between animals that survived and animals that died following *Bacillus anthracis* spore challenge and treatment with single intravenous administration of 20 mg/kg or 40 mg/kg raxibacumab in cynomolgus monkeys and rabbits is displayed in Figure 2.2.3.1-4. A comparison of raxibacumab Cmax and AUCinf between animals that survived and animals that died following *Bacillus anthracis* spore challenge and treatment with single intravenous administration of 20 mg/kg or 40 mg/kg raxibacumab in cynomolgus monkeys and rabbits is displayed in Figure 2.2.3.1-5. No distinct differences in raxibacumab serum concentrations, Cmax, or AUCinf were observed between animals that died and animals that survived.

Figure 2.2.3.1-4. Comparison of Raxibacumab Serum Concentrations Between Animals that Survived and Animals that Died Following *Bacillus anthracis* Spore Challenge and Treatment with Single Intravenous Administration of 20 mg/kg or 40 mg/kg Raxibacumab

Graph A: Monkeys



Graph B: Rabbits

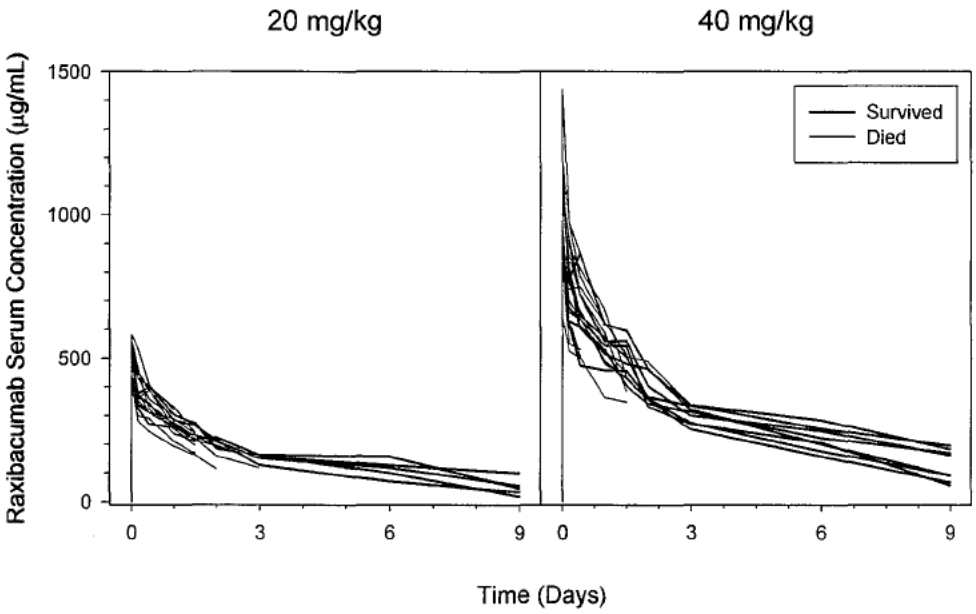
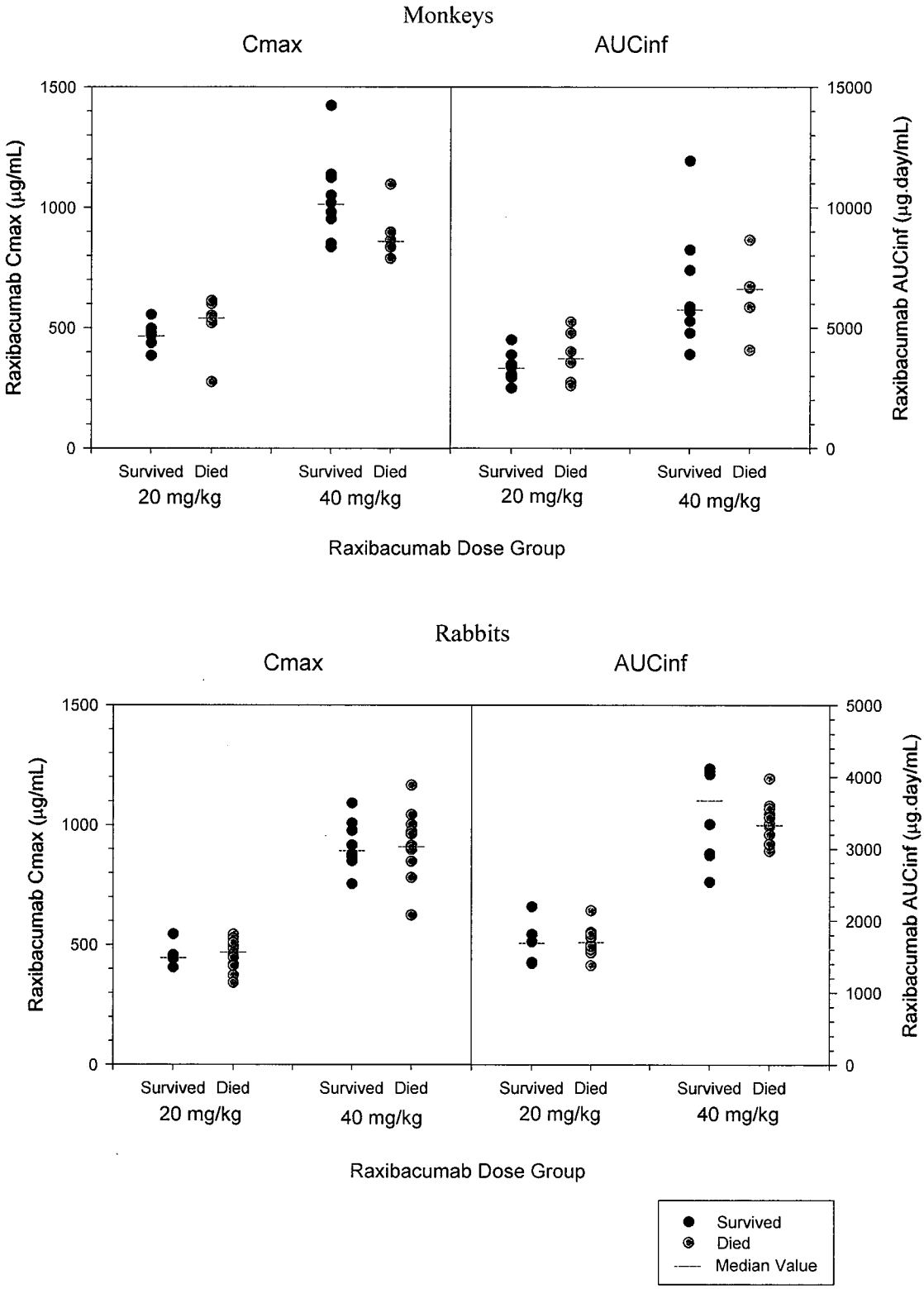


Figure 2.2.3.1-5. Comparison of Raxibacumab Cmax and AUCinf Between Animals that Survived and Animals that Died Following *Bacillus anthracis* Spore Challenge and Treatment with Single Intravenous Administration of 20 mg/kg or 40 mg/kg Raxibacumab

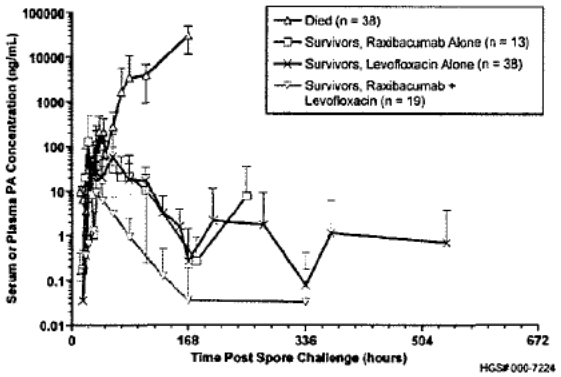


Exposure-PA Relationship

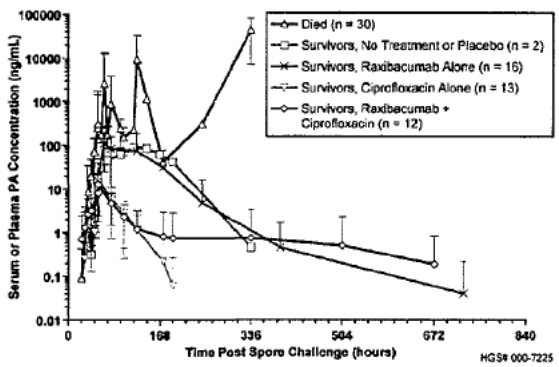
The following two characteristics of an "effective human raxibacumab dose" were proposed by the applicant: 1) a sufficiently high Cmax, and 2) at least 28 days duration of protective serum raxibacumab levels. This was based on a comparison of serum/plasma PA concentration-time profiles for *B. anthracis* inhalation spore-challenged animals that were untreated and died or were administered placebo and died verus animals that survived, as displayed in Figure 2.2.3.1-6. In surviving animals, PA concentrations reach a peak, but then decrease, whereas in animals that died, PA levels continue to increase until death occurs. In addition, the peak PA levels in animals that died are orders of magnitude higher than the peak levels in the animals that survived. As proposed by the applicant, the data suggest that an efficacious human raxibacumab dose needs to be large enough to rapidly attain serum levels equal to or in excess of those needed to bind systemic PA concentrations, to prevent the mortality associated with PA toxemia. In other words, ***a human raxibacumab dose should attain a sufficiently high Cmax to optimize the likelihood of achieving efficacy.***

Figure 2.2.3.1-6 Serum/Plasma PA Concentration Profiles in Rabbits and Monkeys

Rabbits



Monkeys



In the rabbit and monkey efficacy studies, animals that survived generally developed measurable anti-PA Ab concentrations and toxin neutralizing Ab (TNA) titers by 14 to 28 days post spore challenge. Thus, the applicant proposes an efficacious human raxibacumab dose ***should maintain protective systemic raxibacumab exposure for at least one month after administration, to allow the innate immune response to PA to develop.***

These two characteristics for the single 40 mg/kg IV infusion raxibacumab dose in humans were assessed by determining the proportion of human subjects for whom raxibacumab exposure can be considered as protective for survival against inhalation anthrax. Comparisons of raxibacumab PK between humans, rabbits and monkeys are presented in Tables 2.2.3.1-3 and 2.2.3.1-4. Cmax was similar for a 40 mg/kg dose across species. Given the raxibacumab mechanism of action (binding PA to block lethal effects of toxemia) it is reasonable to expect that attaining similar Cmax should result in similar efficacy in the therapeutic intervention setting. The lowest Cmax and AUC0-∞ in a human subject were 589 µg/mL and 8720 µg·day/mL, respectively, while the lowest observed Cmax and AUC0-∞ for surviving spore-challenged rabbits were 405 µg/mL and 1411 µg·day/mL, respectively, and were 385 µg/mL and 2499 µg·day/mL, respectively, for surviving spore-challenged monkeys. Thus, a 40 mg/kg dose to humans can be expected to provide exposure associated with survival for virtually all subjects.

Table 2.2.3.1-3 Raxibacumab PK in Rabbits and in Healthy Humans (Mean ± SD)

	Healthy Rabbits AB50409.INF.0.016		Spore-Challenged Rabbits That Survived Report 682-G005758		Report 781-G923701	Healthy Humans Population Analysis	
	1 mg/kg (n = 4)	10 mg/kg (n = 4)	20 mg/kg[1] (n = 5)	40 mg/kg[1] (n = 8)	40 mg/kg[2] (n = 18)	40 mg/kg[1] (n = 322)	20 mg/kg[3]
C_{max} (µg/mL)	26 ± 1	276 ± 19	460 ± 51	918 ± 105	929 ± 106	960 ± 164	480 ± 82
$AUC_{0-\infty}$ (µg·day/mL)	174 ± 60	1518 ± 408	1711 ± 323	3504 ± 647	4439 ± 856	16667 ± 3198	8334 ± 1599
$t_{1/2,\alpha}$ (h)	0.40 ± 0.22	0.24 ± 0.05	0.23 ± 0.04	0.26 ± 0.05	0.09 ± 0.01	1.76 ± 0.36	1.76 ± 0.36
$t_{1/2,\beta}$ (h)	8.7 ± 4.4	6.9 ± 2.7	3.86 ± 1.11	4.15 ± 1.22	4.58 ± 0.70	22.35 ± 4.04	22.35 ± 4.04
MRT (h)	12.0 ± 6.0	9.7 ± 3.7	5.41 ± 1.54	5.79 ± 1.69	6.55 ± 0.99	30.09 ± 5.76	30.09 ± 5.76
CL (mL/kg/day)	6.2 ± 1.8	6.9 ± 1.8	11.96 ± 2.36	11.60 ± 2.42	9.35 ± 1.91	2.49 ± 0.49	2.49 ± 0.49
CLD_2 (mL/kg/day)	NA	NA	37.18 ± 1.31	36.73 ± 2.12	86.38 ± 6.74	6.56 ± 0.91	6.56 ± 0.91
V_1 (mL/kg)	39.0 ± 2.1	36.4 ± 2.5	43.56 ± 4.21	43.22 ± 4.53	43.62 ± 5.49	42.86 ± 7.28	42.86 ± 7.28
V_2 (mL/kg)	NA	NA	18.34 ± 4.32	20.70 ± 4.55	16.11 ± 3.16	30.06 ± 4.34	30.06 ± 4.34
V_{ss} (mL/kg)	66.9 ± 9.9	63.2 ± 15.4	61.90 ± 6.45	63.92 ± 7.54	59.73 ± 6.34	72.92 ± 10.07	72.92 ± 10.07

Abbreviations: C_{max}, maximum serum drug concentration; $AUC_{0-\infty}$, area under the serum drug concentration-time curve from time 0 to infinite time; $t_{1/2,\alpha}$, elimination half-life for the 1st phase; $t_{1/2,\beta}$, elimination half-life for the 2nd (terminal) phase; MRT, mean residence time; CL, clearance; CLD_2, intercompartmental clearance; V_1, volume of distribution for the central compartment; V_2, volume of distribution for the peripheral compartment; V_{ss}, volume of distribution at steady-state; NA, not available.

[1] Based on individual post hoc estimates.

[2] Based on individual post hoc estimates; raxibacumab was administered with levofloxacin.

[3] Extrapolated values, assuming linear PK.

Table 2.2.3.1-4 Raxibacumab PK in Monkeys and in Healthy Humans (Mean ± SD)

	Healthy Monkeys AB50409.INF.0.017		Spore-Challenged Monkeys That Survived Report 724-G005829		Report 789-G923702	Healthy Humans Population Analysis	
	1 mg/kg (n = 4)	10 mg/kg (n = 4)	20 mg/kg[1] (n = 7)	40 mg/kg[1] (n = 9)	40 mg/kg[2] (n = 12)	40 mg/kg[1] (n = 322)	20 mg/kg[3]
C_{max} (µg/mL)	29 ± 6	262 ± 30	475 ± 50	1042 ± 177	1067 ± 158	960 ± 164	480 ± 82
$AUC_{0-\infty}$ (µg·day/mL)	267 ± 91	2030 ± 172	3379 ± 655	6544 ± 2400	9903 ± 2279	16667 ± 3198	8334 ± 1599
$t_{1/2,\alpha}$ (h)	1.10 ± 0.84	0.69 ± 0.53	0.69 ± 0.10	0.68 ± 0.14	0.64 ± 0.12	1.76 ± 0.36	1.76 ± 0.36
$t_{1/2,\beta}$ (h)	15.8 ± 4.1	11.8 ± 1.9	10.80 ± 1.79	9.95 ± 2.48	15.27 ± 4.53	22.35 ± 4.04	22.35 ± 4.04
MRT (h)	19.8 ± 4.3	15.8 ± 1.7	14.38 ± 2.64	13.06 ± 3.53	20.78 ± 6.33	30.09 ± 5.76	30.09 ± 5.76
CL (mL/kg/day)	4.1 ± 1.4	5.0 ± 0.4	6.09 ± 1.15	6.64 ± 2.00	4.25 ± 1.05	2.49 ± 0.49	2.49 ± 0.49
CLD_2 (mL/kg/day)	NA	NA	19.95 ± 3.11	19.03 ± 2.97	21.34 ± 2.98	6.56 ± 0.91	6.56 ± 0.91
V_1 (mL/kg)	36.0 ± 7.8	38.6 ± 4.3	42.41 ± 4.81	38.80 ± 6.05	38.18 ± 5.14	42.86 ± 7.28	42.86 ± 7.28
V_2 (mL/kg)	NA	NA	43.04 ± 2.60	42.07 ± 5.83	44.88 ± 7.58	30.06 ± 4.34	30.06 ± 4.34
V_{ss} (mL/kg)	78.8 ± 23.7	78.0 ± 8.3	85.45 ± 6.90	80.87 ± 10.40	83.06 ± 10.08	72.92 ± 10.07	72.92 ± 10.07

Abbreviations: C_{max}, maximum serum drug concentration; $AUC_{0-\infty}$, area under the serum drug concentration-time curve from time 0 to infinite time; $t_{1/2,\alpha}$, elimination half-life for the 1st phase; $t_{1/2,\beta}$, elimination half-life for the 2nd (terminal) phase; MRT, mean residence time; CL, clearance; CLD_2, intercompartmental clearance; V_1, volume of distribution for the central compartment; V_2, volume of distribution for the peripheral compartment; V_{ss}, volume of distribution at steady-state; NA, not available.

[1] Based on individual post hoc estimates.

[2] Based on individual post hoc estimates; raxibacumab was administered with ciprofloxacin.

[3] Extrapolated values, assuming linear PK.

Based on the median human serum raxibacumab concentration-time profile and lower 90% prediction interval bound profile for a single 40 mg/kg IV raxibacumab dose, the percentage of PA that could be bound were calculated and are summarized in Table 2.2.3.1-5. A single IV 40 mg/kg raxibacumab dose in humans can be expected to produce serum drug concentrations high enough to bind at least 99% of serum PA for at least 42 days post-dose in at least 95% of the subjects. Greater than 99% of PA would be bound following a 20 mg/kg dose; however, that level of binding would only be maintained for 28 days.

Table 2.2.3.1-5 Serum Raxibacumab Concentrations in Humans and PA Binding

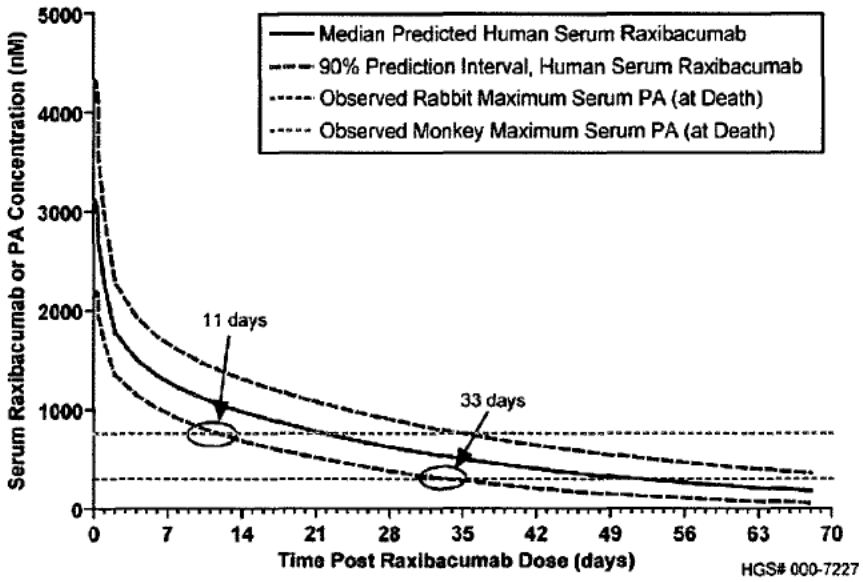
	Serum Raxibacumab Concentrations at:											
	20 mg/kg						40 mg/kg					
	Median			Lower 90% PI			Median			Lower 90% PI		
Time (Days)	µg/mL	nM	% Bound for PA	µg/mL	nM	% Bound for PA	µg/mL	nM	% Bound for PA	µg/mL	nM	% Bound for PA
End of Infusion	452	3115	99.9	317	2185	99.9	903	6230	100.0	634	4370	99.9
1	326	2251	99.9	243	1672	99.9	653	4502	99.9	485	3345	99.9
14	142	978	99.7	99	683	99.6	283	1955	99.9	198	1367	99.8
28	90	623	99.6	55	380	99.4	181	1246	99.8	110	759	99.7
42	58	403	99.4	30	203	98.8	117	806	99.7	59	407	99.4
56	38	260	99.1	15	104	97.7	75	520	99.5	30	209	98.8

Note: This table also includes the percentage of PA that would be bound for a single 20 mg/kg dose based on extrapolation from the 40 mg/kg dose results.

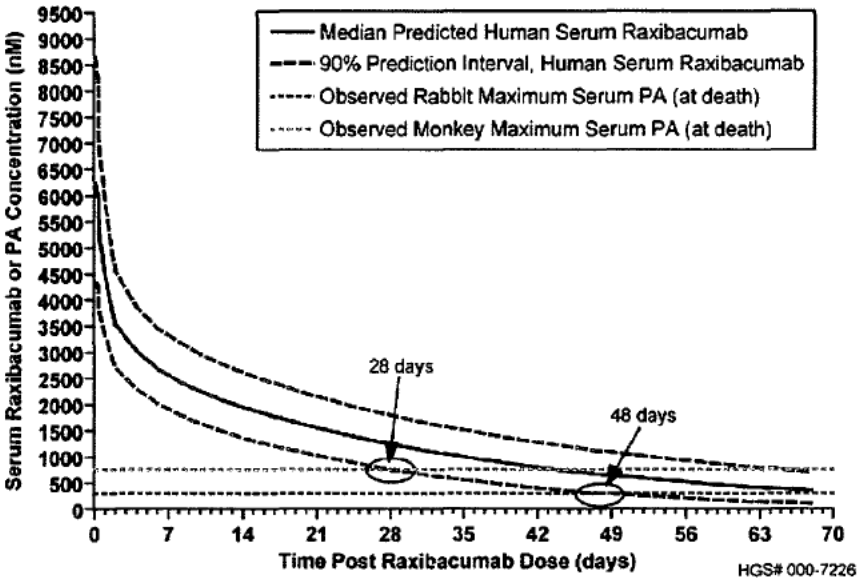
The highest observed serum/plasma PA concentrations observed prior to death in control rabbits and monkeys (ie, not administered raxibacumab or antibiotic) that died were 24752 and 63096 ng/mL (298 and 760 nM), respectively. The median and 90% prediction interval serum raxibacumab profiles for a 20 and 40 mg/kg single IV dose (expressed as nM concentrations) overlaid with the expected highest PA concentrations to be encountered (expressed as nM) is presented in Figure 2.2.3.1-7. Following a 40 mg/kg dose, serum raxibacumab levels are equimolar to or greater than the highest expected PA levels for 28 or 48 days, using PA levels from monkeys and rabbits, respectively. In contrast, following a 20 mg/kg dose, serum raxibacumab levels were equimolar to or greater than the highest expected PA levels for 11 to 33 days.

Figure 2.2.3.1-7 Median and 90% Prediction Interval Serum Raxibacumab Profiles for a 20 and 40 mg/kg Single IV Dose Versus the Highest Expected PA Concentrations to be Encountered

20 mg/kg



40 mg/kg



A reasonable assessment of the inter-relationship between raxibacumab concentrations, PA concentrations and survival for purposes of predicting the acceptability of a proposed dose is a comparison of raxibacumab serum concentrations over time to the concentrations required for 99.0 and

99.9% binding of PA (based on the known mechanism of action and binding kinetics of raxibacumab). Based on in vitro binding kinetics studies, serum raxibacumab concentrations of approximately 40 and 202 µg/mL are required for 99.0 and 99.9% binding of PA, respectively. As presented in Figure 2.2.3.1-8, in human subjects that received 40 mg/kg raxibacumab IV, raxibacumab concentrations remained above 202 µg/mL for 7 days and above 40 µg/mL for 42 days for all human subjects. Thus, in humans a 40 mg/kg dose of raxibacumab would be expected to maintain levels required for virtually complete binding of PA for 7 days. But, despite the theoretical importance of targeting virtually complete binding of PA, the duration of time raxibacumab concentrations remain above the threshold of 202 µg/mL does not appear to impact efficacy, as displayed in Figures 2.2.3.1-9 and 2.2.3.1-10. In both rabbits and monkeys, the amount of time serum concentrations of raxibacumab remained above 202 µg/mL generally did not differ between survivors and non-survivors.

Figure 2.2.3.1-8 Individual Serum Concentrations of Raxibacumab in Human Subjects Following Administration of 40 mg/kg IV Compared to the Concentrations Required for 99 and 99.9% Binding of PA

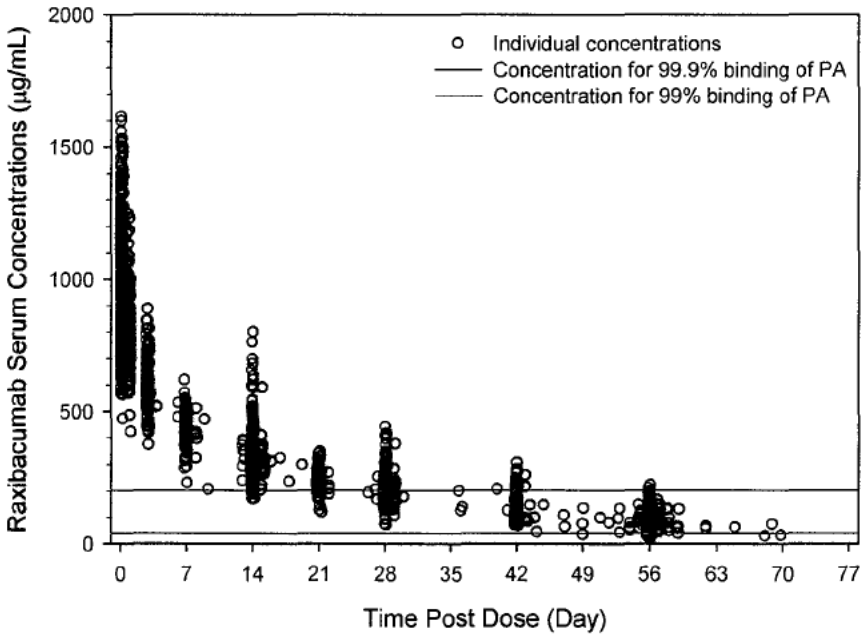


Figure 2.2.3.1-9 Spaghetti Plot of Serum Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Rabbits

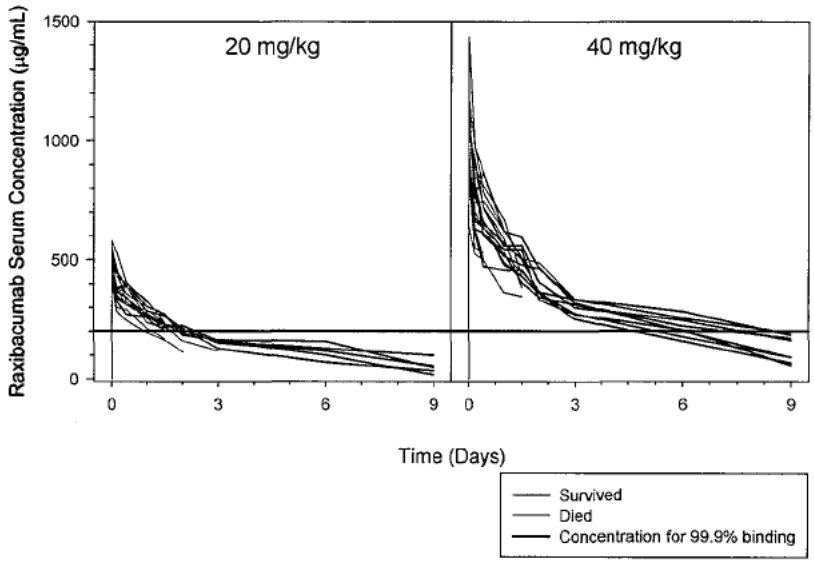
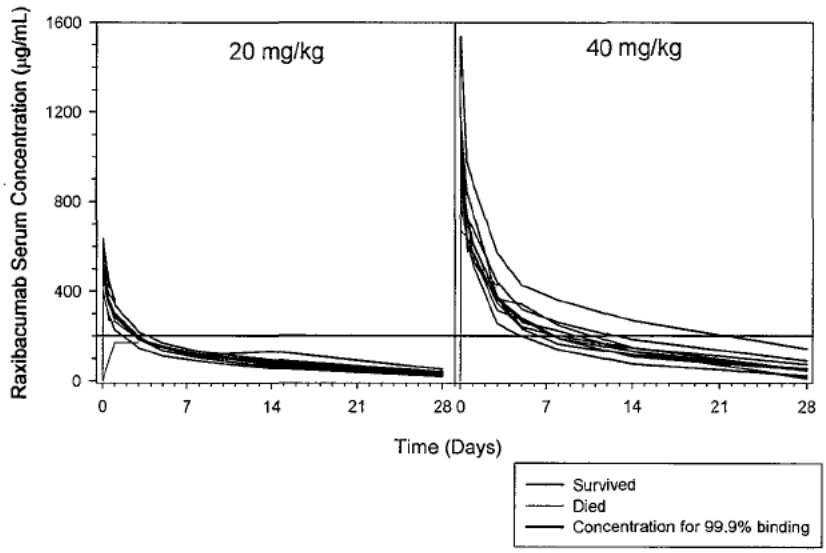


Figure 2.2.3.1-10 Spaghetti Plot of Serum Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Monkeys



In summary, an evaluation of the relationship between PA concentrations, raxibacumab concentrations, and outcome (survival or death) in monkeys and rabbits suggests the following:

1. The exposure-response data suggest a relationship between raxibacumab dose, concentration, and survival. Although the applicant proposes a sufficiently high Cmax optimizes the likelihood of efficacy, a threshold of Cmax was not identified.

2. Based on in vitro binding kinetics studies, the proposed dose of 40 mg/kg raxibacumab in humans would be expected to maintain levels required for virtually complete binding (99.9%) of PA for 7 days and those required for 99% binding for up to 42 days. Despite the theoretical importance of targeting virtually complete binding of PA, the duration of time raxibacumab concentrations remain above the threshold of 202 µg/mL does not appear to impact efficacy in inhalational anthrax models.
3. Based on predicted human PK profiles, more than 95% of humans administered a 40 mg/kg IV raxibacumab dose can be expected to have serum raxibacumab concentrations that are equimolar to or in excess of the highest observed serum/plasma PA concentration in any animal that died in the therapeutic intervention studies (for 48 and 28 days relative to rabbits and monkeys, respectively). In addition, a single 40 mg/kg IV raxibacumab dose in humans is adequate to bind at least 99.7% of serum PA for up to 28 days after administration, in at least 95% of in simulated humans.

2.2.4.2. What are the characteristics for exposure-response relationships (dose-response, concentration-response) for safety?

Adverse Events in Humans

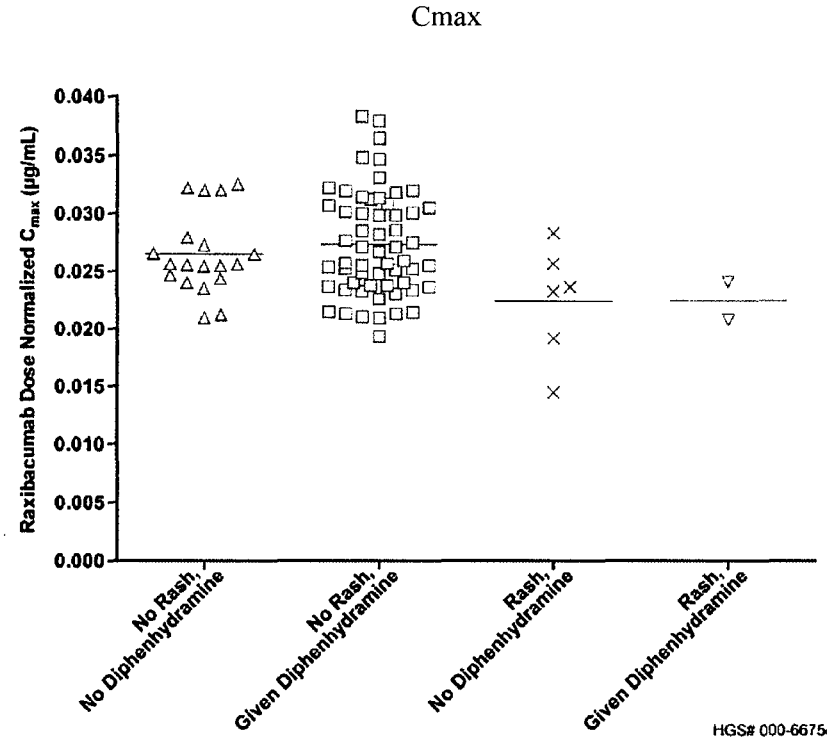
The safety of raxibacumab was evaluated in healthy volunteers in four clinical studies: a first-in-human placebo-controlled, dose escalation study (PAM-NH-01), a drug interaction study with ciprofloxacin (HGS1021-C1064), a repeat-dose study with the second raxibacumab dose administered ≥ four months after the first dose (HGS1021-C1069), and a placebo-controlled study evaluating single doses and two doses administered 14 days apart (HGS1021-C1063). The latter three studies used raxibacumab produced by the manufacturing process and with the formulation proposed for licensure. Incidence of adverse events events (AEs) in the placebo-controlled studies (PAM-NH-01 and HGS1021-C1064) were as follows: placebo (N = 99), 45.5%; raxibacumab 20 mg/kg (N = 73), 58.9%; and raxibacumab 40 mg/kg (N = 224), 49.1%. The incidence of related AEs across studies was 15%. The incidence of serious and of severe AEs across studies was low (0.6% and 1.8%, respectively). There was one possibly raxibacumab-related serious AE of cholecystitis in a single-dose subject from Study HGS1021-C1063. In addition, subjects in placebo-controlled studies who received two raxibacumab doses did not have a higher rate of AEs, related AEs, or SAEs than subjects who received a single dose. Overall, no discernable dose-response relationship for raxibacumab and incidence of adverse events was observed in the placebo-controlled trials.

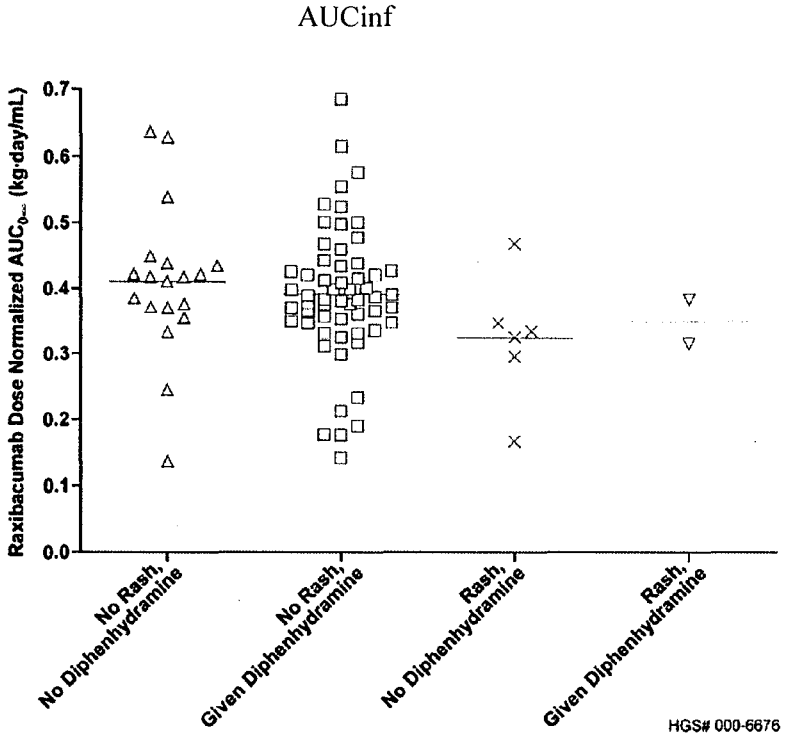
The incidence of AEs among subjects who received two raxibacumab doses (44.2%) was similar to that of the overall population of raxibacumab-treated subjects (49.7%). The incidence of raxibacumab-related AEs in double-dose cohorts was 7.0%. Incidences of at least one AE were similar among double-dose subjects in both of these studies. The incidence of at least one related AE was 5% in study HGS1021-C1069 compared with 8.7% in study HGS1021-C1063. In summary, incidences of AEs and related AEs were similar for subjects who received two raxibacumab doses ≥ four months apart compared with those who received two raxibacumab doses 14 days apart.

Nervous system disorders (primarily headache) and infections and infestations (primarily upper respiratory tract infection) were the most commonly observed AEs in single-dose raxibacumab-treated subjects (14.7% and 12.1%, respectively) and in single-dose placebo- treated subjects (16.2% and 17.2%, respectively) in placebo-controlled trials. Treatment with two raxibacumab doses was associated with comparable incidences of these most common AEs compared with single-dose treatment. In addition, the subjects treated with raxibacumab 40 mg/kg had comparable incidences of these most common AEs compared with the subjects treated with placebo.

Some subjects treated with raxibacumab experienced mild to moderate rashes in Study HGS1021-C1064 (N = 8/86 subjects; 9.1%). There were no rashes of Grade 3 or higher. All raxibacumab-related rashes resolved within 1 to 15 days despite the persistence of significant raxibacumab levels during this interval. To assess potential relationships between occurrence of rash and raxibacumab exposure, plots of Cmax/Dose or AUC0-∞/Dose were generated, as presented in Figure 2.2.4.2-1. Subjects were grouped according to whether they experienced a rash or not, and further subsetted according to whether or not they were administered diphenhydramine. Diphenhydramine was administered within 60 minutes prior to raxibacumab administration in 61/86 subjects. All treatment groups were combined for the assessment of rash versus raxibacumab exposure. Mean Cmax/Dose for subjects who experienced rash was lower (by approximately 17%) than those for the subjects who did not experience rash, while diphenhydramine use had little, if any, impact on Cmax/Dose; this difference was statistically significant ($p = 0.0334$), but none of the Bonferroni's multiple comparison post tests achieved significance ($p > 0.05$). Mean AUC0-∞/Dose for subjects who experienced rash tended to be lower (by approximately 11% to 21%) than those for the subjects who did not experience rash, while diphenhydramine use had no consistent impact on AUC0-∞/Dose; this difference was not statistically significant ($p = 0.3312$). Overall, raxibacumab exposure appears to be somewhat lower in the subjects who experienced rash, relative to those who did not. Raxibacumab exposure does not appear to differ between subjects who were administered diphenhydramine and those who were not.

Figure 2.2.4.2-1. Individual and Mean Raxibacumab Dose Normalized Cmax and AUCinf Following a Single 40 mg/kg Raxibacumab IV Infusion Dose, Administered with or without PO or IV Ciprofloxacin Coses, in Subjects Who Did or Did Not Experience Rash, and Who Were or Were Not Administered Diphenhydramine (Study HGS1021-C1064)





HGS1021-C1064 Pharmacokinetic Report, Section 3.6

CNS Findings in Animals (Clinical Pharmacology Reviewer Analyses)

In the pivotal animal studies in both rabbits and monkeys (Studies 682-G005758 and 724-G005829), animals that were treated with raxibacumab and died exhibited a higher incidence of histopathologic findings in the brain versus placebo treated animals, as presented in Figure 2.2.4.2-2. Histopathologic findings were also of higher severity in raxibacumab treated animals versus animals receiving placebo treatment, as presented in Figure 2.2.4.2-3. The raxibacumab 20 mg/kg dose group had a higher incidence and severity of CNS findings versus the 40 mg/kg group, suggesting an absence of clear dose-response relationship for brain histopathology.

Figure 2.2.4.2-2 Histopathology Findings by Treatment in Animals that Died

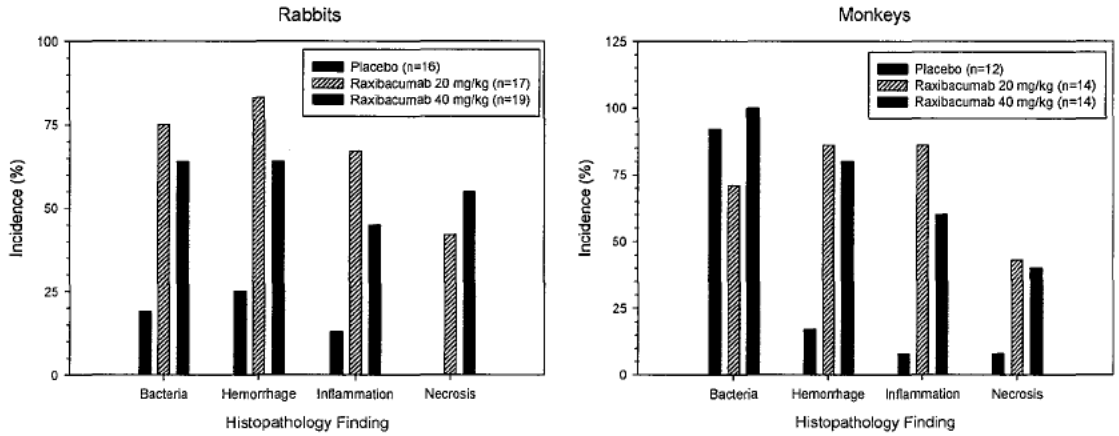
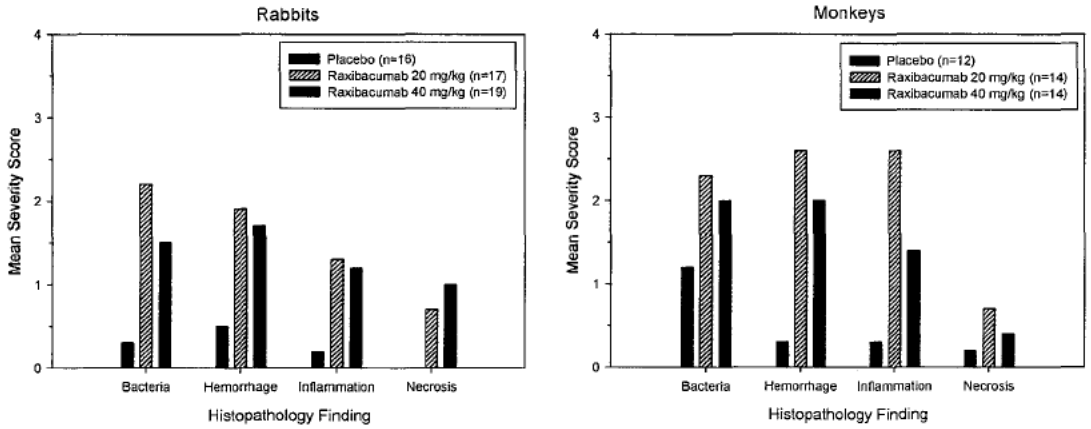


Figure 2.2.4.2-3 Severity of Histopathology Findings by Treatment in Animals that Died

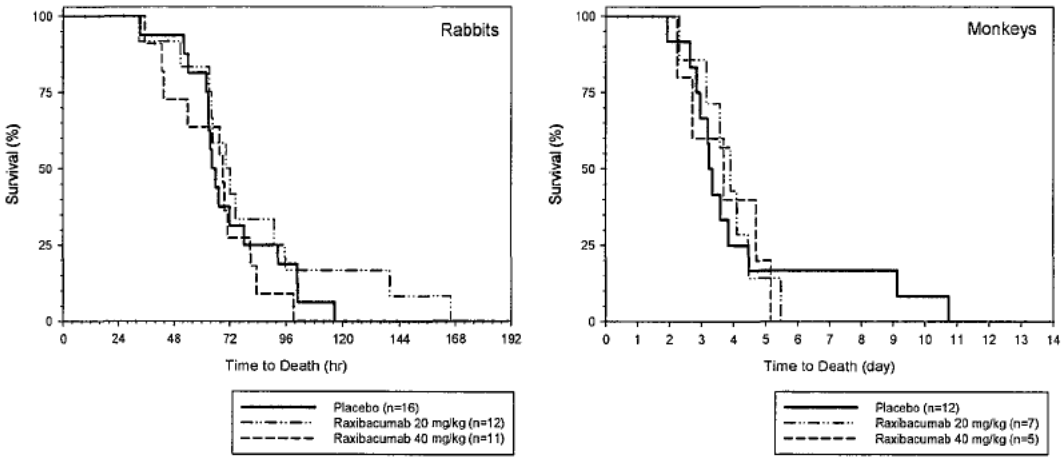


Grading scale is as follows: Grade 1, minimal representing the least detectable lesion; Grade 2, mild representing an easily discernable lesion unlikely to have biological relevance; Grade 3, moderate representing a change affecting a large area of the represented tissue that had the potential to be of some relevance; and Grade 4, marked representing a lesion that approached maximal.

A potential hypothesis was proposed by the applicant to explain why adverse CNS findings were more prevalent in animals that received raxibacumab and died versus animals that received placebo and died is that animals that received raxibacumab survived longer and brain lesions increased over time. Figure 2.2.4.2-3 shows percent survival and survival time by raxibacumab dose for rabbits and monkeys that died in the pivotal studies. The survival plots show that in rabbits that died, placebo animals have comparable survival times versus animals that received

40 mg/kg of raxibacumab. In monkeys that died, animals that received placebo lived longer than those receiving raxibacumab. Thus, the potential for raxibacumab treatment to result in adverse CNS findings is not likely to be related to prolonged survival time.

Figure 2.2.4.2-3 Time to Death in Rabbits and Monkeys That Died



To determine if in animals that died, higher raxibacumab exposure led to longer survival times, Figures 2.2.4.2-4 and 2.2.4.2-5 compare percent survival and survival time by quantiles of exposure (Cmax and AUC) for rabbits and monkeys that died, respectively. In rabbits that died, animals in the lower Cmax and AUC quantiles appeared to live longer than the other groups and placebo, but in monkeys there was no discernable difference between survival times for the higher and lower quantiles of exposure. The placebo group exhibited a longer survival time compared to the raxibacumab groups. This suggests that in animals that died, the magnitude of raxibacumab exposure did not affect survival time.

Figure 2.2.4.2-4 Comparison of Time to Death in Rabbits by Quantile of Raxibacumab Exposure

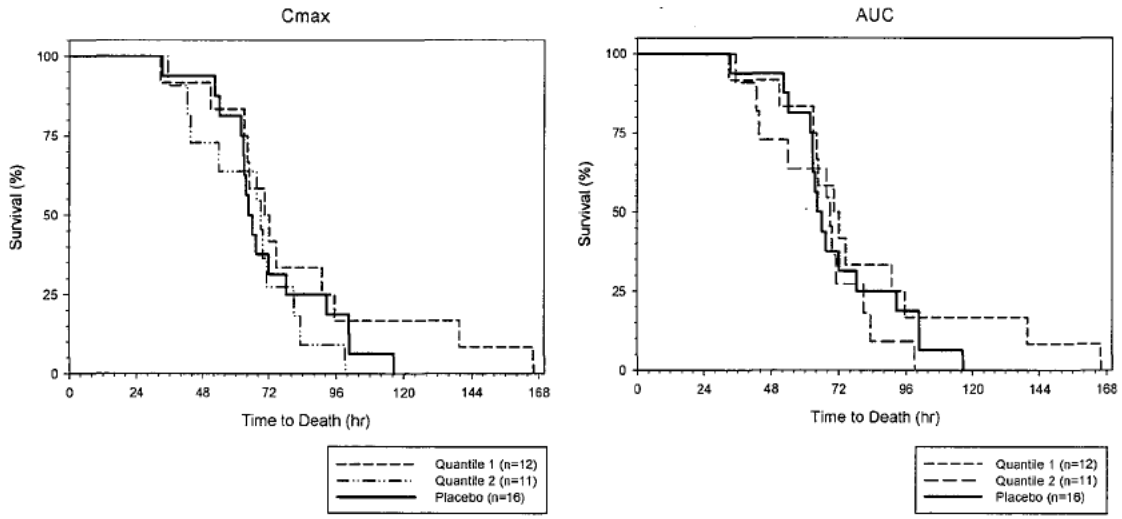
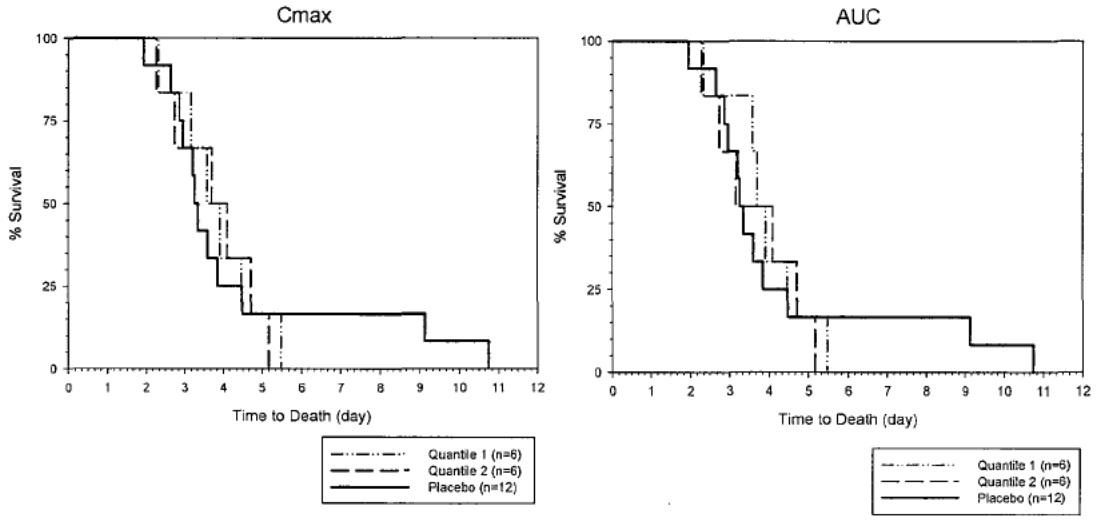
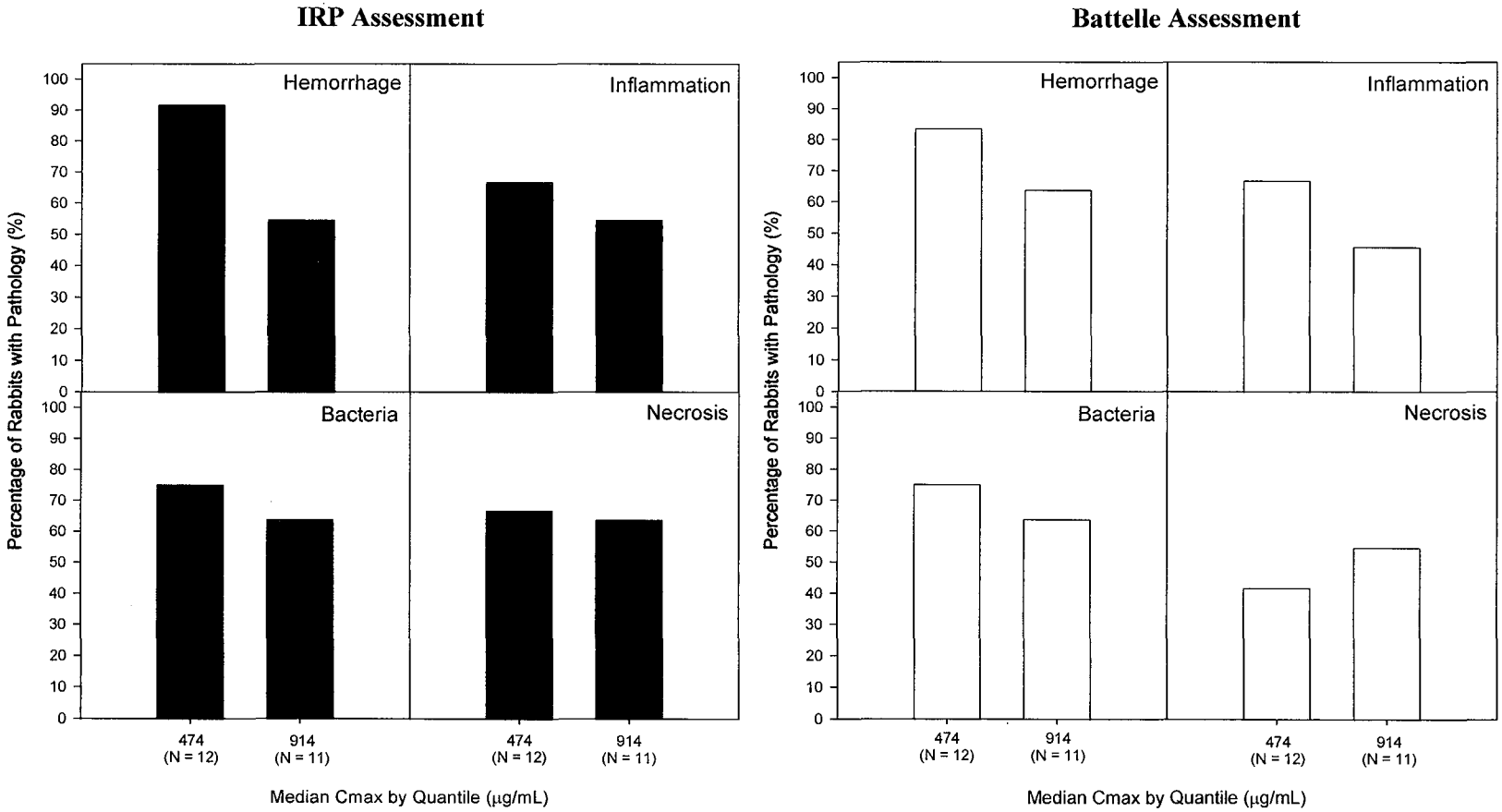


Figure 2.2.4.2-4 Comparison of Time to Death in Monkeys by Quantile of Raxibacumab Exposure



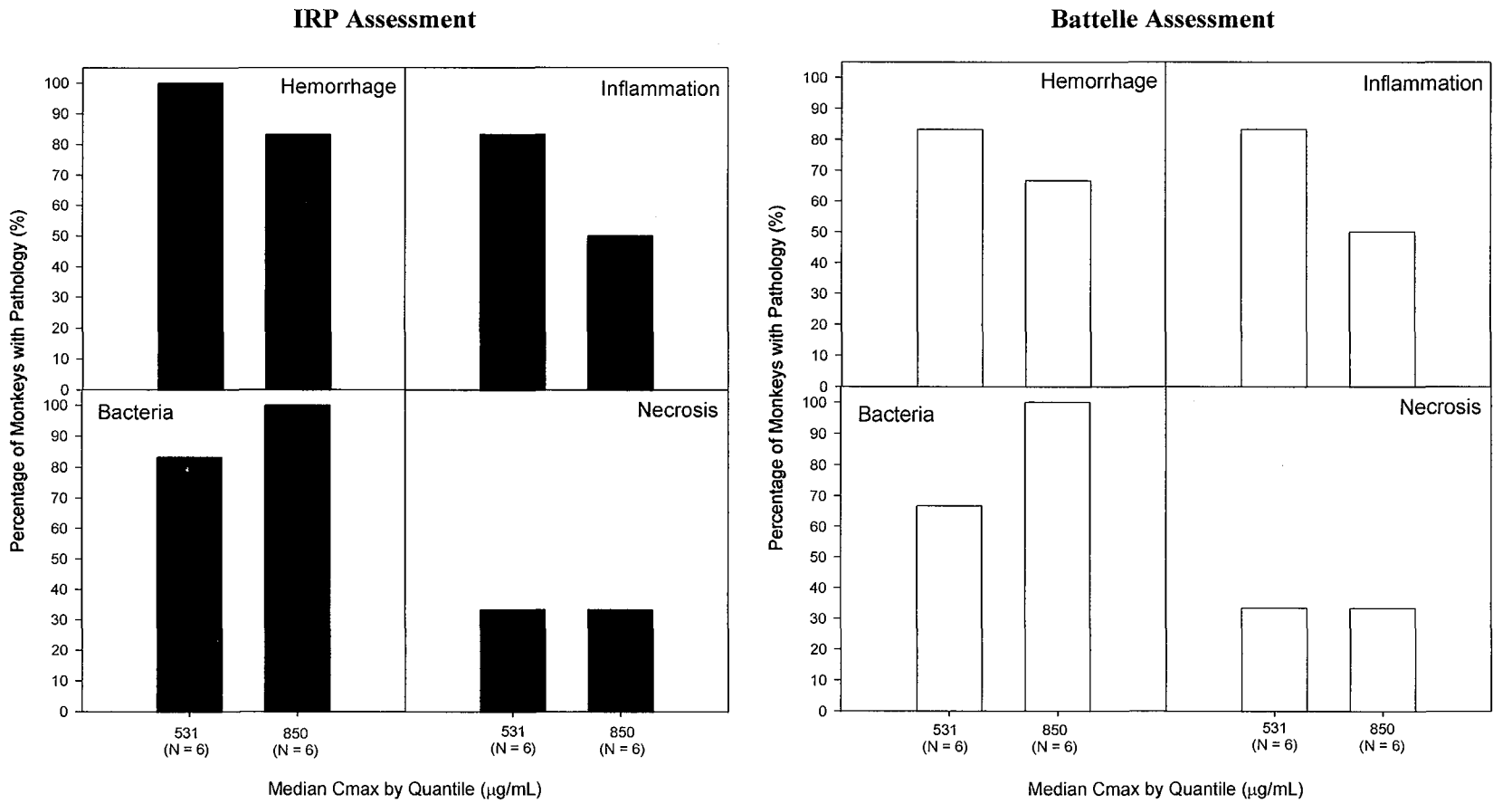
Comparisons of Cmax and AUCinf values by quantile and the incidence of CNS pathology mirrors that of dose, ie a lower incidence of CNS pathology was observed with higher quantiles of exposure. Comparisons of exposure values and the incidence of specific pathological findings (bacteria, hemorrhage, inflammation, necrosis) in rabbits and monkeys are presented in Figures 2.2.4.2-6 through 2.2.4.2-8. In rabbits, a lower incidence of bacteria, hemorrhage, and inflammation, but not necrosis, with higher exposures (both Cmax and AUC) is suggested. In monkeys, a lower incidence of hemorrhage and inflammation with higher exposures (primarily for Cmax). Conversely, the higher quantiles of exposure had a higher incidence of bacteria in the CNS. In monkeys, the incidence of necrosis did not differ between the two quantiles of exposure.

Figure 2.2.4.2-6 Comparison of Incidence of Specific CNS Pathology Findings in Rabbits that Died by Quantile of Raxibacumab Cmax



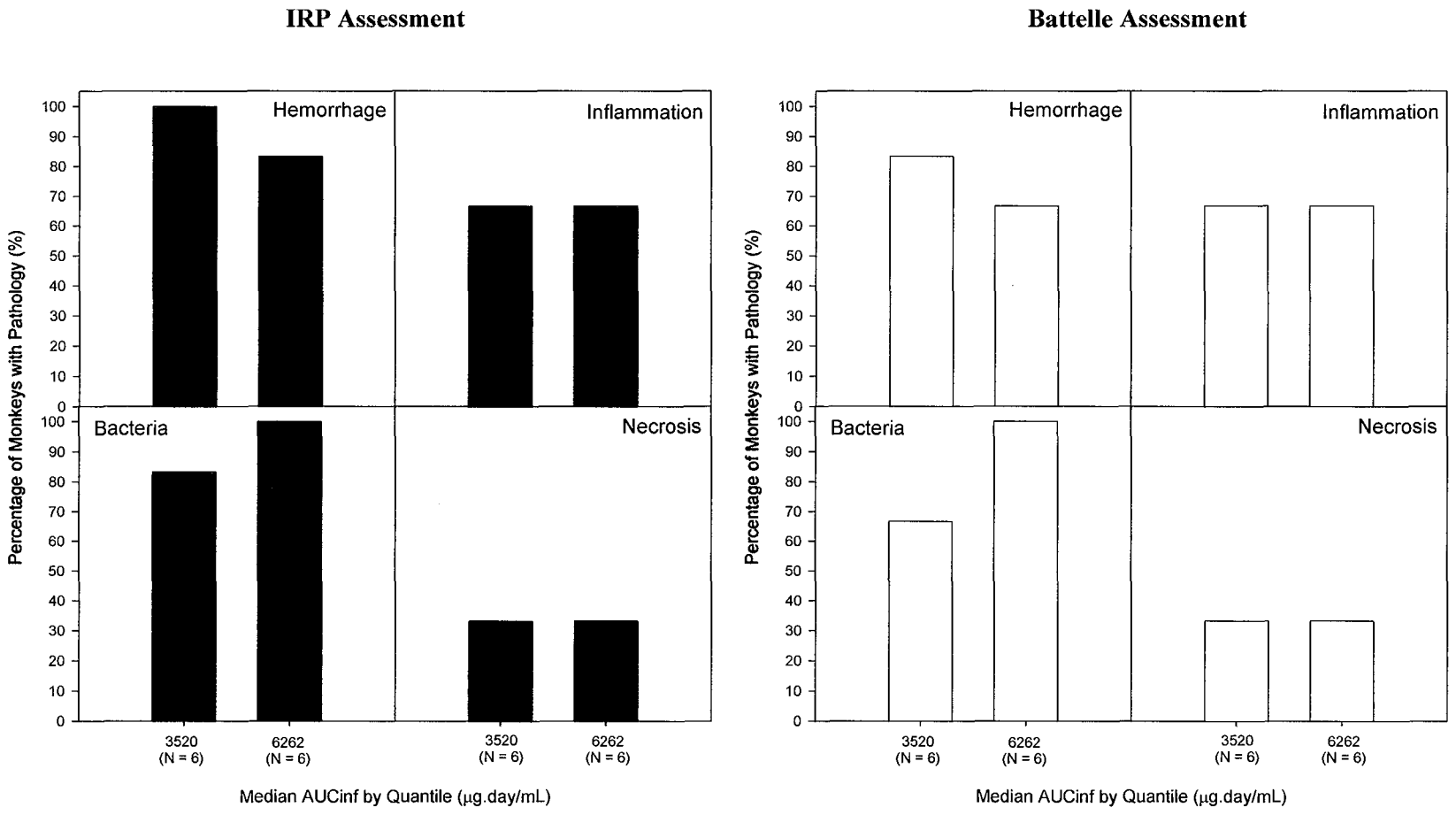
Quantile ranges: Quantile 1 (342 – 543), Quantile 2 (624 – 1166)
IRP, readings performed by independent pathologist review
Battelle, readings performed by board-certified pathologist at study site
A comparison of AUCinf values by quantile showed results identical to the results for Cmax (data not shown).

Figure 2.2.4.2-7 Comparison of Incidence of Specific CNS Pathology Findings in Monkeys that Died by Quantile of Raxibacumab Cmax



Quantile ranges: Quantile 1 (276 – 600), Quantile 2 (613 – 1097)
IRP, readings performed by independent pathologist review
Battelle, readings performed by board-certified pathologist at study site

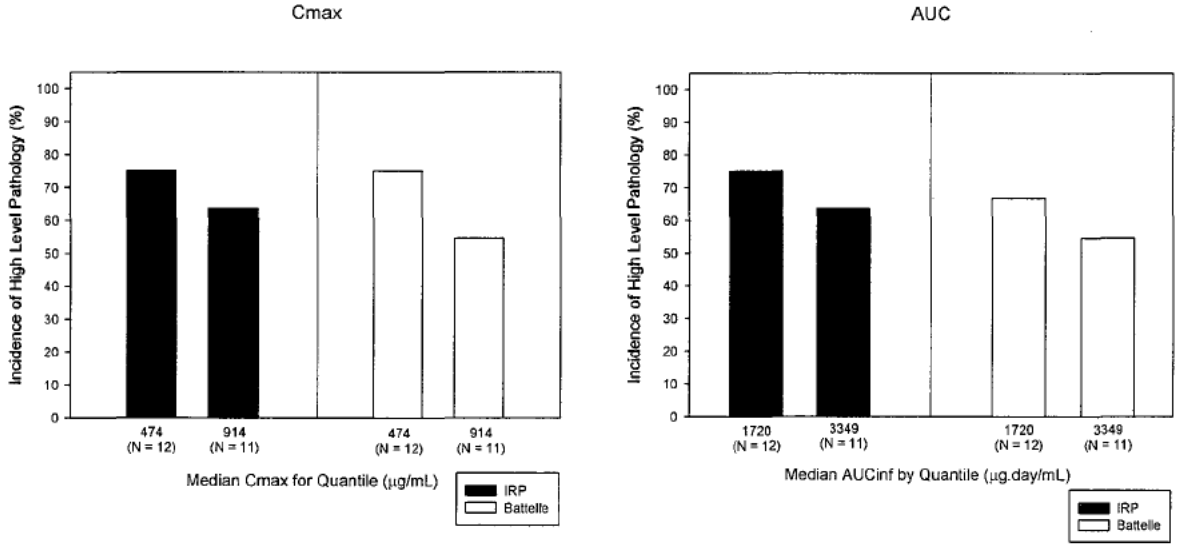
Figure 2.2.4.2-8 Comparison of Incidence of Specific CNS Pathology Findings in Monkeys that Died by Quantile of Raxibacumab AUC



Quantile ranges: Quantile 1 (2597 – 4080), Quantile 2 (4780 – 8653)
IRP, readings performed by independent pathologist review
Battelle, readings performed by board-certified pathologist at study site

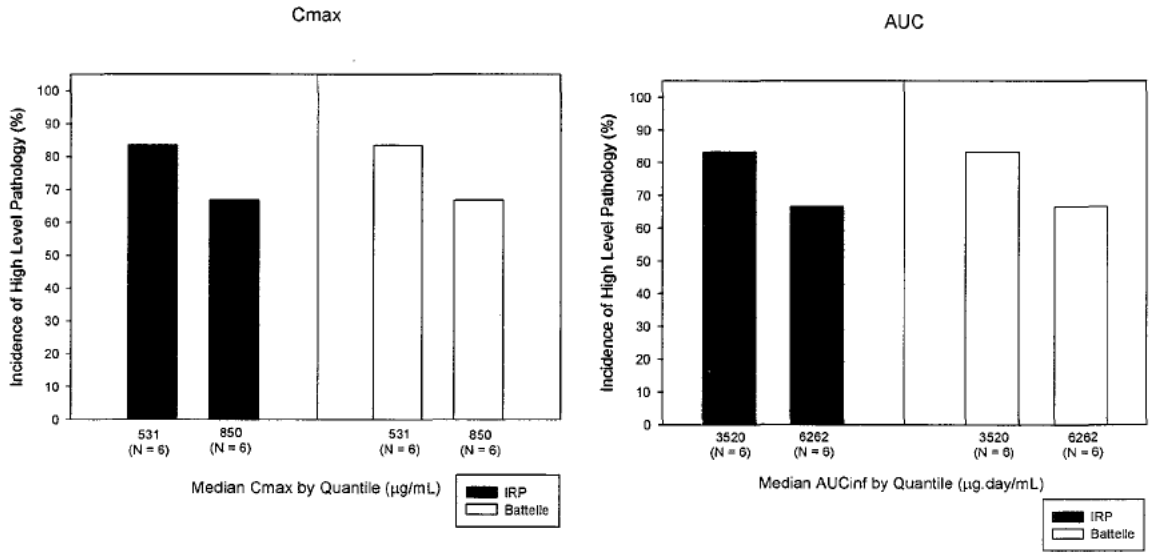
Comparisons of exposure values and the incidence of high level CNS pathology (defined as pathology Grades ≥ 3) are presented in Figures 2.2.4.2-9 and 2.2.4.2-10. Although a trend towards lower incidence of high level CNS pathology in higher quantiles of exposure was observed, the number of animals resulting in these differences is small.

Figure 2.2.4.2-9 Comparison of Incidence of High Level (Grades ≥ 3) CNS Pathology in Rabbits that Died by Quantile of Raxibacumab Exposure



Cmax Quantile ranges: Quantile 1 (342 – 543), Quantile 2 (624 – 1166) ;
AUC Quantile ranges: Quantile 1 (1380 – 2144), Quantile 2 (2971 – 3978)
IRP, readings performed by independent pathologist review; Battelle, readings performed by board-certified pathologist at study site

Figure 2.2.4.2-10 Comparison of Incidence of High Level (Grades ≥ 3) CNS Pathology in Monkeys that Died by Quantile of Raxibacumab Exposure



Cmax Quantile ranges: Quantile 1 (276 – 600), Quantile 2 (613 – 1097)
AUC Quantile ranges: Quantile 1 (2597 – 4080), Quantile 2 (4780 – 8653)
IRP, readings performed by independent pathologist review; Battelle, readings performed by board-certified pathologist at study site

In conclusion, no clear relationship between exposure and CNS pathology can be discerned based on the limited amount of data in animals that died. Overall, the raxibacumab 20 mg/kg dose group had a higher incidence and severity of CNS findings than the 40 mg/kg group in both rabbits and monkeys. Due to the small numbers of animals that exhibited specific pathology findings, no definitive conclusions about exposure-response for bacteria, hemorrhage, inflammation, or necrosis could be made.

The potential hypothesis proposed by the applicant that animals that received raxibacumab survived longer (and thus had more brain lesions that increased over time) does not appear to be a likely explanation for the higher CNS pathology rates in animals treated with raxibacumab. In rabbits that died, placebo animals have comparable survival times versus animals that received the higher raxibacumab dose (40 mg/kg). In monkeys that died, animals that received placebo lived longer than those receiving raxibacumab.

In rabbits that died, animals in the lower Cmax and AUC quantiles appeared to live longer than the other groups and placebo, but in monkeys there was no discernable difference between survival times for the higher and lower quantiles of exposure. The placebo group exhibited a longer survival time compared to the raxibacumab groups. This suggests that in animals that died, the magnitude of raxibacumab exposure did not affect survival time.

The potential hypothesis proposed by the applicant that time to death in animals with moderate to marked inflammation was longer than that for animals without moderate to marked inflammation (i.e. that more severe inflammatory brain findings are associated with a longer time to death) is plausable. This appears to be true only for the raxibacumab treated animals; in animals that received placebo, rabbits with low grade CNS findings lived longer than those with high grade pathology.

2.2.4.3. Is the dose and dosing regimen selected by the applicant consistent with the known relationship between dose-concentration-response, and are there any unresolved dosing or administration issues?

The dose and dosing regimen selected by the applicant, raxibacumab 40 mg/kg IV single dose, is consistent with the known relationship between dose-concentration-response (survival). The proposed dose of 40 mg/kg raxibacumab in humans would be expected to maintain levels required for virtually complete binding (99.9%) of PA for 7 days and those required for 99% binding for up to 42 days, and more than 95% of humans administered a 40 mg/kg IV raxibacumab dose can be expected to have serum raxibacumab concentrations that are equimolar to or in excess of the highest observed serum/plasma PA concentration in any animal that died in the therapeutic intervention studies (for 48 and 28 days relative to rabbits and monkeys, respectively).

Dose and concentration response for the endpoint of survival indicates potential to achieve higher response rates at higher doses. If future trials are conducted, higher doses (e.g. 60 or 80 mg/kg) should be explored along with 40 mg/kg. In any future trials studying higher doses in animals, safety assessments should include an examination of CNS pathology.

2.2.5. What are the PK characteristics of raxibacumab?

2.2.5.1. What are the single dose and multiple dose PK parameters?

The pharmacokinetics of raxibacumab (manufactured by the M11 process) following intravenous administration was evaluated in humans, monkeys and rabbits in the studies described in Table 2.2.5.1-1.

Table 2.2.5.1-1. Animal and Clinical Pharmacology Studies for Raxibacumab

Study Number and Title	Phase (number of centers)	Study Population	IV dose (mg/kg)	Regimen	Number of evaluable subjects
Human Studies					
HGS1021-C1063 A Randomized, Single-Blind, Placebo-Controlled Study to Evaluate the Safety and Tolerability of Raxibacumab (Human Monoclonal Antibody to *Bacillus anthracis* Protective Antigen) in Healthy Subjects	Phase 3 (6 US centers)	Healthy volunteers (54% females; 12% Hispanic, 21% non-white; **≥ 18 years of age, 9% ≥ 65 years of age)**	SD Grp:40	Single dose	217
			DD Grp: 40	Two doses given 14 days apart	23
HGS1021-C1064 An Open-Label Study to Evaluate the Pharmacokinetics and Safety of Raxibacumab (Human Monoclonal Antibody to *Bacillus anthracis* Protective Antigen) Administered in Combination with Ciprofloxacin in Healthy Subjects	Phase 2/3 (3 US centers)	Healthy volunteers (43 males, 45 females; 53 White, 31 Black or African American, 4 other; 18 to 60 years of age)	Grp 1[a]: 40	Single dose	30
			Grp 2: 40	Single dose	28
			Grp 3[b]: 40	Single dose	28
HGS1021-C1069 An Open-Label Study to Evaluate the Immunogenicity and Safety of Raxibacumab (Human Monoclonal Antibody to *Bacillus anthracis* Protective Antigen) Administered in Healthy Subjects	Phase 2/3 (2 US centers)	Healthy volunteers recruited from HGS1021-C1064 subjects (12 males, 8 females; 13 White, 7 Black or African American; 23 to 61 years of age)	40	Two doses given ≥ four months after prior dose (in HGS1021-C1064)	20[c]
Animal Studies					
Study Number and Title	Target Spore Challenge (× LD_{50})	Animal Characteristics	Doses Studied (mg/kg)	Treatment	Number of Animals[2]
682-G005758 Evaluation of Raxibacumab Efficacy as Therapeutic Treatment Against Inhalation Anthrax in the Rabbit Model	200	8 males 8 females	20 40	IV raxibacumab vehicle	16
724-G005829 Evaluation of Raxibacumab Efficacy as Therapeutic Treatment Against Inhalation Anthrax in the Cynomolgus Macaque	200	6 males 6 females	20 40	IV raxibacumab vehicle	12

SD, single dose; Grp, group; DD, double dose

[a] Raxibacumab was administered concurrent with oral (PO) 500 mg q12 hour x 15 ciprofloxacin doses; the raxibacumab dose was administered immediately following the 11th ciprofloxacin dose.

[b] Raxibacumab was administered concurrent with IV 400 mg q12 hour x 2 ciprofloxacin doses, which were followed by PO 500 mg q12 hour x 13 ciprofloxacin doses; the raxibacumab dose was administered immediately following the 1st IV ciprofloxacin dose.

[c] Number of subjects not included in the total (these subjects were enrolled in the previous study HGS1021-C1064).

Single Dose Pharmacokinetics of Raxibacumab

Pharmacokinetic data from the human study HGS1021-C1064 and the pivotal animal studies 724-G005829 and 682-G005758 were used for the primary comparison of single dose raxibacumab exposure across species. Serum concentration-time profiles for raxibacumab following single intravenous administration of 40 mg/kg in healthy, male and female subjects, monkeys with inhalation anthrax, and rabbits with inhalation anthrax are presented in Figure 2.2.4.1-1. Pharmacokinetic parameters for raxibacumab following single intravenous administration of 40 mg/kg in healthy, male and female subjects, monkeys with inhalation anthrax, and rabbits with inhalation anthrax are summarized in Table 2.2.4.1-2.

Following single intravenous administration of raxibacumab 40 mg/kg in healthy, male and female human subjects, raxibacumab appeared to distribute to tissues; mean Vss were greater than plasma volume, ranging between 65 to 72 mL/kg. Serum concentration-time profiles for raxibacumab suggest biphasic elimination with a mean terminal phase elimination half-lives (t1/2,z) range from 20 to 22 days. Mean clearance (CL) ranged from 2.6 to 3.0 mL/day/kg among the dose groups studied. CL values were much smaller than the glomerular filtration rate indicating that there is virtually no renal clearance of raxibacumab.

Serum raxibacumab concentrations in monkeys administered raxibacumab were best fit to a 2-compartment model with 1st-order elimination. The lack of difference in PK between treatment groups is consistent with linear PK over the dose range studied. Following IV raxibacumab administration, V1 was approximately 53 and 40 mL/kg for a 3 kg male and female animal, respectively, similar to plasma volume. The steady-state volume of distribution (Vss) was nearly 2-fold greater than V1, suggesting that although distribution of raxibacumab may initially be restricted to the plasma volume, raxibacumab does subsequently distribute to tissues. The disappearance of raxibacumab from serum is multiphasic. The mean CL of raxibacumab in the monkey was much less than the glomerular filtration rate (2995 mL/day/kg), indicating that, similar to humans, there is virtually no renal clearance of raxibacumab. Inter-individual variability in raxibacumab PK was low, with CV% of 26% or less for the primary PK parameters. V1 and CLD2 are dependent on sex, with females having smaller values for each parameter than males.

Similar to monkeys, serum raxibacumab concentrations in rabbits were best fit a 2-compartment model with 1st-order elimination. The lack of difference in PK between treatment groups is consistent with linear PK over the dose range studied. Following IV raxibacumab administration, V1 was the same as the plasma volume. The steady-state volume of distribution (Vss) is about 43% greater than V1, suggesting that although distribution of raxibacumab may initially be restricted to the plasma volume, raxibacumab does subsequently distribute to tissues. Similar to humans and monkeys, mean CL of raxibacumab was much smaller than the glomerular filtration rate (4493 mL/day/kg) indicating that there is virtually no renal clearance of raxibacumab. Inter-individual variability in raxibacumab PK was low, with CV% of 27% or less for the primary PK parameters.

Figure 2.2.4.1-1. Mean (SD) Serum Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 40 mg/kg in Healthy, Male and Female Subjects, Monkeys with Inhalation Anthrax, and Rabbits with Inhalation Anthrax

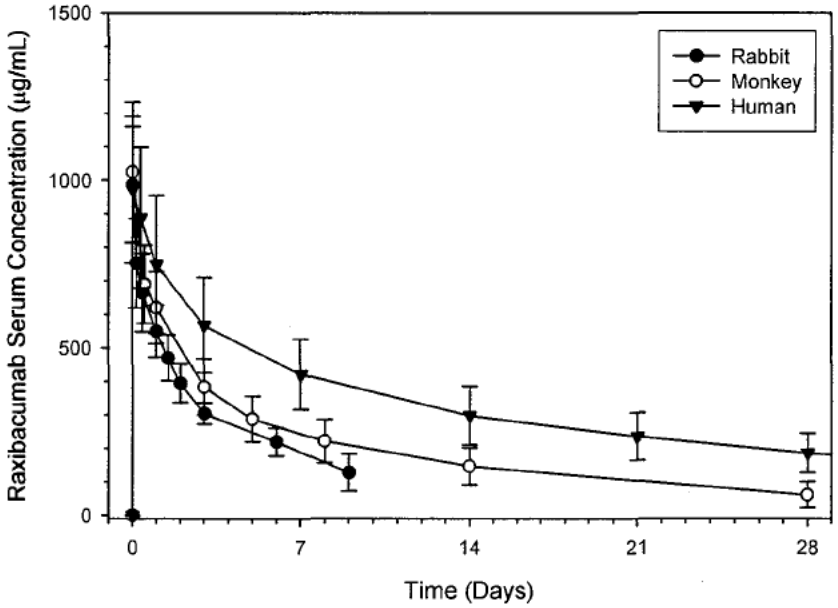


Table 2.2.5.1-2. Pharmacokinetic parameters for raxibacumab following single intravenous administration of 40 mg/kg in healthy, male and female subjects, monkeys with inhalation anthrax, and rabbits with inhalation anthrax

Species	Dose Group	N	Cmax (µg/ml)	AUCinf (µg·day/ml)	Half life (days)	CL (ml/day/kg)	Vss (ml/kg)
Rabbit	**20 mg/kg**	17	459.9 ± 57.8 (342.5 - 544.8)	1719.1 ± 223.5 (1380.4 - 2196.7)	3.97 ± 0.73 (2.63 – 5.44)	11.6 ± 1.71 (9.10 – 15.5)	63.11 ± 7.91 (52.9 – 82.0)
Rabbit	**40 mg/kg**	19	918.8 ± 124.0 (623.8 – 1166.0)	3424.0 ± 464.1 (2541.4 – 3348.7)	4.10 ± 0.85 (2.99 – 6.10)	11.7 ± 1.81 (8.83 – 15.23)	63.8 ± 8.48 (51.9 – 87.9)
Monkey	**20 mg/kg**	14	490.5 ± 87.7 (275.6 – 613.2)	3575.9 ± 827.9 (2499.0 – 5240.4)	11.1 ± 1.94 (7.95 – 14.61)	5.85 ± 1.29 (3.80 – 8.00)	84.2 ± 10.3 (73.8 – 110.3)
Monkey	**40 mg/kg**	14	989.8 ± 170.1 (788.8 – 1422.9)	6490.6 ± 2095.5 (3899.4 – 11934.5)	10.1 ± 2.35 (5.98 – 14.3)	6.61 ± 1.90 (3.30 – 10.2)	82.8 ± 9.10 (64.5 – 93.3)
Human Group 1	**40 mg/kg + cipro PO**	27[a]	1143.3 ± 169.5 (833.6 – 1517.8)	14871.9 ± 3821.1 (5494.5 – 21116.6)	19.8 ± 8.18 (4.27 – 45.8)	2.99 ± 1.38 (1.85 – 7.30)	64.9 ± 11.9 (44.1 – 99.8)
Human Group 2	**40 mg/kg**	27[b]	1020.3 ± 140.6 (766.2 – 1293.7)	15845.8 ± 4333.5 (7614.9 – 25464.6)	20.6 ± 6.54 (5.93 – 32.3)	2.73 ± 0.84 (1.57 – 5.24)	69.7 ± 13.7 (45.6 – 106.7)
Human Group 3	**40 mg/kg + cipro IV/PO**	28	1047.8 ± 180.3 (772.5 – 1458.4)	16349.3 ± 4255.7 (7117.5 – 27465.2)	21.5 ± 8.92 (6.53 – 42.9)	2.63 ± 0.82 (1.46 – 5.62)	67.2 ± 12.6 (47.6 – 93.2)

Data presented represents mean ± one standard deviation and (minimum – maximum)

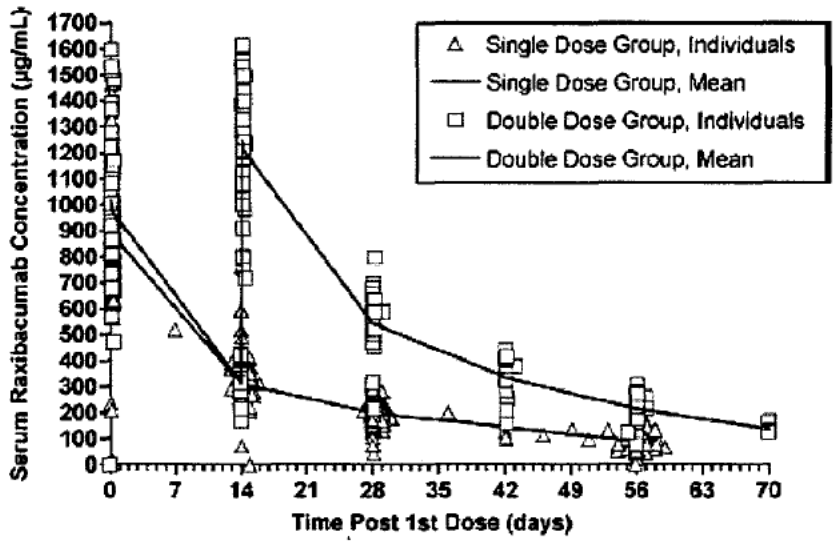
[a] Subjects US003-000006 and US003-000028 excluded for receiving only partial doses of raxibacumab. Subject US003-00029 excluded for PK profile uncharacteristic of IV administration.

[b] Subject US003-000002 excluded for receiving only a partial dose of raxibacumab.

Multiple Dose Pharmacokinetics of Raxibacumab

Although raxibacumab is intended for single dose administration, Study HGS1021-C1063 was designed to evaluate the safety and tolerability of repeat-dose administration of raxibacumab, and multiple dose pharmacokinetics of raxibacumab were assessed as a secondary objective. Serum concentrations for raxibacumab following single and repeat intravenous administration (after 14 days) of 40 mg/kg in healthy, male and female subjects are presented in Figure 2.2.4.1-2. For purposes of comparison, the mean serum raxibacumab concentration-time profile following a single 40 mg/kg IV infusion raxibacumab in Study HGS1021-C1064 is presented. As expected with a half-life of approximately 20 days, accumulation of raxibacumab concentrations was observed upon repeat dosing at 14 days. Serum raxibacumab concentrations were consistent between the single- and double-dose groups up to the time of the second dose.

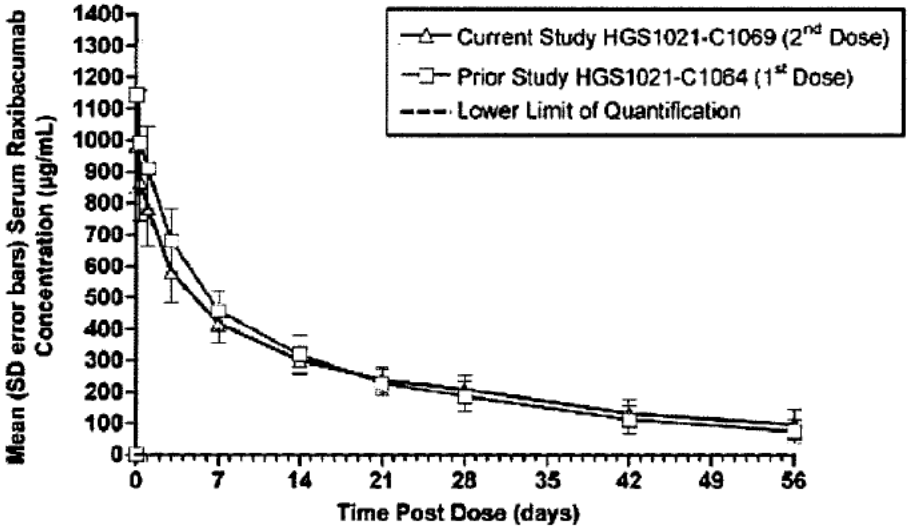
Figure 2.2.4.1-2. Individual serum concentrations and mean (± SD) serum concentration-time profiles for raxibacumab following single (single-dose group) and multiple (double-dose group) intravenous administration of 40 mg/kg in healthy, male and female subjects



Source: Study Report HGS1021-C1063, Section 10.4.4

In the human study assessing the immunogenicity of raxibacumab (Study HGS1021-C1069), subjects who had received raxibacumab in a prior to study (in Study HGS1021-C1064) received a second dose of raxibacumab equal to that of their previous dose ≥ 4 months following their first dose. Serum raxibacumab concentration-time profiles were very similar for these first and second doses, as displayed in Figure 2.2.4.1-3.

Figure 2.2.4.1-3. Mean (± SD) serum concentration-time profiles for raxibacumab following the first (Study HGS1021-C1064) and second (Study HGS1021-C1069) doses of raxibacumab 40 mg/kg IV administered at least four months apart in healthy, male and female subjects



Source: Study Report HGS1021-C1069, Section 9.2.4

Pharmacokinetic parameters for raxibacumab following the first (Study HGS1021-C1064) and second (Study HGS1021-C1069) doses of raxibacumab 40 mg/kg IV administered at least four months apart in healthy, male and female subjects are summarized in Table 2.2.5.1-3. There were no statistically significant differences in mean AUCinf, t1/2,z, or CL between the second dose administered in study HGS1021-C1069 and the first dose administered in study HGS1021-C1064. The mean Cmax for the second dose was significantly lower (-15%, p = 0.0008) than that for the first dose. Although the difference in mean Cmax between two doses administered ≥ **four months apart** was statistically significant, the difference is less than 20%. An examination of individual Cmax values does not indicate a consistent decrease in Cmax values for the second dose relative to the first dose among the subjects, as displayed in Figure 2.2.4.1-4. Thus, the clinical significance of this difference is questionable. Also, the median Tmax for the second dose was significantly shorter than that for the first dose (-3%, 0.0019). Review of the individual Tmax values show that the slightly larger tmax for the first dose may be driven by two outlier subjects, as depicted in Figure 2.2.4.1-5. Thus, this small difference in median tmax (-3%) is unlikely to be clinically meaningful.

Table 2. Summary of raxibacumab pharmacokinetic parameters following first (Study HGS1021-C1064) and second (Study HGS1021-C1069) doses of raxibacumab 40 mg/kg IV administered at least four months apart in healthy, male and female subjects

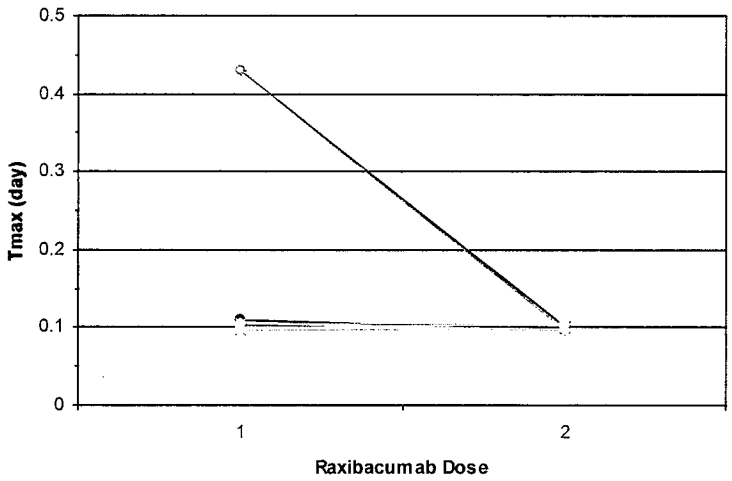
Parameter	HGS1021-C1064 (first dose) (N = 20)	HGS1021-C1069 (second dose) (N = 20)	p-value
Cmax (µg/mL)	1152 (176)	979 (148)	0.0008
AUCinf (µg·day/mL)	16440 (4140)	18239 (6179)	0.1798
Tmax (day)[a]	0.102 (0.097 – 0.431)	0.99 (0.097 – 0.104)	0.0019[b]
T1/2 (day)	21.20 (8.62)	25.68 (11.19)	0.1535
CL (mL/day/kg)	2.59 (0.77)	2.37 (0.63)	0.3017
Vss (mL/kg)	64.73 (14.02)	75.72 (11.42)	0.0122

Data presented as mean (SD), unless otherwise specified.
P-value from paired t-test, unless otherwise specified.
[a] Data presented at median (range).
[b] P-value from Wilcoxon matched pairs test.

Source: HGS1021-C1069, Section 3.5.1.4

Figure 2.2.4.1-4. Individual Cmax values for raxibacumab following the first (Study HGS1021-C1064) and second (Study HGS1021-C1069) doses of raxibacumab 40 mg/kg IV administered at least four months apart in healthy, male and female subjects

Figure 2.2.4.1-5. Individual Tmax values for raxibacumab following the first (Study HGS1021-C1064) and second (Study HGS1021-C1069) doses of raxibacumab 40 mg/kg IV administered at least four months apart in healthy, male and female subjects



2.2.5.2. How does raxibacumab exposure following single 40 mg/kg IV dosing compare between species?

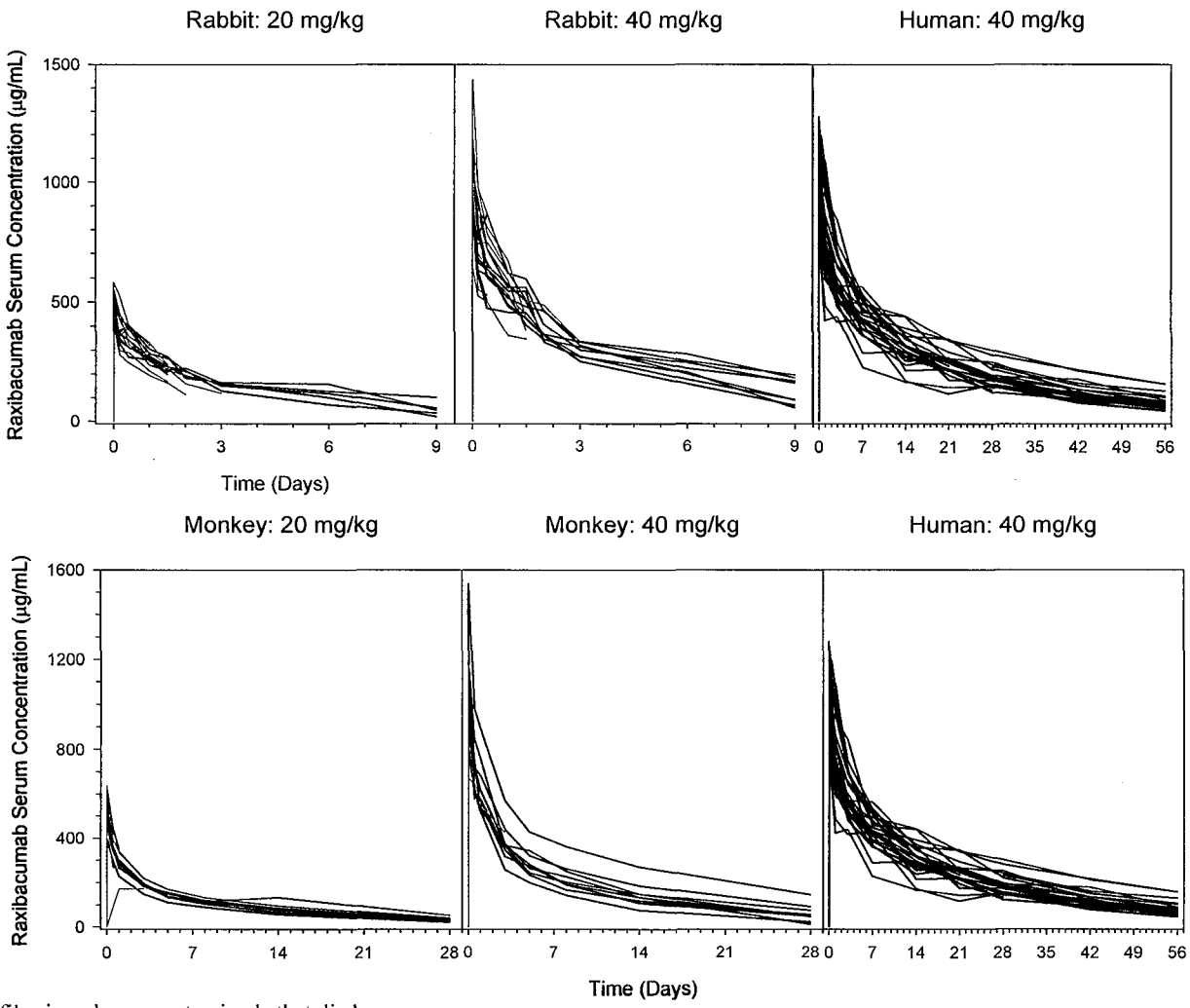
To addresses the sufficiency of the clinical pharmacology data to support the use of a 40 mg/kg raxibacumab dose for therapeutic treatment of inhalation anthrax, measures of exposure (serum concentrations, Cmax, and AUC) were compared across humans, monkeys, and rabbits receiving single IV doses of raxibacumab (M11). Individual serum concentration-time profiles for raxibacumab following single intravenous administration of 40 mg/kg in healthy, male and female subjects and 20 mg/kg and 40 mg/kg in monkeys and rabbits with inhalation anthrax are presented in Figure 2.2.4.2-1. The range of raxibacumab serum concentrations observed in both monkeys and rabbits following single 20 mg/kg and 40 mg/kg IV doses are compared to individual human serum concentration-time profiles following a single 40 mg/kg dose in Figures 2.2.4.2-2 and 2.2.4.2-3. Point plots of individual Cmax and AUCinf values following single intravenous administration of 40 mg/kg in healthy, male and female subjects and 20 mg/kg and 40 mg/kg in monkeys and rabbits with inhalation anthrax are illustrated in Figure 2.2.3.2-4.

Serum concentrations of raxibacumab following a single IV dose of 40 mg/kg alone in humans fell within or exceeded the range of concentrations observed in rabbits and monkeys receiving single 40 mg/kg IV doses, except for two subjects at the 24-hour sampling time point. Mean raxibacumab Cmax in humans following a single 40 mg/kg IV dose was similar to or greater than mean Cmax values in monkeys and rabbits. Individual raxibacumab Cmax values in humans following a single IV dose of 40 mg/kg alone fell within the range of Cmax values observed in rabbits. All but one Cmax in humans fell within the range of values observed in the monkeys. Mean raxibacumab AUCinf in humans following a single 40 mg/kg IV dose was 2.4 and 4.6-fold that of the mean Cmax values in monkeys and rabbits, respectively. Individual raxibacumab AUCinf values in humans following a single IV dose of 40 mg/kg alone fell within or exceeded the range of AUCinf values observed in rabbits and monkeys. Mean total clearance of raxibacumab was substantially slower in human as compared to rabbits by a factor of approximately 5 and in monkeys by a factor of approximately 3. Consequently, mean half-life of raxibacumab was longer in humans compared to mean half-lives observed in monkeys and rabbits (20.6 ± 6.5 days versus 10.1 ± 2.4 days and 4.1 ± 0.85 days, respectively). Variability in Cmax was

similar across species; %CV values ranged between approximately 13 to 17%. Variability in AUCinf was wider, ranging between 13 and 32% across species.

In summary, humans achieve similar to or greater exposure to raxibacumab following a single 40 mg/kg IV dose compared to rabbits and monkeys receiving the same dose. Humans achieve much greater exposure to raxibacumab following a single 40 mg/kg IV dose compared to rabbits and monkeys receiving a 20 mg/kg dose.

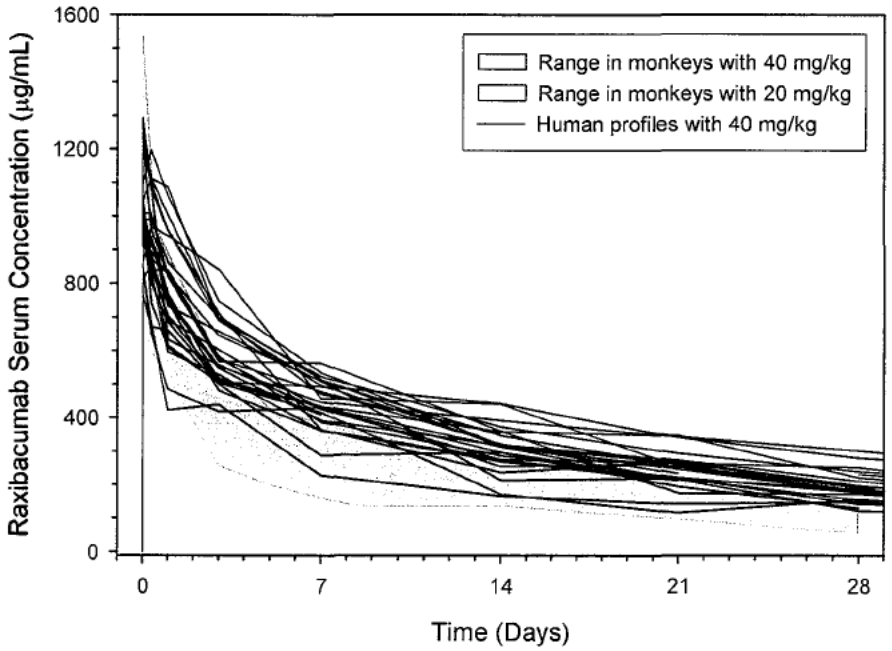
Figure 2.2.4.2-1. Individual Serum Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 40 mg/kg in Healthy, Male and Female Subjects and 20 mg/kg and 40 mg/kg in Monkeys and Rabbits with Inhalation Anthrax



Serum concentration profiles in red represent animals that died.

Figure 2.2.4.2-2. Range of Serum Concentrations of Raxibacumab in Monkeys Following 20 mg/kg and 40 mg/kg Single IV Doses Versus Individual Serum Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 40 mg/kg in Humans

Graph A: data to 28 days



Graph B: data to 7 days

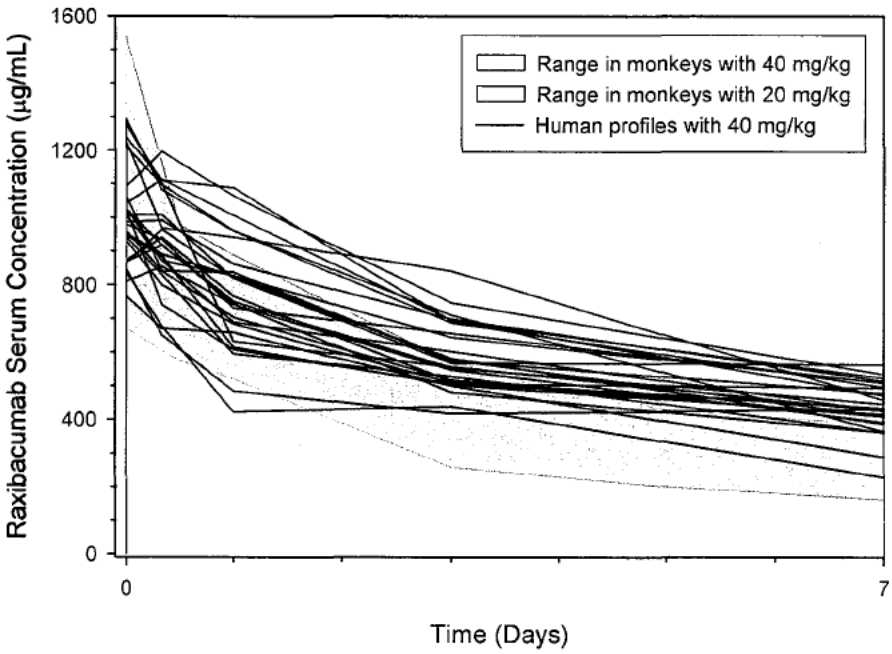


Figure 2.2.4.2-3. Range of Serum Concentrations of Raxibacumab in Rabbits Following 20 mg/kg and 40 mg/kg Single IV Doses Versus Individual Serum Concentration-Time Profiles for Raxibacumab Following Single IV Administration of 40 mg/kg in Humans

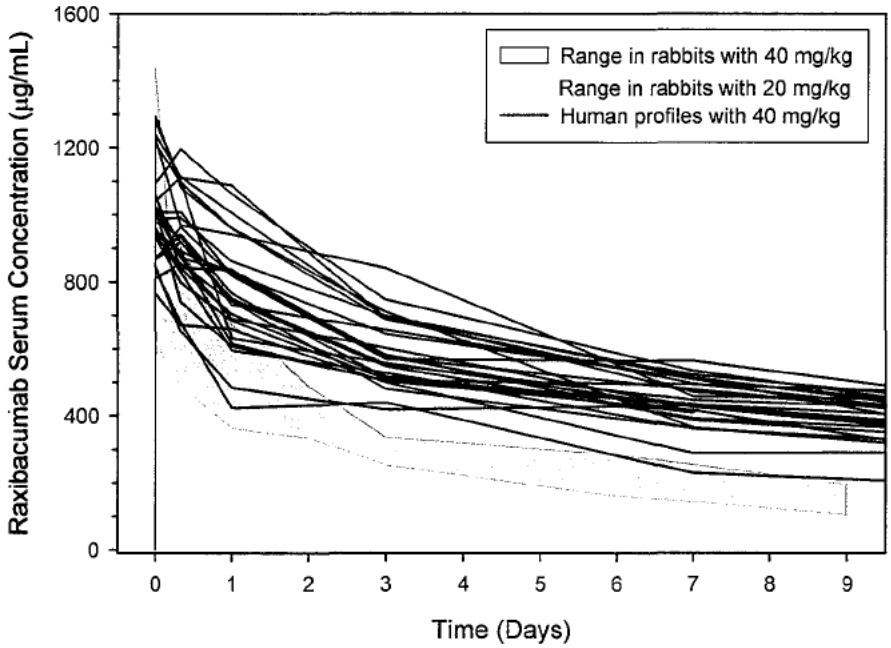
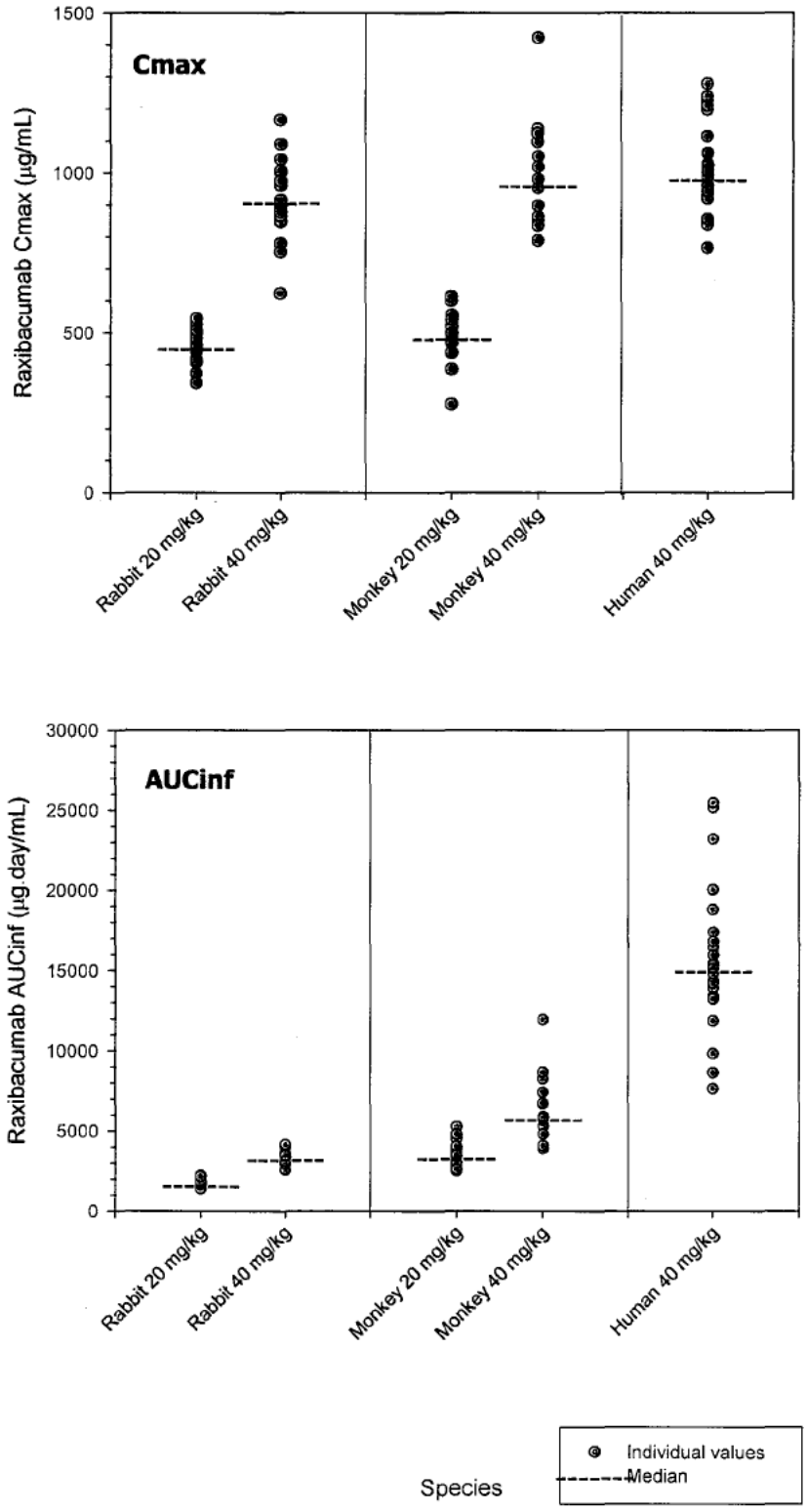


Figure 2.2.4.2-4. Individual Raxibacumab Cmax and AUCinf Values Following Single Intravenous Administration of 40 mg/kg in Healthy, Male and Female Subjects and 20 mg/kg and 40 mg/kg in Monkeys and Rabbits with Inhalation Anthrax

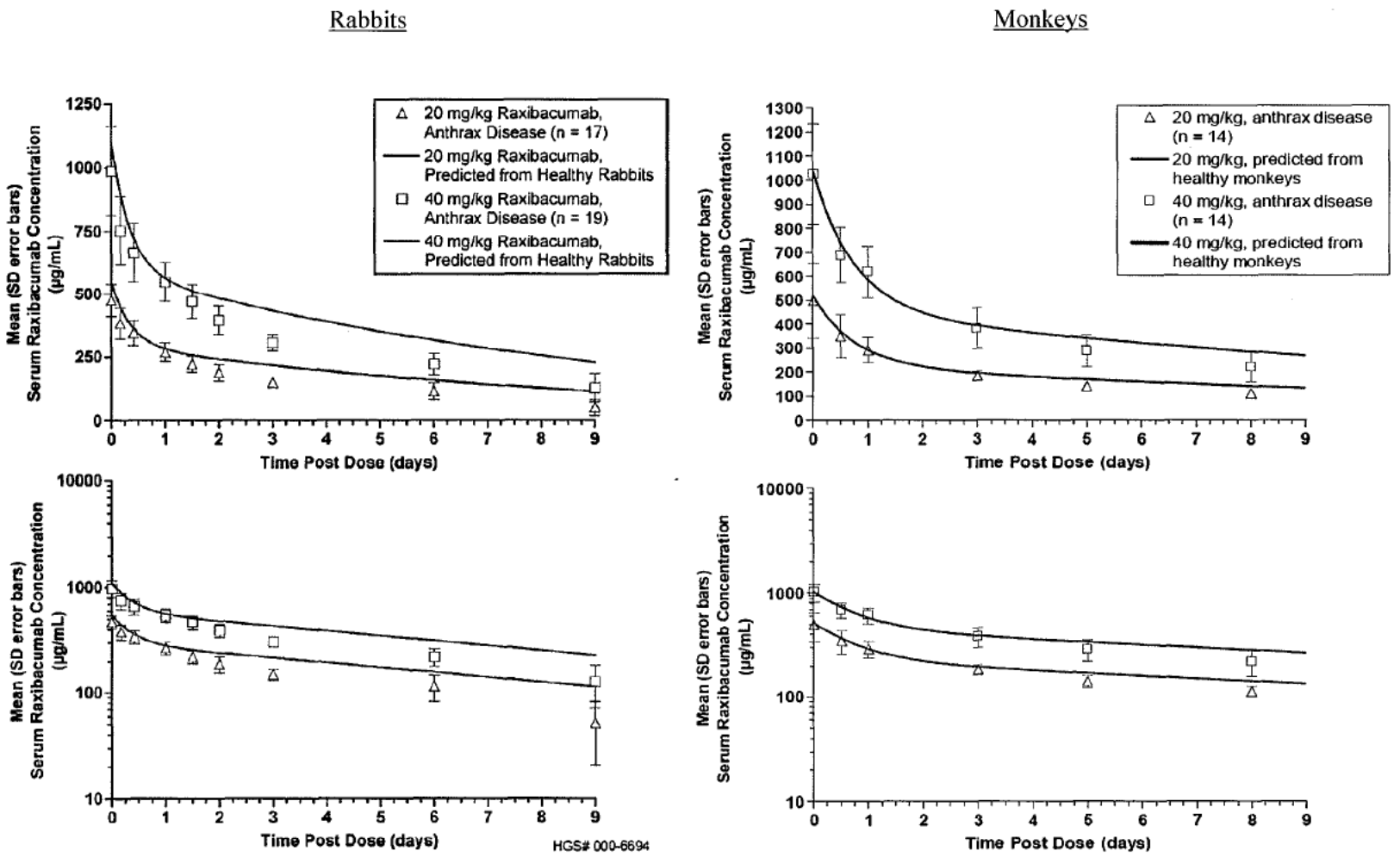


2.2.5.3. *How does the PK of the drug and its major active metabolites in healthy volunteers compare to that in patients?*

Due to the nature and incidence of the indication (inhalational anthrax) and since evaluation of treatment for inhalational anthrax is not possible in controlled clinical trials in humans, the PK of raxibacumab has not been evaluated in patients.

Raxibacumab PK in healthy and anthrax-infected rabbits and monkeys was examined for possible drug-disease effects on raxibacumab disposition. Serum raxibacumab concentration-time profiles from the pivotal efficacy studies in anthrax-infected rabbits and monkeys are compared with predicted PK profiles in healthy animals in Figure 2.2.5.3-1. The profiles suggest that raxibacumab was cleared from the plasma more rapidly (shorter terminal half-life) in animals with anthrax versus healthy animals. Raxibacumab CL in healthy rabbits ranged from 6.2 to 6.9 mL/kg/day, versus that for rabbits with anthrax, at 11.7 mL/kg/day. Similarly, $t1/2,\beta$ in healthy rabbits ranged from 6.9 to 8.7 days, while in rabbits with anthrax, $t1/2,\beta$ was shorter, at 3.8 days. Similarly, raxibacumab CL in healthy monkeys, which ranged from 4.1 to 5.0 mL/kg/day, versus that for monkeys with anthrax, at 6.7 mL/kg/day. Similarly, $t1/2,\beta$ in healthy monkeys ranged from 11.8 to 15.8 days, while in monkeys with anthrax, $t1/2,\beta$ was shorter, at 9.4 to 10.2 days. These differences in clearance of raxibacumab may be a reflection of altered physiology in anthrax disease, but extrapolation of these findings to humans is difficult. Distributional changes, binding of raxibacumab to PA, or a combination of these factors could influence raxibacumab disposition. It is not possible to differentiate the contributions of these possible causes based on the results of these studies.

Figure 2.2.5.3-1. Raxibacumab serum concentration-time profiles in rabbits and monkeys administered single IV bolus 20 or 40 mg/kg raxibacumab compared with the profiles predicted based on raxibacumab pharmacokinetics for healthy rabbits and monkeys



2.2.5.4. What are the characteristics of drug absorption?

Description of absorption characteristics is not applicable, as raxibacumab is formulated for intravenous administration.

2.2.5.5. What are the characteristics of drug distribution?

The steady state volume of distribution for raxibacumab is approximately 58 to 76 mL/kg. The Vss in rabbits, monkeys and humans is similar across species and indicates distribution beyond the vascular space to tissues.

Raxibacumab is a fully human monoclonal IgG1λ antibody and is not expected to bind other plasma proteins.

2.2.5.6. Does the mass balance study suggest renal or hepatic as the major route of elimination?

A mass balance study was not conducted for raxibacumab.

2.2.5.7. What are the characteristics of drug metabolism?

As a monoclonal antibody, raxibacumab is expected to be degraded into small peptides and individual amino acids. No biotransformation studies have been conducted with raxibacumab.

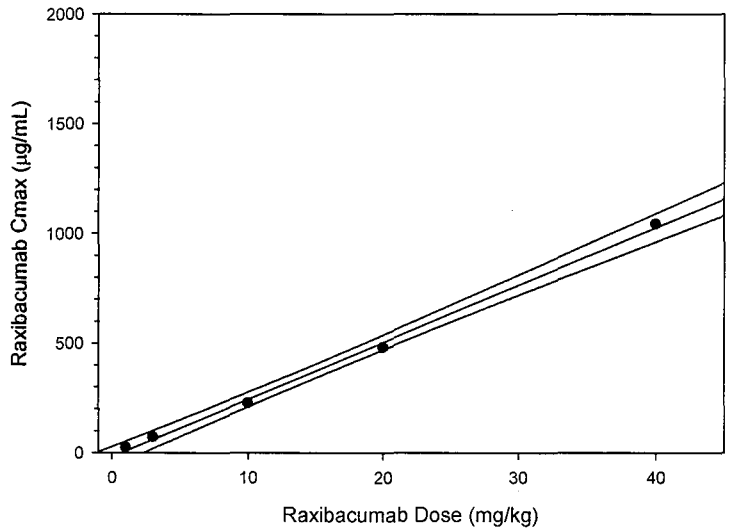
2.2.5.8. What are the characteristics of drug excretion?

Raxibacumab clearance in humans ranges from 2.4 to 2.9 mL/day/kg, values much smaller than the glomerular filtration rate. Therefore, there is virtually no renal clearance of this monoclonal antibody.

2.2.5.9. Based on PK parameters, what is the degree of linearity or nonlinearity in the dose-concentration relationship?

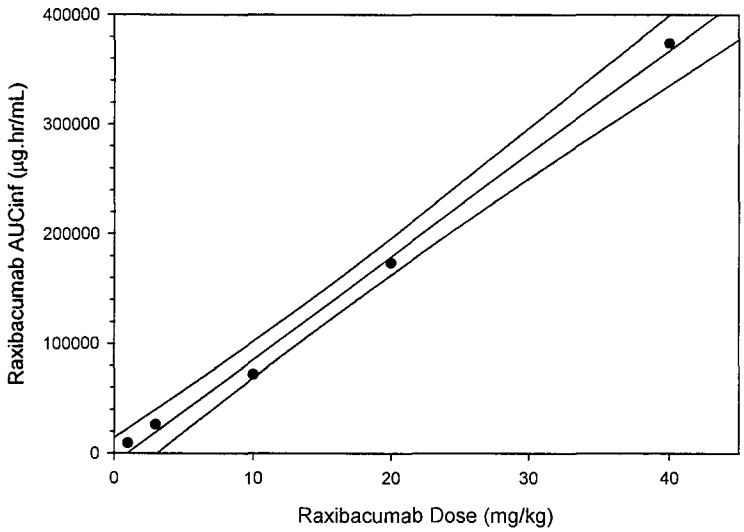
The linearity of raxibacumab PK was not evaluated with the final M11 formulation. In a Phase 1, dose-escalation study of a developmental formulation of raxibacumab, 1.0, 3.0, 10, 20, and 40 mg/kg single IV doses were evaluated in healthy subjects (Study PAM-NH-01). Linear increases in raxibacumab exposure (Cmax and AUCinf) in relation to dose were observed over the dose range, as depicted in Figures 2.2.5.9-1 and 2.2.5.9-2. Dose normalized values for Cmax were similar, ranging between 0.0226 to 0.0261 kg/mL. Dose normalized values for AUCinf ranged between 7.15 to 9.33 kg·h/mL.

Figure 2.2.5.9-1 Effect of Increasing Dose of Raxibacumab on Raxibacumab Cmax



Lines represent linear regression and 95% confidence intervals

Figure 2.2.5.9-2 Effect of Increasing Dose of Raxibacumab on Raxibacumab AUCinf



Lines represent linear regression and 95% confidence intervals

2.2.5.10. How do the PK parameters change with time following chronic dosing?

Raxibacumab is intended for single dose administration; therefore the PK of raxibacumab following chronic dosing was not evaluated.

2.2.5.11. What is the inter- and intra-subject variability of PK parameters in volunteers and patients, and what are the major causes of variability?

Raxibacumab exhibited low degree of intersubject variability (< 30% CV) in PK parameters in healthy volunteers. Variability in systemic clearance in healthy volunteers was 20% across pooled Phase 1 studies in the population PK analysis. In the population PK analysis, the variability in V1, CL, V1, V2 and CLD2 was estimated as approximately 16%, 20%, 21.2%, and 33.4% CV, respectively. Evaluation of subject covariates demonstrated that body weight, sex and race were statistically significant covariates contributing to variability in raxibacumab PK.

2.2.5.12. Does the clinical pharmacology information presented by the applicant support the use of a 40 mg/kg raxibacumab dose for therapeutic treatment of inhalation anthrax?

Based on a comparison of exposure across species to form the basis of extrapolation of efficacy findings from animals to humans, the clinical pharmacology information presented by the applicant support the use of a 40 mg/kg raxibacumab dose for therapeutic treatment of inhalation anthrax in the event of an emergency. Humans achieve ***similar to or greater exposure*** to raxibacumab following a single 40 mg/kg IV dose compared to rabbits and monkeys receiving the same dose. As reported by the applicant, raxibacumab doses of 20 and 40 mg/kg IV provided a statistically significant survival benefit in monkeys and rabbits in the setting of therapeutic treatment of inhalation anthrax. The 40 mg/kg raxibacumab dose afforded 44% survival in rabbits and 64% survival in cynomolgus monkeys compared with 0% survival in placebo-treated animals. These efficacy findings in animals receiving the 40 mg/kg dose can be extrapolated to humans which achieve similar to or greater exposure following the same dose.

2.3. Intrinsic Factors

2.3.1. What intrinsic factors influence exposure and/or response, and what is the impact of any differences in exposure on efficacy or safety responses?

The effect of intrinsic factors on the pharmacokinetics of raxibacumab was explored in the applicant's population PK analysis. As anticipated based on experience with other monoclonal antibodies, weight is a significant covariate for raxibacumab PK (specifically on CL, V1, CLD2 and V2). The applicant has proposed weight-based dosing for raxibacumab, thus the impact of this covariate on efficacy and safety responses should be minimal.

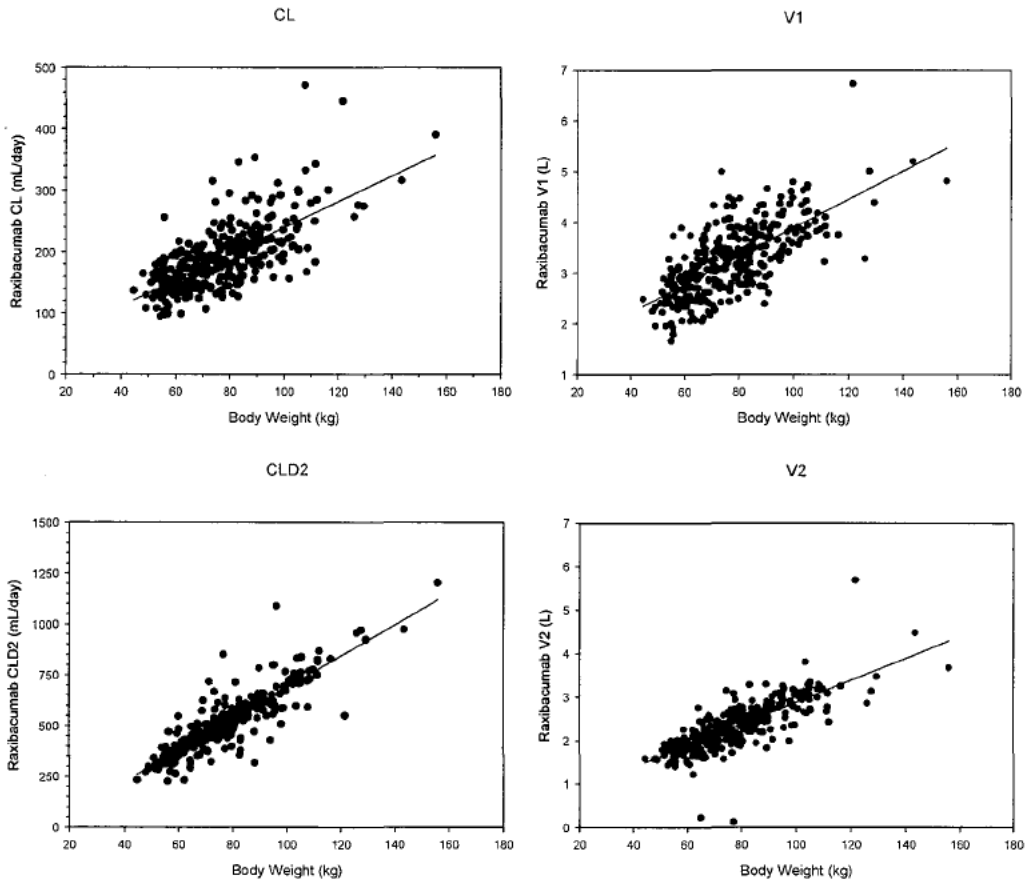
No other covariates were clinically significant.

2.3.2. Based upon what is known about exposure-response relationships and their variability and the groups studied, healthy volunteers vs. patients vs. specific populations (examples shown below), what dosage regimen adjustments, if any, are recommended for each of these groups? If dosage regimen adjustments are not based upon exposure-response relationships, describe the alternative basis for the recommendation.

2.3.2.1. Weight

As anticipated based on experience with other monoclonal antibodies, weight is a significant covariate for raxibacumab PK, specifically on CL, V1, CLD2 and V2 as depicted in Figure 2.3.2.1-1.

Figure 2.3.2.1-1 Effect of Body Weight on Raxibacumab PK Parameters



Based on simulated subjects receiving the same 40 mg/kg dose, Cmax increases by 61% over a weight range of 45 to 156 kg, while AUC0-∞ increases by 22%. Therefore, the applicant's proposed weight-based dosing of 40 mg/kg accounts for increases in clearance seen with higher body weights. The simulated exposures for a body weight of 156 kg are within the range of exposures observed in human subjects receiving raxibacumab 40 mg/kg in clinical trials.

2.3.2.2. Elderly

The age range for subjects included in the applicant's population analysis was 18 to 87 years. Age was not a significant covariate for raxibacumab PK in this analysis. Thus, no dosage regimen adjustments based on age are recommended.

2.3.2.3. Pediatric patients

Raxibacumab has not been studied in children (individuals under 18 years of age). The applicant has not proposed dosing recommendations specific to pediatric patients, but has recommended that raxibacumab should be administered to children only if the potential benefit justifies the potential risk.

2.3.2.4. Gender

In the population PK analysis, sex was a covariate for V1. Based on simulated subjects receiving the same 40 mg/kg dose, Cmax values were 11% higher in females than in males (886 and 983 μg/mL for males and females, respectively). Since CL was unaffected by sex, AUCinf did not differ in males and females. There were small differences in t1/2,α, t1/2,β, andVss due to sex (< 7%). Overall, sex does not have a clinically meaningful impact on raxibacumab PK and no dosage regimen adjustments based on sex are recommended.

2.3.2.5. Race

In the population PK analysis, Black race was a covariate for CLD2. Based on simulated subjects receiving the same 40 mg/kg dose, there were minimal differences in t1/2,α and t1/2,β (< 3% and < 1%, respectively). Overall, race does not have a clinically meaningful impact on raxibacumab PK and no dosage regimen adjustments based on race are recommended.

2.3.2.6. Renal impairment

The effect of renal impairment on raxibacumab PK has not been investigated. Raxibacumab clearance in humans ranges from 2.4 to 2.9 mL/day/kg, values much smaller than the glomerular filtration rate. Therefore, there is virtually no renal clearance of this monoclonal antibody.

2.3.2.7. Hepatic impairment

The effect of hepatic impairment on raxibacumab PK has not been investigated.

2.3.2.8. Pregnancy/Lactation

Raxibacumab has not been studied in pregnant or nursing women. It is not known if raxibacumab is excreted in human milk. The applicant has recommended that caution should be exercised when raxibacumab is administered to a nursing woman and that raxibacumab should be used during preganancy only if clearly needed.

2.3.3. Immunogenicity

Evaluation of a potential immunogenic response to raxibacumab in humans was conducted as part of all four human clinical studies (PAM-NH-01, HGS1021-C1063, HGS1021-C1064, and HGS1021-C1069).

2.3.3.1. What is the incidence (rate) of formation of the anti-product antibiodies (APA), including the rate of pre-existing antibodies, the rate of APA formation during and after treatment, time profiles and adequacy of the sampling schedule?

Raxibacumab was non-immunogenic in humans and there were no subjects with a positive antiraxibacumab antibody response in these studies following single or repeat (every 14 days or following ≥ 4 months) doses.

Table 2.3.3.1-1 summarizes the sampling schedule for anti-raxibacimab antibodies in each human study. The sampling schedules were adequate.

Table 2.3.3.1-1 Immunogenicity Sampling in Raxibacumab Clinical Trials

Study	*Sampling Schedule for Anti-Raxibacumab Antibodies*
PAM-NH-01	Day 0 predose, and Days 14, 28, and 56/exit
HGS1021-C1063*	• Double-dose groups (Group 1 and Group 3): blood samples were collected prior to dosing on Day 0 and Day 14, and on Days 42 and 70/End of Study. • Single-dose groups (Group 2 and Group 4): blood samples were collected prior to dosing on Day 0, and on Days 28 and 56/End of Study.
HGS1021-C1064*	• Group 1: Blood samples were collected on Days 0 (prior to raxibacumab dose), 33, and 61/End of Study. • Group 2 and Group 3: Blood samples were collected on Days 0 (prior to prior to raxibacumab dose), 28, and 56/End of Study.
HGS1021-C1069*	Screening, prior to raxibacumab dosing on Day 0, and on Days 28, 56, and 70 (End of Study)

*Subjects found to have a positive anti-raxibacumab antibody titer following raxibacumab administration were to have serum collected approximately 6 months after the dose for follow-up immunogenicity assessment.

2.3.3.2. Does the immunogenicity affect the PK and/or PD of the therapeutic protein?

Immunogenicity had no discernable impact on raxibacumab PK. Since raxibacumab was non-immunogenic, no impact on PD is anticipated.

2.3.3.3. Do the anti-product antibodies have neutralizing activity?

Since raxibacumab was non-immunogenic and no anti-product antibodies were detected, neutralizing activity could not be assessed.

2.3.3.4. What is the impact of anti-product antibodies on clinical efficacy?

Since raxibacumab was non-immunogenic, no impact on clinical efficacy is anticipated.

2.3.3.5. What is the impact of anti-product antibodies on clinical safety (e.g. infusion-related reactions, hypersensitivity reactions, cross-reactivity to endogenous counterparts, etc.)?

Premedication with diphenhydramine was mandated following the occurrence of infusion-related rashes in some subjects during Study HGS1021-C1064. Eight (9.1%) subjects experienced mild to moderate infusion-related rashes during this study: one subject in Group 1, six subjects in Group 2, and one subject in Group 3. There were no rashes of Grade 3 or higher. No other types of infusion reactions were observed and some of the rashes were observed outside the peri-infusion period. All raxibacumab-related rashes resolved within 1 to 15 days despite the persistence of significant raxibacumab levels during this interval. There were no constitutional symptoms evidenced by vital sign changes or abnormal laboratory values in the subjects with rash that would indicate a systemic hypersensitivity or anaphylactoid reaction beyond the localized cutaneous reactions. Since no anti-product antibodies were detected in these subjects, the impact on anti-product antibodies on clinical safety is expected to be minimal.

2.4. Extrinsic Factors

2.4.1. What extrinsic factors (drugs, herbal products, diet, smoking, and alcohol use) influence dose-exposure and/or -response and what is the impact of any differences in exposure on response? Based upon what is known about exposure-response relationships and their variability, what dosage regimen adjustments, if any, do you recommend for each of these factors?

No extrinsic factors have been shown to influence raxibacumab exposure-response. Therefore, no dosage adjustments for extrinsic factors are recommended.

2.4.2. Drug-Drug Interactions

2.4.2.1. Is there an in vitro basis to suspect in vivo drug-drug interactions?

There is no in vitro basis to suspect in vivo drug-drug interactions with raxibacumab.

2.4.2.2. Is the drug a substrate of CYP enzymes? Is metabolism influenced by genetics?

As a monoclonal antibody, raxibacumab is expected to be degraded into small peptides and individual amino acids. Therefore, it is not expected to be a substrate of CYP enzymes nor have metabolism influenced by genetics.

2.4.2.3. Is the drug an inhibitor and/or an inducer of CYP enzymes?

As a monoclonal antibody, raxibacumab is not expected to be an inhibitor and/or an inducer of CYP enzymes.

2.4.2.4. Is the drug a substrate and/or an inhibitor of P-glycoprotein transport processes?

As a monoclonal antibody, raxibacumab is not expected to be a substrate and/or an inhibitor of P-glycoprotein transport processes.

2.4.2.5. Are there other metabolic/transporter pathways that may be important?

Since raxibacumab is a monoclonal antibody, other metabolic/transporter pathways are not expected to be of importance.

2.4.2.6. Does the label specify co-administration of another drug (e.g., combination therapy in oncology) and, if so, has the interaction potential between these drugs been evaluated?

The DOSAGE AND ADMINISTRATION section of the proposed labeling suggests premedication with 25-50 mg diphenhydramine administered within 1 hour prior to raxibacumab infusion, due to the possibility of infusion and hypersensitivity reactions. The effect of diphenhydramine on raxibacumab PK was evaluated in the human clinical study HGS1021-1064. Of the 86 subjects enrolled in the study, 8 subjects experienced mild to moderate rashes in this study (n = 8/86 subjects). Cmax/Dose and AUC0-∞/Dose were calculated with subjects grouped according to whether they experienced a rash or not, and

further subsetted according to whether or not they were administered diphenhydramine. Diphenhydramine was administered within 60 minutes prior to raxibacumab administration in 61/86 subjects enrolled. Mean Cmax/Dose for subjects who experienced rash was lower (by approximately 17%) than those for the subjects who did not experience rash, while diphenhydramine use had little, if any, impact on Cmax/Dose; this difference was statistically significant ($p = 0.0334$), but none of the Bonferroni's multiple comparison post tests achieved significance ($p > 0.05$). Mean AUC0-∞/Dose for subjects who experienced rash tended to be lower (by approximately 11% to 21%) than those for the subjects who did not experience rash, while diphenhydramine use had no consistent impact on AUC0-∞/Dose; this difference was not statistically significant ($p = 0.3312$). Overall, raxibacumab exposure appears to be somewhat lower in the subjects who experienced rash, relative to those who did not. Raxibacumab exposure does not appear to differ between subjects who were administered diphenhydramine and those who were not.

This finding was confirmed in the population PK analysis where diphenhydramine use was assessed as a potential covariate, but was found to not have a significant impact on raxibacumab PK.

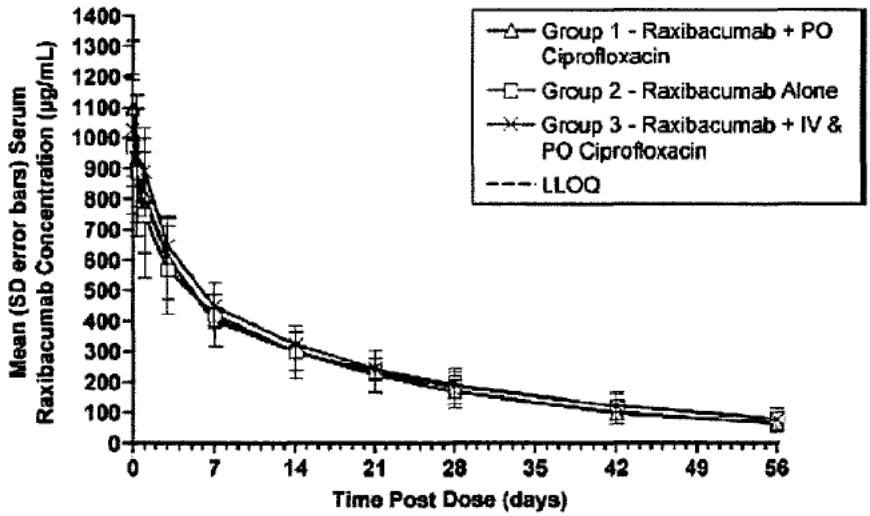
2.4.2.7. What co-medications are likely to be administered to the target patient population?

In the course of treatment of a *Bacillus anthracis* infection and in reponse to an anthrax-related emergency, raxibacumab will likely be administered concurrently with a fluroquinolone antibiotic active against *Bacillus anthracis*. To assess the influence of co-administration of the fluroquinolone antibiotic on dose-exposure, the applicant conducted a Phase 1 drug interaction study with ciprofloxacin in healthy subjects (HGS1021-C1064).

HGS1021-C1064 was an open-label study to evaluate the effect of raxibacumab on ciprofloxacin PK as well as the safety and PK of raxibacumab in combination with ciprofloxacin in healthy adult male and female subjects. Three treatment groups were evaluated. Group 1 received PO ciprofloxacin (500 mg Q12h, Days 0 to 7), with a single raxibacumab (40 mg/kg) dose IV on Day 5. Group 2 received a single raxibacumab (40 mg/kg) dose IV on Day 0. Group 3 received a single IV ciprofloxacin (400 mg) dose on Day 0 immediately followed by a single IV raxibacumab (40 mg/kg) dose, a 2nd IV ciprofloxacin (400 mg) dose 12 hours later, and then PO ciprofloxacin (500 mg Q12h, Days 1 to 7) for a total of 13 doses.

Serum concentration-time profiles for raxibacumab following single intravenous administration of 40 mg/kg with and without coadministration of ciprofloxacin PO (Group 1) or IV/PO (Group 3) in healthy, male and female subjects are presented in Figure 2.4.2.7-1. Pharmacokinetic parameters and results of statistical comparisons for raxibacumab following single intravenous administration of 40 mg/kg with and without coadministration of ciprofloxacin PO (Group 1) or IV/PO (Group 3) in healthy, male and female subjects are summarized in Table 2.4.2.7-1. Serum raxibacumab concentration-time profiles were very similar among treatment groups, with overlapping SD error bars. Statistically significant differences in PK parameters were not encountered for Group 3 (raxibacumab + IV ciprofloxacin) vs the raxibacumab alone group. Overall, exposure to ciprofloxacin appears to have no consistent or meaningful impact on raxibacumab PK.

Figure 2.4.2.7-1. Mean (± SD) Serum Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 40 mg/kg with and without Ciprofloxacin PO or IV in Healthy, Male and Female Subjects



Source: HGS1021-C1064 Pharmacokinetic Report, Section 3.5.2.4

Table 2.4.2.7-1 Summary of Raxibacumab PK Following a Single 40 mg/kg Raxibacumab IV Infusion Administered with or without PO or IV Ciprofloxacin

Parameter	Group 1[a] (n = 27)	Group 2[b] (n = 27)	Group 3 (n = 28)
Cmax (µg/ml)	1143.3 ± 169.5 (833.6 – 1517.8)	1020.3 ± 140.6 (766.2 – 1293.7)	1047.8 ± 180.3 (772.5 – 1458.4)
AUCinf (µg·day/ml)	14871.9 ± 3821.1 (5494.5 – 21116.6)	15845.8 ± 4333.5 (7614.9 – 25464.6)	16349.3 ± 4255.7 (7117.5 – 27465.2)
Half life (days)	19.8 ± 8.18 (4.27 – 45.8)	20.6 ± 6.54 (5.93 – 32.3)	21.5 ± 8.92 (6.53 – 42.9)
CL (ml/day/kg)	2.99 ± 1.38 (1.85 – 7.30)	2.73 ± 0.84 (1.57 – 5.24)	2.63 ± 0.82 (1.46 – 5.62)
Vss (ml/kg)	64.9 ± 11.9 (44.1 – 99.8)	69.7 ± 13.7 (45.6 – 106.7)	67.2 ± 12.6 (47.6 – 93.2)

Group 1, raxibacumab 40 mg/kg + cipro PO; Group 2, raxibacumab 40 mg/kg only; Group 3, raxibacumab 40 mg/kg + cipro IV/PO

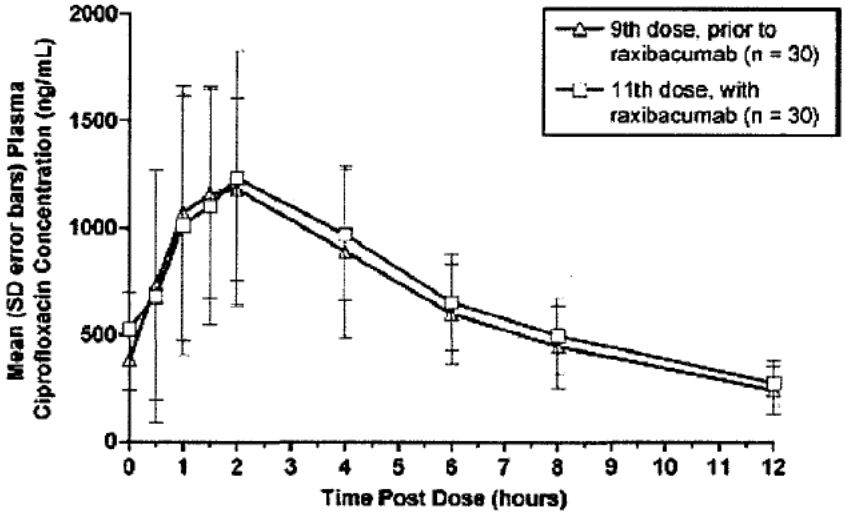
Data presented represents mean ± one standard deviation and (minimum – maximum)

[a] Subjects US003-000006 and US003-000028 excluded for receiving only partial doses of raxibacumab. Subject US003-00029 excluded for PK profile uncharacteristic of IV administration.

[b] Subject US003-000002 excluded for receiving only a partial dose of raxibacumab.

Plasma concentration-time profiles for ciprofloxacin following multiple oral administration with and without raxibacumab 40 mg/kg (Group 1) and following IV and oral administration with and without raxibacumab 40 mg/kg (Group 3) in healthy, male and female subjects are presented in Figures 2.4.2.7-2 and 2.4.2.7-3. Pharmacokinetic parameters ciprofloxacin following oral administration and IV followed by oral administration with and without coadministration of a raxibacumab 40 mg/kg single dose in healthy, male and female subjects are summarized in Table 2.4.2.7-2.

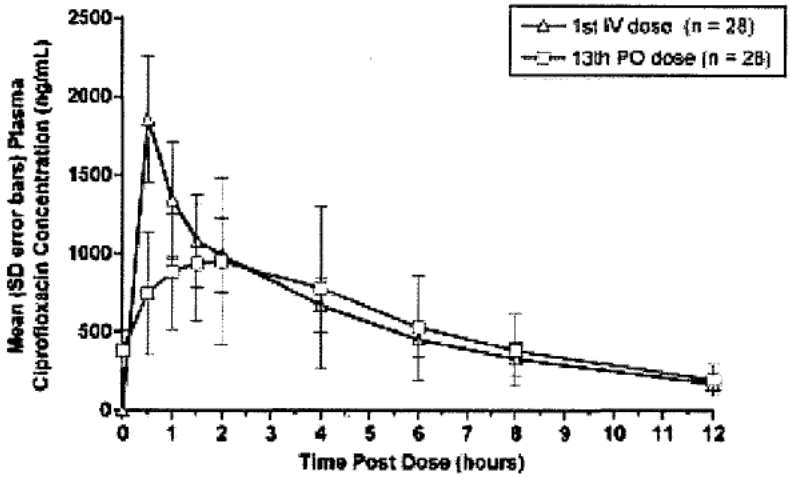
Figure 2.4.2.7-2. Mean (± SD) Plasma Concentration-Time Profiles for Ciprofloxacin Following Oral Administration with and without a Single IV Dose of Raxibacumab 40 mg/kg in Healthy, Male and Female Subjects (Group 1)



Graph depicts plasma ciprofloxacin concentrations following the 9th and 11th of 15 consecutive 500 mg PO ciprofloxacin doses given q12h; a single 40 mg/kg raxibacumab IV infusion dose was administered just prior to the 11th ciprofloxacin dose.

Source: HGS1021-C1064 Pharmacokinetic Report, Section 3.5.1.4

Figure 2.4.2.7-3. Mean (± SD) Plasma Concentration-Time Profiles for Ciprofloxacin Following Intravenous and Oral Administration with and without a Single Intravenous Dose of Raxibacumab 40 mg/kg in Healthy, Male and Female Subjects (Group 3)



Graph depicts plasma ciprofloxacin concentrations following the 1st of two 400 mg IV ciprofloxacin doses given 12 hours apart, and following the 13th consecutive 500 mg PO ciprofloxacin doses given q12h starting 12 hours after the 2nd IV dose; a single 40 mg/kg raxibacumab IV infusion dose was administered immediately after the 1st IV ciprofloxacin dose.

Source: HGS1021-C1064 Pharmacokinetic Report, Section 3.5.1.4

Table 2.4.2.7-2. Summary of Ciprofloxacin Pharmacokinetic Parameters Following Oral (Group 1) or IV Followed by Oral Administration (Group 3) with and without a Single IV Dose of Raxibacumab 40 mg/kg in Healthy, Male and Female Subjects

Parameter	Group 1		Group 3	
	PO without Raxibacumab (n = 30)	PO with Raxibacumab (n = 30)	IV with Raxibacumab (n = 28)	PO with Raxibacumab (n = 28)
Cmax (ng/ml)	NA	NA	1854 (402)	NA
Css,max (ng/ml)	1436 (519)	1419 (599)	NA	1195 (566)
AUCinf (ng·hr/ml)	NA	NA	8770 (1877)	NA
AUCtau (ng·hr/ml)	7694 (2680)	8151[a] (2673)	NA	6615 (3224)
Half life (hr)	4.74 (2.09)	5.25[a] (2.47)	4.53 (0.89)	4.62[b] (1.07)
CL or CL/F (L/hr)	72.4 (24.0)	66.8[a] (21.9)	47.6 (9.8)	91.1 (36.9)
Vss (L)	NA	NA	285.9 (725.9)	NA
Vz or Vz/F (L)	510.7 (340.3)	486.0[a] (212.8)	312.0 (933.7)	630.7[b] (308.6)

Data presented as mean (SD).
Group 1, raxibacumab 40 mg/kg + cipro PO
Group 3, raxibacumab 40 mg/kg + cipro IV/PO
[a] n = 29
[b] n = 27

Source: HGS1021-C1064 Pharmacokinetic Report, Section 3.5.1.4

Upon statistical comparison for Css,max and AUCτ, the 90% CI fell within the 80% to 125% range, demonstrating that for those primary parameters Dose 11 (with raxibacumab) was equivalent to Dose 9 (prior to raxibacumab administration). The secondary parameter CL/F also had a 90% CI, indicating equivalence before and after raxibacumab dosing. Based on the results obtained for Css,max and AUCτ, this analysis indicates that ciprofloxacin exposure is equivalent for ciprofloxacin administered alone and when administered with raxibacumab.

2.4.2.8. Are there any in vivo drug-drug interaction studies that indicate the exposure alone and/or exposure-response relationships are different when drugs are co-administered?

There are no in vivo drug interaction studies that indicate exposure or E/R relationships are different when the drugs are co-administered.

2.4.2.9. Is there a known mechanistic basis for pharmacodynamic drug-drug interactions, if any?

There is no known mechanistic basis for pharmacodynamic drug-drug interactions.

2.4.2.9.1. Are there any unresolved questions related to metabolism, active metabolites, metabolic drug interactions, or protein binding?

There are no unresolved issues related to metabolism, active metabolites, metabolic drug interactions or protein binding for this monoclonal antibody.

2.4.3. What issues related to dose, dosing regimens, or administration are unresolved and represent significant omissions?

Dose and concentration response for the endpoint of survival indicates potential to achieve higher response rates at higher doses. If future trials are conducted, higher doses (60 or 80 mg/kg) should be explored along with 40 mg/kg.

2.5. General Biopharmaceutics
Not applicable.

2.6. Analytical Section
This section summarizes the bioanalytical methods utilized to assess therapeutic protein concentrations and the formation of the anti-product antibodies. Details for the bioanalytical methodology used to determine ciprofloxacin plasma concentrations in Study HGS1021-C1064 are presented in the individual study review in Section 4.1.

2.6.1. What bioanalytical methods are used to assess the therapeutic protein concentrations? Briefly describe the methods and summarize the assay performance.

Raxibacumab concentrations in serum of rabbits, monkeys, and humans were quantitated by a validated electrochemiluminescence assay. **Raxibacumab is a recombinant, fully human, IgG1λ** monoclonal antibody and is expected to be degraded into small peptides and individual amino acids. Therefore, no metabolites were measured. The electrochemiluminescence (ECL) assay used to quantify raxibacumab concentrations measured total raxibacumab. Raxibacumab binds PA and inhibits its biological activity. The assay did not distinguish between raxibacumab bound to PA versus free raxibacumab. For purposes of comparing total exposure between rabbits, monkeys and humans for determination of an effective human dose, this method is acceptable.

Assay specifics and performance are summarized in Table 2.6.1-1.

Table 2.6.1-1. Qualification and Performance of Bioanalytical Assays Used to Quantify Raxibacumab Concentrations

Method (Studies)	TR-21-07-016 (Study 682-G005758)	TR-21-07-057 (Study 724-G005829)	TR-21-07-015 (Studies HGS1021-C1063, HGS1021-C1069, HGS1021-C1064)
Description	Rabbit PK assay	Monkey PK assay	Human PK assay
LOD (ng/mL)	< 0.005	0.0041	< 0.0046
LLOQ (ng/mL)	0.027	0.038	0.03 (0.8 in undiluted serum)
ULOQ (ng/mL)	65	65	70 (1.6 in undiluted serum)
Linearity Range (ng/mL)	0.027 – 65	0.038 – 65	0.03 - 70
Dilutional linearity range (µg/mL)	0.75 - 1500	0.94 - 1300	0.8 - 1600
Accuracy (%)	90.9 – 118.1	91.1 – 122.7	83.9 – 96.0
Intra-day precision (%CV)	0 – 5.9	3.5 – 8.9	4.0 – 9.0
Inter-day precision (%CV)	0 – 5.7	0.98 – 4.9	3.0 – 17.0
Relative error (%)	2.3 – 12.0	3.2 – 13.0	3.0 – 19.0

LOD, limit of detection
LLOQ, lower limit of quantitation
ULOQ, upper limit of quantitation

2.6.1.1. What is the range of the standard curve? How does it relate to the requirements for clinical studies? What curve fitting techniques are used?

The range of the standard curve was determined using accuracy and precision data from all raxibacumab concentrations. The highest and lowest values that meet the acceptable total error were assigned as the ULOQ and LLOQ and thus define the range (see Table 2.6.1-1). The standard curve was obtained by fitting the experimental data from raxibacumab standards to a 4-parameter logistic (4PL) function with concentration of analyte (X) as the independent variable and absorbance (Y) as the dependent variable:

$$y = ((A - D)/(1 + (x/C)^{B})) + D$$

In this expression, A refers to the left (lower) asymptote, D refers to the right (upper) asymptote, C refers to the analyte concentration that produces 50% of the maximum response and B is a scale parameter related to the shape and steepness of the curve. The curve fit was performed using variable weighting.

The raxibacumab assays were further characterized for parameters of sample analysis to demonstrate the applicability of the assay system to quantitation of raxibacumab in rabbit, and human serum within the concentration ranges expected in PK studies. Summaries of this dilutional qualification are as follows:

- For rabbits, pooled rabbit serum spiked with raxibacumab at concentrations of 1500, 1250, 1000, 550, 225, 20, 5, 1, 0.85, 0.75, 0.5, and 0.3 µg/mL were analyzed at a dilution of 1:24,000. The assay was linear within this tested concentration range ($R^2 = 0.9984$, $p < 0.0001$). Data from the lowest concentrations, 0.5 and 0.3 µg/mL, fell outside the linear range of the standard curve. Thus, 0.75 µg/mL was deemed the lowest

concentration in human serum that may be quantified in rabbit serum. Accuracy was also determined by measuring the concentration of raxibacumab in diluted pooled rabbit serum samples spiked with raxibacumab at concentrations of 20 to 5200 µg/mL selected to cover the high-concentration range expected in some PK studies. The calculated accuracies of the spiked samples ranged from 100.3% to 111.3%.

- For the monkey assay, pooled monkey serum spiked with raxibacumab at concentrations of 1400, 1300, 1100, 640, 255, 25, 2.4, 0.94, 0.72, 0.54 and 0.26 µg/mL were analyzed at a dilution of 1:24,000. The assay was linear within this tested concentration range ($R^2 = 0.9942$, $p < 0.0001$). Data from the lowest concentrations, 0.72, 0.54 and 0.26 µg/mL, fell outside the linear range of the standard curve. Consequently, the data from these concentrations were not included in determining precision, accuracy and linearity. The lowest neat spike concentration that met acceptance criteria (CV ≤ 0%, relative error ≤ 0% and total error ≤ 0%) was 0.94 µg/mL; this was deemed the lowest concentration that may be quantified. The highest neat spike concentration that met acceptance criteria (CV ≤ 0%, relative error ≤ 0% and total error ≤ 30%) was 1300 µg/mL; this was deemed the highest concentration that may be quantified.
- For the human assay, pooled normal human serum spiked with raxibacumab at concentrations of 1600, 1200, 600, 240, 22, 11, 2.3, 1.2, 0.92, 0.8, 0.5, and 0.26 µg/mL were analyzed at a dilution of 1:24,000. The assay was linear within this tested concentration range ($R^2 = 0.9991$, $p < 0.0001$). The two lowest spiked concentrations (0.5 and 0.26 µg/mL) did not meet the accuracy and precision criteria and were therefore not included in the linear regression analysis. The lowest neat spike concentration that met the criteria (CV ≤ 20%, relative error ≤ 20%, and total error ≤ 30%) was 0.8 µg/mL; thus, this was deemed the lowest concentration in human serum that may be quantified.

The ranges of standard curves for the rabbit, monkey and human assays are adequate for purposes of determining serum concentrations of raxibacumab in the pivotal animal and clinical studies.

2.6.1.2. What are the lower and upper limits of quantification (LLOQ/ULOQ)?

Refer to Table 2.6.1-1 for the LLOQ and ULOQ for each raxibacumab assay.

2.6.1.3. What are the accuracy, precision, and selectivity at these limits?

Refer to Table 2.6.1-1 for the accuracy and precision for each raxibacumab assay.
Assay specificity was determined from the assessment of a series of serum samples spiked with an unrelated human monoclonal antibody (belimumab). All samples resulted in measurements below the LLOQ and LOD.

2.6.1.4. What is the sample stability under the conditions used in the study (long-term, freeze-thaw, sample-handling, sample transport, autosampler)?

Raxibacumab sample stability was assessed by evaluating raxibacumab in 100% pooled serum under various storage conditions: (1) a control group stored at -80°C; (2) a group that was stored for 7 days at 4°C; (3) a group that was frozen at -80°C and thawed 5 times; (4) a group that was stored for 1 day at 22°C; and (5) a group that was stored at -20°C. The mean concentration of each storage condition was compared with the mean concentration of the control condition. Each stability sample was tested by each operator on 2 plates over 12 total measurements for the rabbit assay and 6 measurements for the human and monkey assays. All of the conditions tested were found to be within 90-110% of the control mean.

2.6.1.5. What is the QC sample plan?

Three positive controls with known concentrations of raxibacumab (44 ng/mL, 0.92 ng/mL and 0.043 ng/mL for the rabbit assay; 49 ng/mL, 0.98 ng/mL and 0.054 ng/mL for the monkey assay; 70 ng/mL, 1.6 ng/mL and 0.08 ng/mL for the human assay) were analyzed on each plate. Four out of six positive controls were required to measure within 25% (75-125%) of the nominal concentration. At least one determination of each of the duplicate controls at each concentration was required to be within 25% (75-125%) of the nominal concentration.

2.6.2. What bioanalytical methods are used to assess the formation of the anti-product antibodies? Briefly describe the methods and summarize the assay performance, including sensitivity, specificity, precision, cut point, interference and matrix, etc.

2.6.2.1. What is the performance of the binding assay(s)?

The method for detection of anti-raxibacumab antibodies in human serum in Studies HGS1021-C1063, HGS1021-C1064, and HGS1021-C1069 consists of two ECL assays: a screening assay and a confirmatory (inhibition of binding) assay. In the screening assay, undiluted serum samples in duplicate wells are acidified with hydrochloric acid for one hour to dissociate raxibacumab from anti-raxibacumab antibodies. Acidified serum samples are then neutralized in the presence of raxibacumab-biotin (capture) and raxibacumab-Sulfo-TAG (detector) and allowed to re-equilibrate for four hours at room temperature in a streptavidin-coated assay plate. Following re-equilibration, plates are washed and read for ECL counts. A rabbit polyclonal antibody is used as a positive control. The inhibition of binding assay is performed similarly to the screening assay with the exception that the samples are tested in parallel with excess unlabeled raxibacumab. The excess unlabeled raxibacumab is added in the capture/detector solution so that samples are simultaneously exposed to excess competing drug and labeled capture/detector.

To determine assay specificity, two irrelevant polyclonal antibodies raised against wtPA (including belimumab) were tested. There was no confirmed binding of these two antibodies at 4 µg/mL. A series of anti-raxibacumab control spikes was prepared in pooled human serum at 1, 0.5, 0.25, 0.125, 0.0625, and 0.03125 µg/mL and variance component analysis was performed for precision analysis. The overall %CV of spikes above the LOD was 7.9-10.4%. To determine the LOD, anti-raxibacumab polyclonal antibody was spiked in pooled human serum at 1, 0.5, 0.25, 0.125, 0.1, 0.0625, and 0.03125 µg/mL, along with unspiked pooled human serum as the negative control. The overall LOD was estimated as the lowest anti-raxibacumab polyclonal antibody concentration that produced a mean ECL where the lower bound of the 95% prediction interval exceeds the cut-point. The lowest anti-raxibacumab antibody concentration that met that criterion was 0.1 µg/mL, thus the LOD was 0.1 µg/mL. Linearity was characterized using the same control spikes used for analysis of precision. Linear regression analysis was performed using the spiked concentration of the control spikes as the independent variable and the ECL count as the dependent variable. The assay provides a linear response within the tested concentration range. The linearity was observed with high degree of correlation ($R^2 = 0.999$) and confidence ($p < 0.0001$).

The method for determining the cut point (above which samples are deemed potentially positive) is called the Progressive Cut Point method. The method was adopted to stabilize the variances of the original qualification data and the % drop values in the confirmatory assay. This is accomplished through the use of a logarithm transformation of the original data which takes the data from a log distribution to a Gaussian distribution. When the data from samples with inhibitor are divided by the data from samples without inhibitor, the resulting ratio (% drop) also has a

near-Gaussian distribution. The benefit of transforming the data is that standard parametric statistics can be applied to determine the cut point. The ratio of the log of the sample mean ECL count to the log of the negative control mean ECL count is calculated for each sample. The upper 95% prediction interval (PI) of all ratios was calculated and samples above that value were excluded and the upper 95% PI of the remaining ratios was set as the cut point which was determined to be 1.046.

The positive control (PC) was used to confirm the stability of samples stored in different conditions by comparing ECL counts after storage. Four conditions were tested: a) the control PC stored at -80°C and thawed at 4°C prior to analysis; b) the PC removed from -80°C and stored at -20°C overnight prior to analysis; c) the PC removed from -80°C and stored at 4°C for 24 hours prior to analysis; and d) a PC following freeze thaw cycles where the PC was removed from -80°C, thawed at 4°C for 1 hour, returned to -80°C for 1 hour, then thawed at 4°C prior to analysis. Three operators ran 2 replicates of each condition, for a total of 6 measurements of each. None of the tested conditions were significantly different from the control condition.

Assay specifics and performance are summarized in Table 2.6.2-1.

Table 2.6.2-1. Qualification and Performance of Bioanalytical Assay Used to Detect Anti-Raxibacumab Antibodies in Human Serum

Qualification Parameter	Assay Result
LOD (µg/mL)	0.1
Linearity Range (µg/mL)	0.03125 – 1 (R^2 = 0.9999, p < 0.0001)
Specificity	No confirmed binding of two irrelevant antibodies at 4 µg/mL
Precision (%CV)	7.9-10.4
Assay Cut-Point	1.046 (ratio of log-sample to log-negative control)
Threshold for Specific Drop in Confirmatory Assay	4.5%
LOD in the presence of drug (raxibacumab)	12.5 µg/mL anti-raxibacumab antibody @ 300 µg/mL drug 3.125 µg/mL anti-raxibacumab antibody @ 150 µg/mL drug 1.56 µg/mL anti-raxibacumab antibody @ 37.5-75 µg/mL drug 0.25 µg/mL anti-raxibacumab antibody @ 5 µg/mL drug 0.125 µg/mL anti-raxibacumab antibody @ 0.625-2.5 µg/mL drug

LOD, limit of detection

2.6.2.2. What is the performance of the neutralizing assay(s)?

Since raxibacumab was non-immunogenic and no anti-product antibodies were detected, neutralizing activity was not assessed.

3. LABELING RECOMMENDATIONS

The following labeling contains clinical pharmacology reviewer recommendations previously incorporated and communicated to the Applicant (i.e. recommendations are unmarked).

21 Page(s) of Draft Labeling have been Withheld in Full as b4 (CCI/TS) immediately following this page

4. APPENDICES

4.1. Individual Study Reviews

AB50409.INF.0.040
Raxibacumab Pharmacokinetics and Protective Antigen Kinetics During the Evaluation of Raxibacumab Efficacy as Therapeutic Treatment Against Inhalation Anthrax in the Monkey Model (Battelle Study No. 724-G005829)

OBJECTIVES:

- To determine the pharmacokinetics (PK) of raxibacumab following a single intravenous (IV) raxibacumab dose in monkeys with inhalation anthrax
- To determine the kinetics of *Bacillus anthracis* protective antigen (PA) in monkeys with inhalation anthrax

STUDY DESIGN:
Forty juvenile (less than 5 years of age) cynomolgus monkeys (20 males and 20 females) weighing 2.3 to 5.1 kg at randomization were randomized to treatment groups, challenge day, and challenge order. Animals were randomized by sex into two raxibacumab dose groups of 14 animals each and one control group of 12 animals, and into three aerosol challenge sets of 14, 13, and 13 animals. The three sets of monkeys were aerosol challenged with a targeted 200 x LD_{50} (1.24×10^7 spores) inhaled dose of *B. anthracis* (Ames strain) spores via a head-only inhalation exposure chamber on three staggered challenge days (Day 0). Following spore challenge, animals were frequently monitored for PA toxemia (every 6 hours from 24 to 54 hours post-spore challenge). Individual monkeys were treated with 1 dose of IM diphenhydramine (1 mg/kg), followed within five minutes by one bolus IV injection of raxibacumab (40 mg/kg or 20 mg/kg according to group assignment) or raxibacumab vehicle (placebo, 1.0 mL/kg) immediately after the first detection of measurable serum PA.

FORMULATIONS:
Raxibacumab (Lot No. 71044) and raxibacumab vehicle (Lot No. 71043) were produced at HGS, and were stored at 2-8°C prior to use. Raxibacumab and vehicle were supplied as ready-to-use sterile liquid formulations. Raxibacumab vehicle was raxibacumab formulation buffer (0.13 mg/mL citric acid, 2.8 mg/mL sodium citrate, 10 mg/mL sucrose, 18 mg/mL glycine, 0.2 mg/mL polysorbate 80, pH 6.5). Raxibacumab was provided as a solution (50 mg/mL) in the formulation buffer. During dosage periods, raxibacumab and vehicle were stored on ice. Prior to administration, raxibacumab was aseptically diluted to concentrations of 40 mg/mL and 20 mg/mL in formulation buffer. The diluted raxibacumab soultions were sealed in sterile dosing vials labeled X and Z. Vehicle control (formulation buffer) was aseptically transferred into sterile dosing vials labeled Y. Dilutions of raxibacumab and transfer of vehicle control were performed the day prior to challenge and the vials were stored at 2 to 8°C prior to dosing.

PHARMACOKINETIC ASSESSMENTS:
Blood specimens for determination of serum raxibacumab concentrations were collected from all monkeys at 3 days prior to spore challenge; just prior to raxibacumab or vehicle dosing; at 5 minutes after dosing; at 12 and 24 hours after dosing; and at 3, 5, 8, 14, and 28 days after dosing. When feasible, a terminal blood sample was taken just prior to euthanasia for animals that were judged to be moribund.

Blood specimens for serum PA kinetics were collected from all monkeys at 3 days prior to spore challenge as well as at 24, 30, 36, 42, 48, and 54 hours after spore challenge, unless treatment occurred prior to the collection time. Additional blood specimens were collected just prior to raxibacumab or vehicle dosing; at 5 minutes after dosing; at 12 and 24 hours after dosing; and at 3, 5, 8, 14, and 28 days after dosing. When feasible, a terminal blood sample was taken just prior to euthanasia for animals that were judged to be moribund.

BIOANALYTICAL ANALYSIS:
Serum samples were analyzed for raxibacumab using an ECL-based assay. The raxibacumab in diluted serum samples binds to the biotinylated-PA on a streptavidin-coated plate and is detected by the addition of goat anti-human antibody labeled with MSD SULFOTAG™, an ECL label. The concentration of raxibacumab in serum samples is interpolated from a reference standard curve. The lower limit of quantitation (LLOQ) is 940 ng/mL of raxibacumab in 100% monkey serum.

Total PA (free + raxibacumab-bound) concentrations in serum samples were determined using an ECL-based bridging assay. In brief, undiluted serum samples in duplicate wells are combined with monkey polyclonal antibody (pAb) anti-PA-biotin (capture) and monkey pAb anti-PA-SULFO-TAG (detector) and allowed to equilibrate in a streptavidin-coated assay plate. Following equilibration, plates are washed and read for ECL counts. The concentration of PA in serum samples and controls is interpolated from a reference standard curve. The LLOQ is 0.65 ng/mL of PA in 100% monkey serum.

Samples were also assayed for anti-PA antibody concentrations and toxin neutralization activity (TNA) titers. Anti-PA Ab concentrations in serum samples were determined using an enzyme-linked immunosorbent assay. In brief, anti-PA Ab from diluted serum samples in duplicate wells are captured on wild-type PA coated plates, with detection using horseradish peroxidase (HRP)-conjugated goat anti-human IgG. The bound HRP activity is quantitated by the color conversion of tetramethylbenzidine, with absorbance measured at 450 nm. The concentration of anti-PA Ab in serum samples and controls is interpolated from a reference standard curve. The LLOQ is < 600 ng/mL of anti-PA Ab in 100% monkey serum. TNA titers in serum samples were determined using a cell killing assay. In brief, this assay is based on the principle that the neutralizing antibodies to PA bind to, and prevent, PA from shuttling another anthrax toxin component, lethal factor (LF, a bacterial metalloprotease), into target J744A.1 macrophages. In the absence of neutralizing antibodies, lethal toxin complexes enter the cells causing cell death. Inhibition of PA binding is correlated with increased cell viability and is used to determine the relative titer of serum samples compared to a known positive control. The LLOQ is < 52 titer in 100% monkey serum.

PHARMACOKINETIC/PHARMACODYNAMIC/STATISTICAL ANALYSIS:
PK analyses of raxibacumab concentration-time profiles for all raxibacumab-dosed monkeys were conducted using population analysis techniques (mixed effect modeling utilizing NONMEM). Inspection of the individual serum raxibacumab concentration-time profiles following IV dosing revealed a profile that was multiphasic; hence, 2- and 3-compartment models with 1st order elimination from the central compartment were evaluated. Following identification of the most appropriate PK structural model and error models for the PK parameters, body weight, sex, age, treatment group, size of spore challenge, duration of spore challenge, survival time, survival status, time to 1st bacteremia by culture, and bacteremia outcome at each collection time were evaluated as potential covariates for the PK parameters. Selection between competing models was based on goodness of fit criteria (reduction in objective function [-2 log likelihood, or

-2LL] value, diagnostic plots). PK parameter estimates for individual animals were derived from the final model.

Kinetic analyses of total PA concentration-time profiles for all monkeys were conducted using population analysis techniques, with the NONMEM software. Body weight, sex, age, raxibacumab dose, size of spore challenge, survival status, time to 1st bacteremia by culture, and raxibacumab PK parameters were evaluated as potential covariates. Prior to modeling, the serum PA results were examined for consistency with the model assumptions and for the presence of outliers. Concentrations below the LLOQ were excluded. Generally, serum PA concentrations measured near or after the time of death appeared unreasonably high. It is unknown if these high concentrations are due to changes in bacterial growth or to changes in the animal's physiology in the period near the time of death. Since the terminal specimen results appeared to be inconsistent with the model, as well as inconsistent with the remainder of the animals' profiles, and may have differed due to factors that could not be accounted for in the model, the terminal specimen results were excluded from the PA kinetics analyses.

In vitro, the dissociation equilibrium constant (Kd) for raxibacumab was estimated to be 2.78 nM. Using the in vitro estimate of Kd, the percentage of serum PA bound following raxibacumab dosing was calculated from the following equation:

$$\%\text{Bound} = 100\left(K_d^{-1}C_{raxi} \Big/ 1 + K_d^{-1}C_{raxi}\right)$$

where Craxi is the serum raxibacumab concentration, expressed as nM.

Serum raxibacumab concentration-time data, serum PA concentration-time data, individual raxibacumab PK parameters, individual PA kinetic parameters, serum anti-PA Ab concentrations, and serum TNA titers were summarized using descriptive statistics. A logistic regression model was used to describe the relationship between the survival probability and maximum serum raxibacumab concentration (Cmax) or area under the serum raxibacumab concentration-time curve to infinite time (AUC0-∞).

RESULTS:

A summary of sex, age and body weight for the monkeys is presented in Table 1.

Table 1. Baseline Characteristics of Monkeys in Study 724-G005829

Group	Number of Animals (N)	Age (yr)	Weight (kg)	Sex
1 (Raxibacumab 40 mg/kg)	14	3.8 (3.0 – 4.3)	3.5 (2.3 – 4.4)	7 Females 7 Males
2 (Raxibacumab 20 mg/kg)	14	3.6 (2.8 – 4.3)	3.4 (2.3 – 5.1)	7 Females 7 Males
3 (Placebo)	12	3.9 (3.0 – 4.6)	3.7 (2.6 – 4.8)	6 Females 6 Males

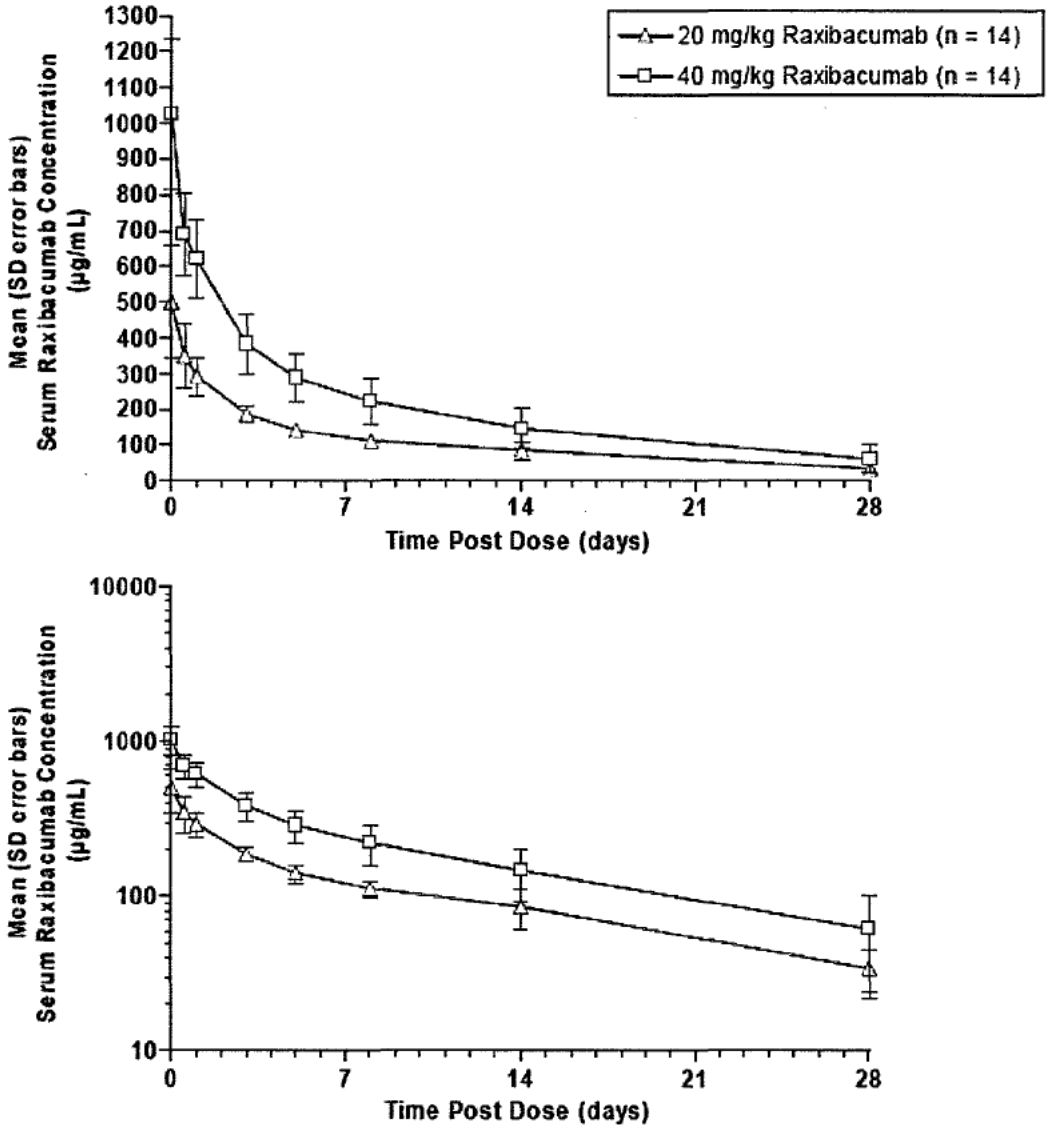
Data presented as mean (range).

Source: Study Report AB50409-INF-0-040, Appendix 1

Raxibacumab Serum Pharmacokinetics

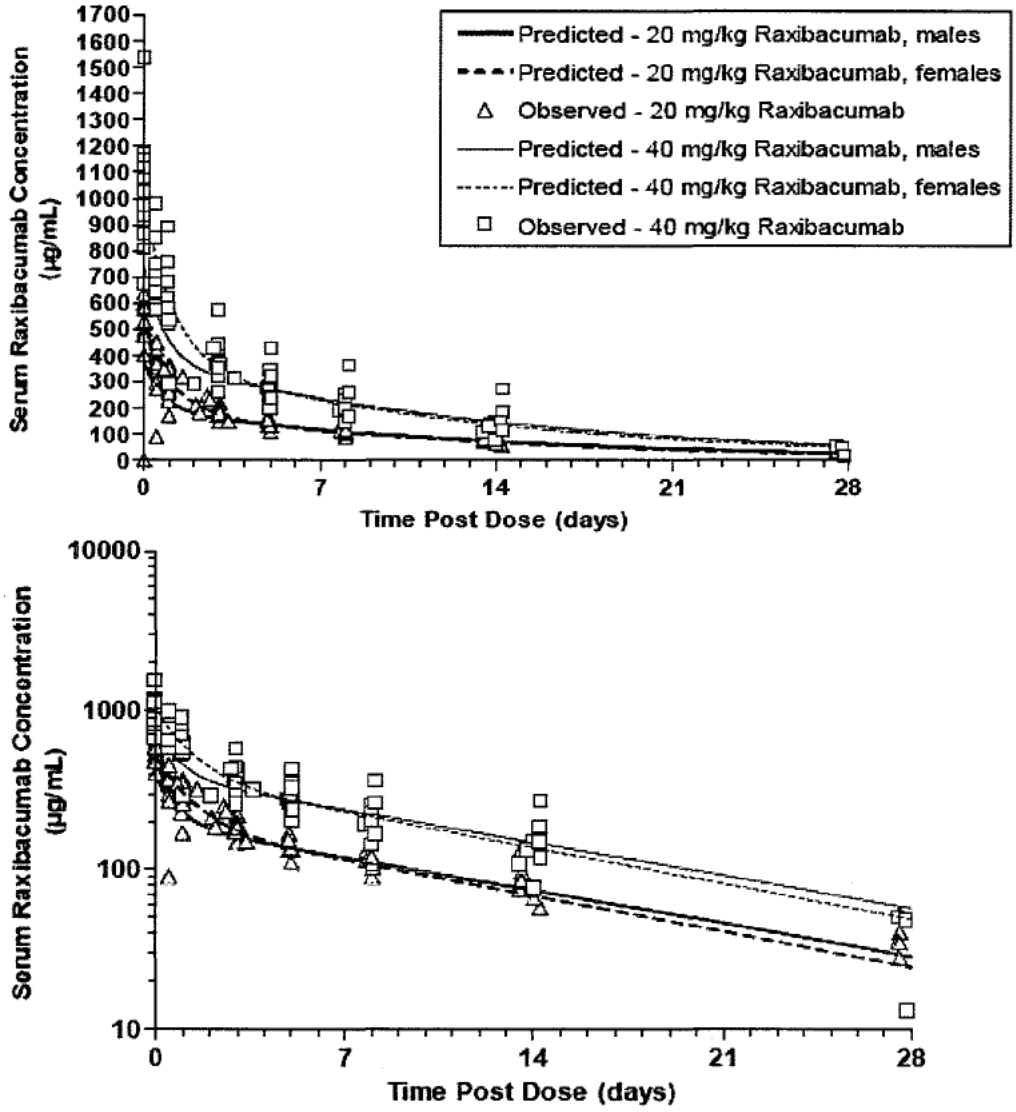
Serum concentration-time profiles for raxibacumab following single intravenous administration of 20 mg/kg and 40 mg/kg in spore-challenged cynomolgus monkeys are presented in Figure 1. Individual observed serum raxibacumab concentrations versus the population average serum raxibacumab concentration-time profiles are presented in Figure 2. Population predicted concentrations versus observed serum raxibacumab concentrations are presented in Figure 3. PK parameter estimates are summarized in Table 2. Serum raxibacumab concentrations were best fit to a 2-compartment open model with 1st-order elimination from the central compartment. Inter-individual variability for clearance (CL), volume of distribution for the central (1st) compartment (V1), and volume of distribution for the peripheral (2nd) compartment (V2) were modeled using exponential terms. Inter-individual variability for intercompartmental clearance (CLD2) was not modeled. Body weight was found to be a significant covariate for V2, while sex was a significant covariate for V1 and CLD2. Other factors assessed (age, treatment group, size of spore challenge, duration of spore challenge, survival time, survival status, time to 1st bacteremia by culture, and bacteremia outcome at each collection time) were not significant covariates accounting for inter-individual differences in PK. The lack of difference in PK between treatment groups is consistent with linear PK over the dose range studied.

Figure 1. Mean (± SD) Serum Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Cynomolgus Monkeys (Linear and Log Scale)



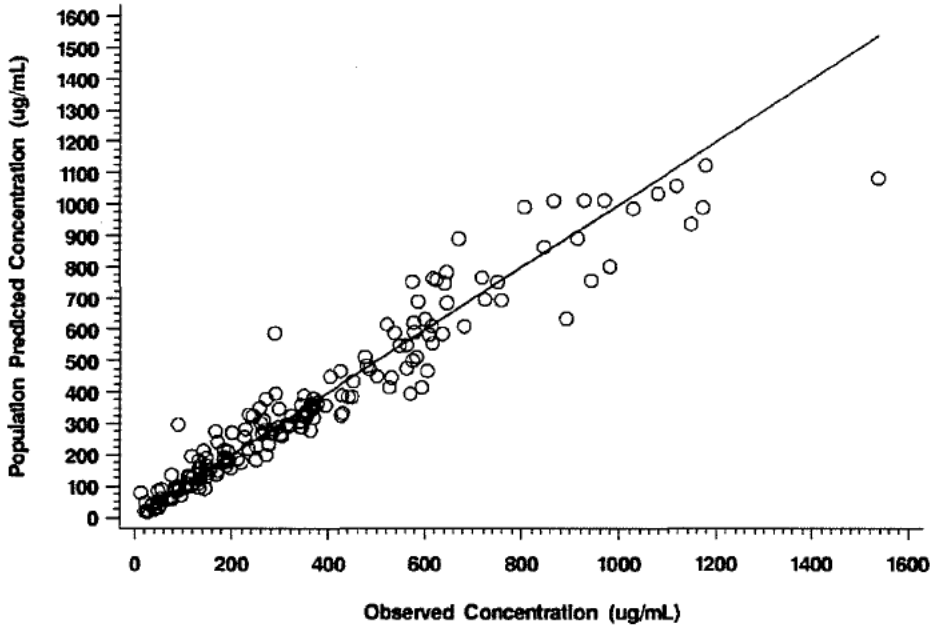
Source: Study Report AB50409-INF-0-040, Section 5.1
N = 14 per dose group

Figure 2. Predicted Population Average Serum Raxibacumab Concentration-Time Profiles and Individual Observed Serum Raxibacumab Concentrations Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Cynomolgus Monkeys (Linear and Log Scale)



Source: Study Report AB50409-INF-0-040, Section 5.1
N = 14 per dose group

Figure 3. Population Predicted Serum Raxibacumab Concentrations Versus Individual Observed Serum Raxibacumab Concentrations Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Cynomolgus Monkeys



Source: Study Report AB50409-INF-0-040, Appendix 8

Table 2. Summary of Model-Derived Raxiacumab Pharmacokinetic Parameters Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Cynomolgus Monkeys

Primary Parameters	Mean	CV%
V_1 (mL)	160	15.8
Effect of sex on V_1 (mL)[1]	$V_1 = 159.8 \times (1 + (-0.25202 \times sex))$	
Males	160	
Females	120	
CL (mL/day)	20	25.9
V_2 (mL)	125[1]	16.0
Effect of weight on V_2 (mL)	$V_2 = 125.02 + (43.545 \times (weight - 3))$	
At 2.5 kg	103	
At 2.75 kg	114	
At 3.25 kg	136	
At 3.5 kg	147	
CLD_2 (mL/day)	87	NA
Effect of sex on CLD_2 (mL/day)[1]	$CLD_2 = 86.789 \times (1 + (-0.44941 \times sex))$	
Males	87	
Females	48	

Secondary Parameters	Mean for 20 mg/kg		Mean for 40 mg/kg	
	Males	Females	Males	Females
C_{max} (µg/mL)[2]	375	751	502	1004
$AUC_{0-\infty}$ (µg·day/mL)[2]	2966	5931	2966	5931
$t_{1/2,\alpha}$ (days)	0.53	0.79	0.53	0.79
$t_{1/2,\beta}$ (days)	10.22	9.40	10.22	9.40
MRT (days)	14.08	12.09	14.08	12.09
V_{ss} (mL)	285	245	285	245

Abbreviations: CV%, coefficient of variation; V_1, volume of distribution for the central compartment; CL, clearance; V_2, volume of distribution for the peripheral compartment; CLD_2, intercompartmental clearance; NA, not applicable; C_{max}, maximum serum drug concentration; $AUC_{0-\infty}$, area under the serum drug concentration-time curve from time 0 to infinite time; $t_{1/2,\alpha}$, elimination half-life for the 1st phase; $t_{1/2,\beta}$, elimination half-life for the 2nd (terminal) phase; MRT, mean residence time; V_{ss}, volume of distribution at steady-state.

1 For sex coded as 0 = male and 1 = female.

2 Assuming a typical monkey weighing 3 kg.

Source: Study Report AB50409-INF-0-040, Section 5.1

Following IV raxibacumab administration, V1, at 160 and 120 mL for males and females, respectively (53 and 40 mL/kg for a 3 kg male and female animal, respectively), is similar to the plasma volume (44.8 mL/kg). The steady-state volume of distribution (Vss) is nearly 2-fold greater than V1, at 285 and 245 mL for males and females, respectively. These results suggest that although distribution of raxibacumab may initially be restricted to the plasma volume, raxibacumab does subsequently distribute to tissues. Both V1 and V2 increase with increasing weight. The disappearance of raxibacumab from serum is multiphasic, with an initial phase elimination half-life (t1/2,α) of 0.5 to 0.8 days. The terminal phase elimination half-life (t1/2,β) is 10.2 and 9.4 days for males and females, respectively. The mean CL of raxibacumab was

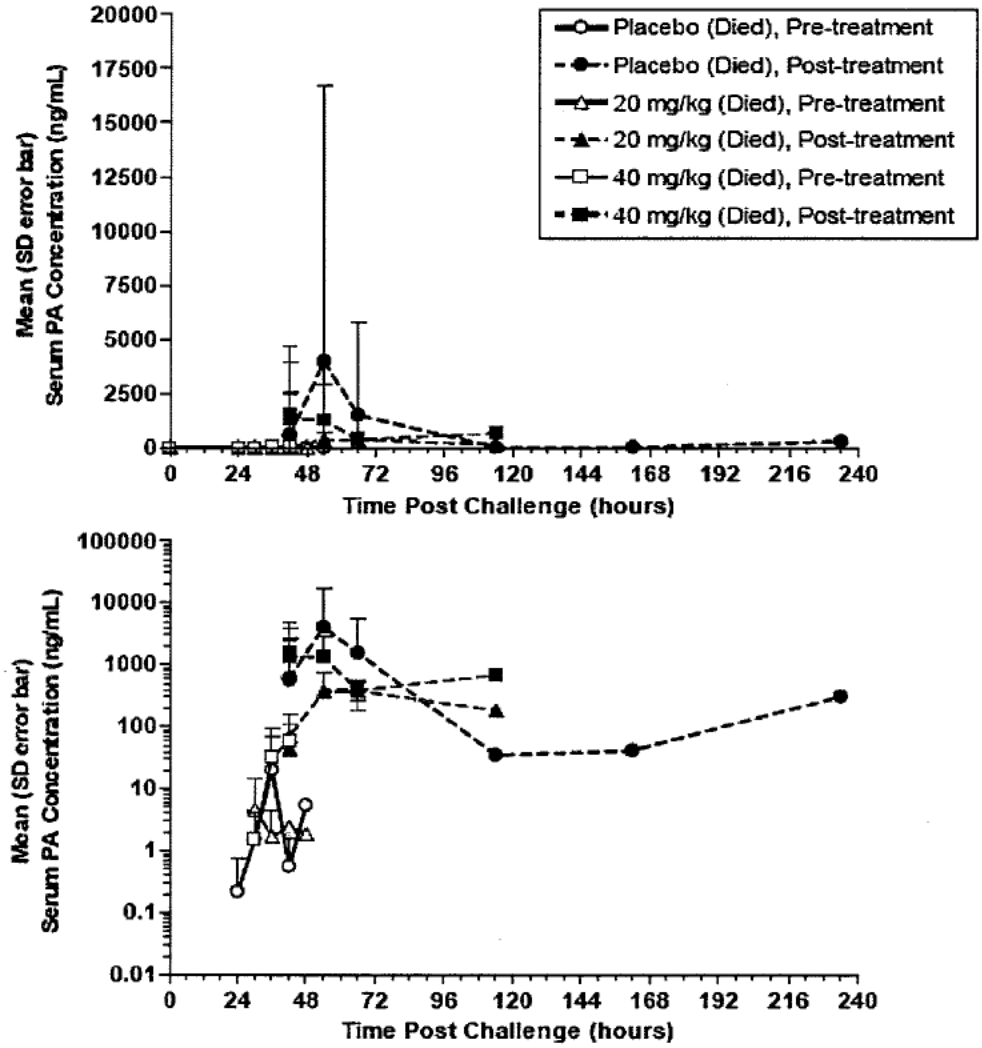
20 mL/day (6.7 mL/day/kg for a 3 kg monkey), which is much less than the glomerular filtration rate (2995 mL/day/kg), indicating that, as expected, there is virtually no renal clearance of this monoclonal antibody. Inter-individual variability in raxibacumab PK was low, with CV% of 26% or less for the primary PK parameters. As noted previously, for CLD2 inter-individual variability was not estimated. However, CLD2 is lower for female monkeys (48 mL/day) than for male monkeys (87 mL/day).

Protective Antigen (PA) Kinetics

Mean (± SD) serum PA concentration-time profiles for animals that died within each treatment group are illustrated in Figure 4. Examination of the profiles for the individual animals that died in all treatment groups revealed serum PA concentration-time profiles generally followed the pattern of rise then plateau, with the exception that some animals died prior to attaining the plateau phase of the serum PA concentration-time profile. Although the mean serum PA concentrations for the monkeys that died differ among the treatment groups, there was substantial overlap of the SD bars among the 3 treatment groups.

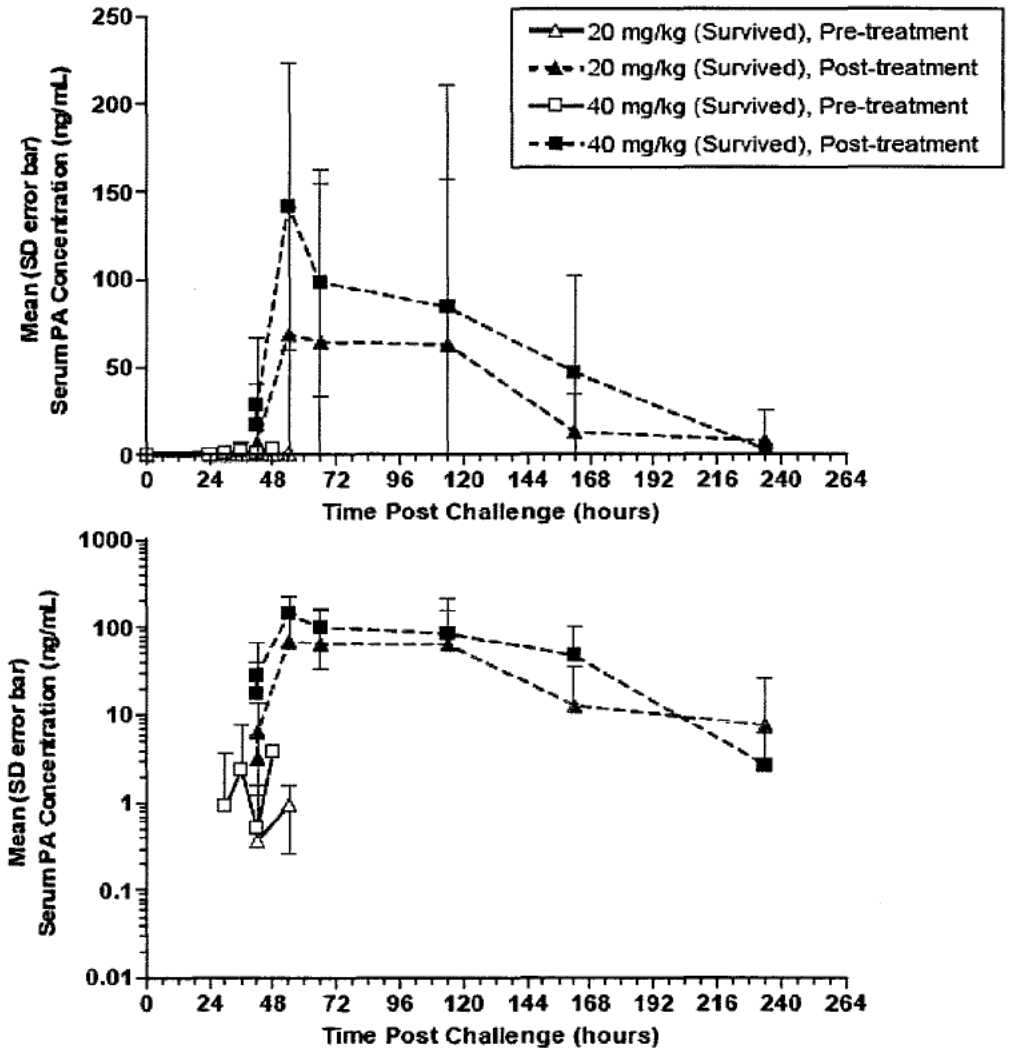
Mean (± SD) serum PA concentration-time profiles for animals that survived within each treatment group are illustrated in Figure 5. Mean serum PA concentration-time profiles for the animals that survived in the 20 and 40 mg/kg raxibacumab groups showed the initial rise, but before the plateau phase was attained, serum PA concentrations began to decline. The onset of declining serum PA concentrations was generally associated with attainment of negative bacteremia results. For all surviving animals in the 20 mg/kg dose group, serum PA concentrations declined to below the LLOQ by the end of the study. For the 40 mg/kg dose group, all but 1 of the 9 surviving animals attained serum PA concentrations below the LLOQ by the end of the study.

Figure 4. Mean (± SD) Serum PA Concentration-Time Profiles in Cynomolgus Monkeys That Died Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab and *Bacillus anthracis* Spore Challenge (Linear and Log Scale)



Source: Study Report AB50409-INF-0-040, Section 5.2

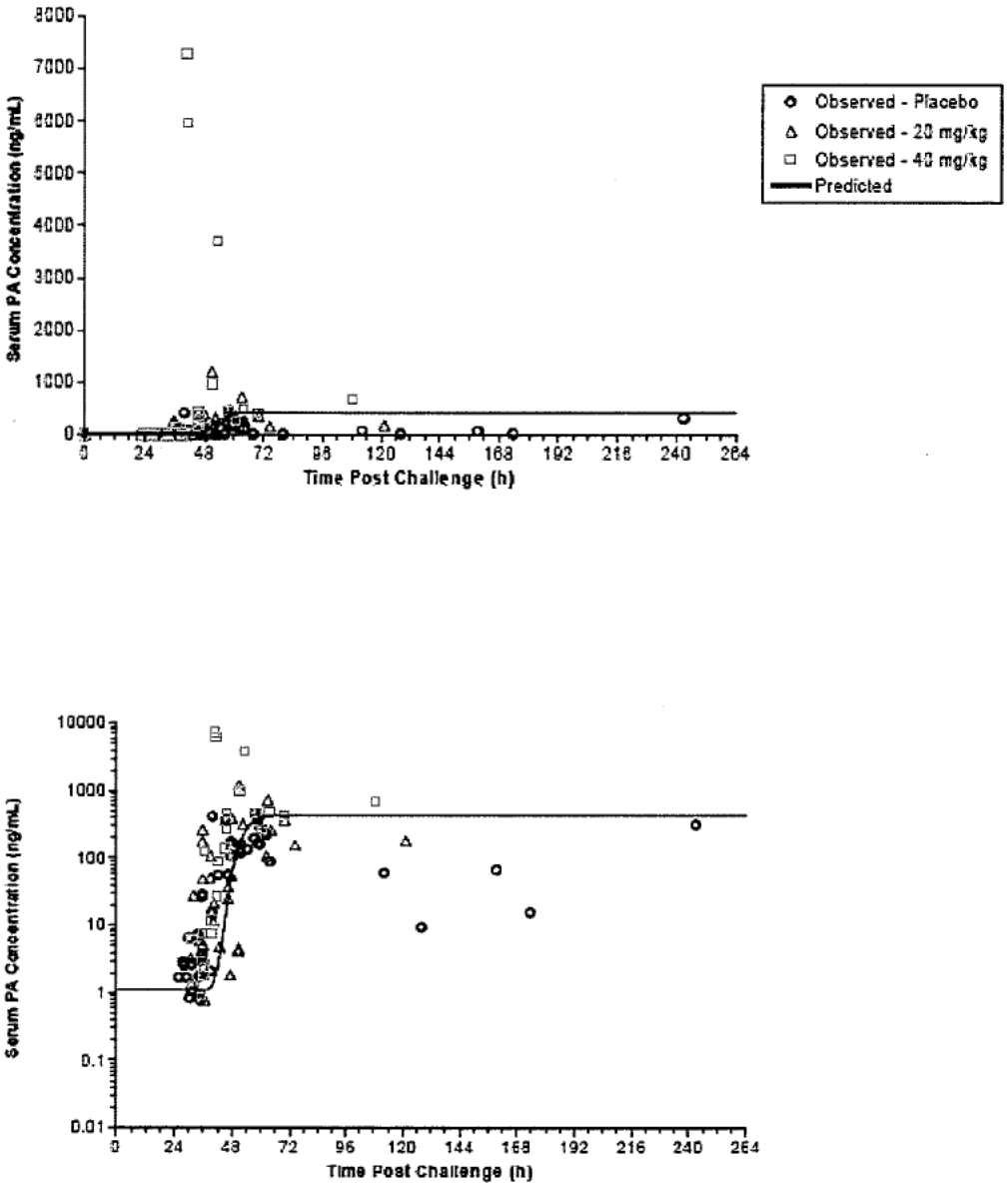
Figure 5. Mean (± SD) Serum PA Concentration-Time Profiles in Cynomolgus Monkeys That Survived Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab and *Bacillus anthracis* Spore Challenge (Linear and Log Scale)



Source: Study Report AB50409-INF-0-040, Section 5.2

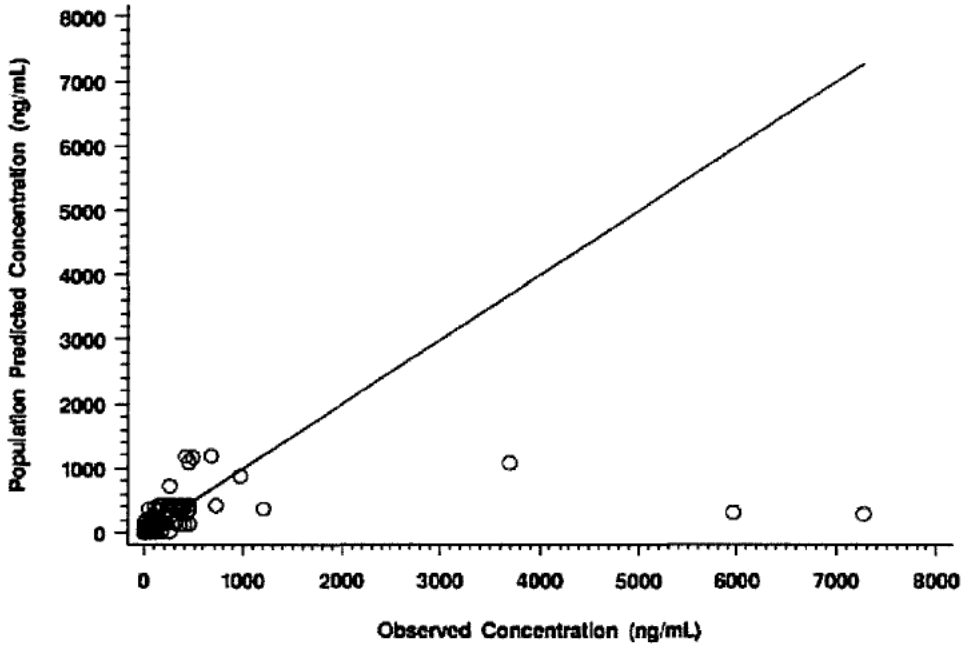
Individual observed serum PA concentrations versus the predicted population average serum PA concentration-time profile are displayed in Figure 6. Population predicted PA concentrations versus observed serum PA concentrations are presented in Figure 7. Parameter estimates are summarized in Table 3. For the PA kinetics modeling, the data for all animals that died were only consistent with a standard Gompertz model, rather than a diauxic Gompertz model; that is, there was no general evidence of the 2nd rising or plateau phases of the diauxic Gompertz model. The surviving animals in the 20 and 40 mg/kg dose groups had declining serum PA levels at times post dosing, and hence were inconsistent with and could not be fit to the Gompertz model. Therefore, the data for all animals that died were fit to the standard Gompertz model (ie, N_0, μ_m, A, and λ were estimated).

Figure 6. Predicted Population Average Serum PA Concentration-Time Profiles and Individual Observed Serum PA Concentrations Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Cynomolgus Monkeys (Linear and Log Scale)



Source: Study Report AB50409-INF-0-040, Section 5.2

Figure 7. Population Predicted Serum PA Concentrations Versus Individual Observed Serum PA Concentrations Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Cynomolgus Monkeys



Source: Study Report AB50409-INF-0-040, Appendix 15

Table 3. Summary of Model-Derived PA Kinetic Parameters Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Cynomolgus Monkeys

Parameters	Mean	CV%
N_0 (ng/mL)	1.12	3.9
λ (h)	41	NA
Effect of time to 1st bacteremia by culture (t_B) on λ	$\lambda = 41 \cdot (t_B/43.7)^{0.595}$	
At 26 h	26	
At 43.7 h	41	
At 247 h	194	
μ_m (h^{-1})	0.591	187.8
A (unitless)	5.95	NA
Effect of Dose on A	A = 5.95 + 0.0511 x (Dose-20)	
At 0 mg/kg	4.93	
At 20 mg/kg	5.95	
At 40 mg/kg	6.97	
Residual variability for Placebo Group (CV%)	36.6	
Residual variability for Raxibacumab Groups (CV%)	317.8	

Abbreviations: CV%, coefficient of variation; N_0, PA concentration at time 0; A, natural log of the ratio of the PA concentration in the asymptotic phase to N_0; μ_m, maximum specific growth rate; λ, lag time; NA, not applicable.

Source: Study Report AB50409-INF-0-040, Section 5.2

There was substantial inter-individual variability in serum PA levels. Further examination of the individual serum PA concentration-time profiles also suggests that a substantial intra-individual variability component may also exist. The PA kinetic results should be interpreted with caution, due to the poor definition of the variability components and the relatively small sample size. As expected, some variability in λ and A could be explained by time to 1st bacteremia by culture and raxibacumab dose, respectively. Smaller values of λ (lag time for serum PA concentrations) were related to shorter times to 1st bacteremia by culture, and larger values of λ were related to longer times to 1st bacteremia by culture. For A (relative magnitude of serum PA levels in the initial plateau phase), higher raxibacumab doses were associated with higher values for A, and no raxibacumab dose was associated with the lowest value of A. This relationship likely reflects the ability of raxibacumab to block the lethal effects of PA. That is, the monkeys with no exposure to raxibacumab died at lower serum PA levels than were tolerated by monkeys administered raxibacumb, and for the monkeys that were administered raxibacumab but died, higher serum PA concentrations were attained prior to death in the high dose group.

It should also be noted that during qualification of the PA assay it was observed that the amount of PA measured increases in the presence of raxibacumab, which may, explain the higher levels for the raxibacumab-treated groups. As noted previously, for the surviving animals serum PA concentrations declined after raxibacumab treatment. Review of the serum PA concentration-time results for the surviving animals, in comparison with the results of bacteremia by culture tests, revealed that the decline in serum PA concentrations coincided with the sterilization of bacteremia after raxibacumab treatment.

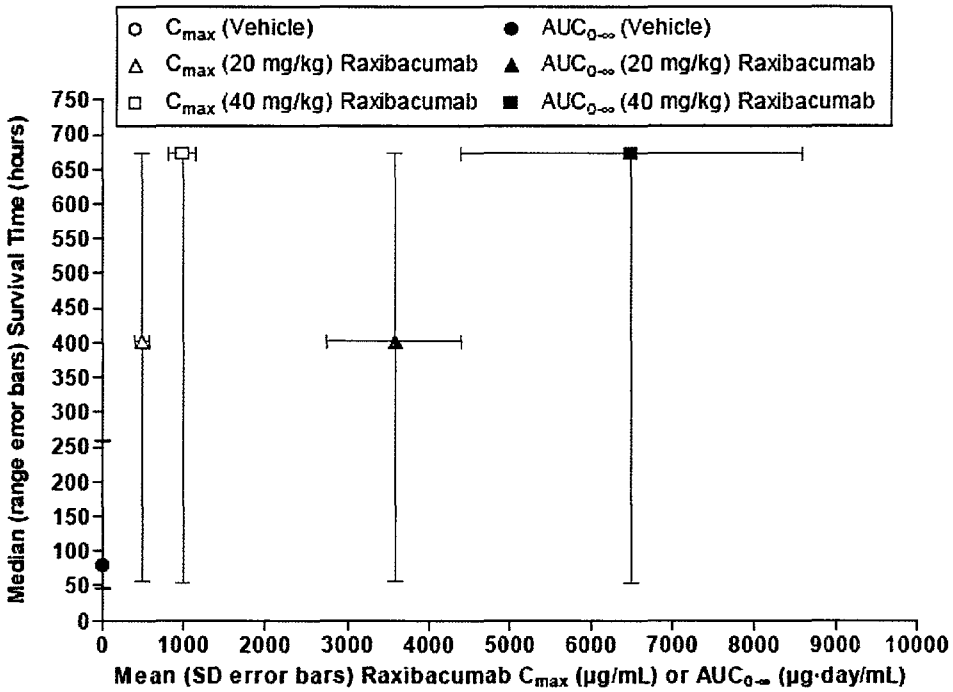
Half-life of PA was calculated for animals that had monotonically decreasing serum PA concentrations at times post raxibacumab dose, when the animal was bacteremia negative for each successive collection time. For the purposes of this analysis, if a bacteremia by culture result was not available for a collection time of interest, the bacteremia result of interest was imputed to be negative if the result at the preceding and subsequent collection times were negative; otherwise, the result was imputed to be positive. Estimation of t1/2 was only possible for 4 surviving monkeys (1 in the 20 mg/kg dose group and 3 in the 40 mg/kg dose group). The overall mean t1/2 for serum PA was 54 hours, with a median of 31 hours and range from 23 to 131 hours.

Exposure-Survival Analysis

Based on the lowest measurable observed serum raxibacumab concentrations for any monkey in either of the 2 raxibacumab treatment groups (20 mg/kg group – Monkey C21462, 21.576 µg/mL [149 nM] at 28 days post dose; 40 mg/kg group - Monkey C24754, 12.984 µg/mL [90 nM] at 28 days post dose), the minimum percentage of serum PA bound was calculated to be 98.4% for the 20 mg/kg dose group and 97.3% for the 40 mg/kg dose group. Relative to this, the highest serum PA concentration observed in a monkey from the 20 or 40 mg/kg dose groups prior to death was 7275.7 ng/mL (115 nM; Monkey No. C24847, 40 mg/kg dose group). On a nM basis, the lowest serum raxibacumab concentrations observed for either dose group, as detailed above, were similar to or 30% greater in the 40 mg/kg and 20 mg/kg dose groups, respectively, than the highest serum PA concentration observed prior to death in any monkey. Since serum raxibacumab concentrations at 5 minutes post dosing are many-fold higher than the lowest concentrations used to calculate the PA binding of 97 to 98%, it can be concluded that the exposures attained for both raxibacumab doses in this study were sufficient for virtually complete binding of PA throughout the duration of the study.

Plots of survival time as a function of raxibacumab Cmax and AUC0-∞ are displayed in Figure 8. Increase in dose (from 20 mg/kg to 40 mg/kg) was accompanied by an increase in median survival time, although there was substantial overlap of the survival time ranges between the two raxibacumab dose groups. There was a marked increase in median survival times between either raxibacumab treatment group and the vehicle treatment group. This indicates that raxibacumab binding of PA is successful in alleviating mortality due to anthrax toxemia. Overall, these results indicate that the relationship between extent of raxibacumab exposure and survival time in the raxibacumab-treated groups exists, but is not well-defined, and that survival times were increased in raxibacumab-treated animals.

Figure 8. Survival Time Versus Exposure Following Single Intravenous Administration of Vehicle, 20 mg/kg or 40 mg/kg Raxibacumab in Spore-Challenged Cynomolgus Monkeys



For purposes of this graph, animals in the vehicle treatment group were assigned a Cmax of 0 µg/mL and an AUC0-∞ of 0 µg·day/mL. Animals that survived were assigned a survival time of 672 hours.

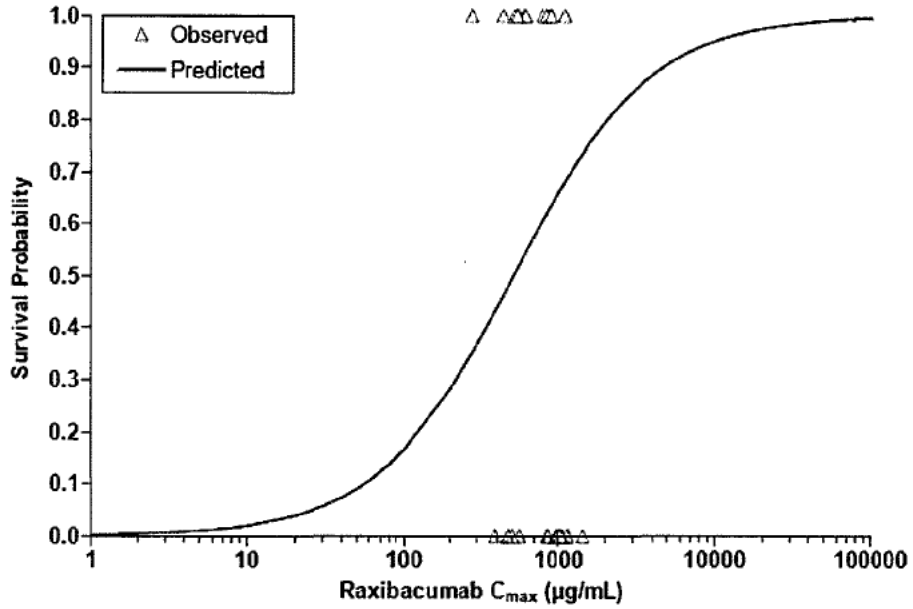
Source: Study Report AB50409-INF-0-040, Section 5.3

Results from the logistic regression model used to describe the relationship between the survival probability and Cmax or AUC0-∞ are presented in Figures 9 and 10, respectively. The results did not show a strong relationship for likelihood of survival to Cmax or AUC0-∞ ($p = 0.3125$ for Cmax and $p = 0.8122$ for AUC0-∞). This is likely due to a lack of differentiation between surviving animals and those that died in the raxibacumab-treated groups, with regard to magnitude of raxibacumab exposure. Exposure predictions for achieving 60, 70, 80, and 90% survival probability are shown in Table 4. The probability of achieving 60% survival corresponds with a Cmax of 765 µg/mL, which would be attained at a dose of 61 mg/kg for a male monkey (a female monkey administered the same dose would have higher exposure). The probability of achieving 60% survival corresponds with an AUC0-∞ of 7655 µg·day/mL, which would be attained at a dose of 103 mg/kg. These estimates of exposures and doses required to attain the tabulated survival probabilities must be interpreted with caution, as they are extrapolations outside the range of observed data.

For this study, reported survival rates were 50% and 64% for 20 and 40 mg/kg, respectively. These survival differences were not statistically significant. The logistic regression predictions for 50% and 64% survival are 509 and 908 µg/mL for Cmax, respectively, and 1374 and 15720 µg·day/mL for AUC0-∞, respectively. For 20 and 40 mg/kg, the mean Cmax were 491 and 990 µg/mL, respectively, and the mean AUC0-∞ were 3576 and 6491 µg·day/mL, respectively. The logistic regression predictions are in reasonable agreement with the observed data for Cmax,

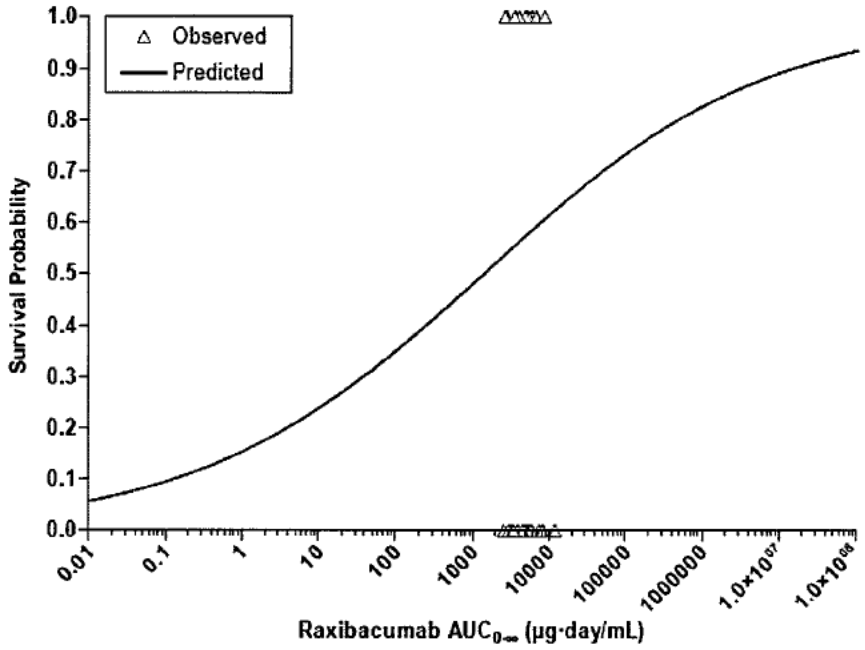
while the logistic regression predictions for AUC0-∞ are in poorer agreement with the observed data. Given the marginal increase in survival as dose was doubled from 20 to 40 mg/kg, it seems reasonable to expect that a much higher dose would be necessary to attain 70% or higher survival. The logistic regression predictions are consistent with that expectation.

Figure 9. Logistic Regression Analysis Between Survival Probability and Cmax Following Single Intravenous Administration of Vehicle, 20 mg/kg or 40 mg/kg Raxibacumab in Spore-Challenged Cynomolgus Monkeys



Source: Study Report AB50409-INF-0-040, Section 5.3

Figure 10. Logistic Regression Analysis Between Survival Probability and AUC0-∞ Following Single Intravenous Administration of Vehicle, 20 mg/kg or 40 mg/kg Raxibacumab in Spore-Challenged Cynomolgus Monkeys



Source: Study Report AB50409-INF-0-040, Section 5.3

Table 4. Predicted Raxibacumab Cmax and AUC0-∞ Corresponding to 60, 70, 80, and 90% Survival Probability Based on Logistic Regression Analysis

	Survival Probability[1]			
	60%	70%	80%	90%
C_{max} (µg/mL)	765	1194	2056	4656
Dose (mg/kg) to attain target C_{max}[2]	61	95	164	371
$AUC_{0-\infty}$ (µg·day/mL)	7655	49736	487669	15128366
Dose (mg/kg) to attain target $AUC_{0-\infty}$[2]	103	671	6577	204024

[1] Results were obtained from the logistic regression modeling.

[2] Assuming linear PK, for a 3 kg male monkey.

Source: Study Report AB50409-INF-0-040, Section 5.3

Anti-PA Antibodies

Serum anti-PA Ab concentration results for the 20 and 40 mg/kg raxibacumab dose groups are summarized in Table 5. Mean serum anti-PA concentration for the 40 mg/kg dose group was more than 2-fold higher than that for the 20 mg/kg dose group. However, the inter-individual variation was large, such that the difference in the means was not statistically significant. The results indicate that increasing exposure to raxibacumab did not decrease the formation of anti-PA Ab by the surviving monkeys in this study.

Table 5. Summary of Serum Anti-PA Antibody Concentrations in Surviving Monkeys Following Single Intravenous Administration of 20 mg/kg or 40 mg/kg Raxibacumab

		Serum Anti-PA Antibody Concentration (µg/mL)	
		Predose	28 Day Postdose
20 mg/kg	N	7	7
	Mean	0	302
	95% CI	(0,0)	(-39,643)
40 mg/kg	N	9	9
	Mean	0	775
	95% CI	(0,0)	(292,1257)

Source: Study Report AB50409-INF-0-040, Section 5.4

Toxin Neutralization Activity

Serum TNA titer results for the 20 and 40 mg/kg raxibacumab dose groups are summarized in Table 6. The mean serum TNA titer for the 40 mg/kg dose group was nearly 3-fold higher than that for the 20 mg/kg dose group. However, the inter-individual variation was large, such that the difference in the means was not statistically significant. Increasing exposure to raxibacumab did not decrease the formation of TNA by the surviving monkeys in this study.

Table 6. Summary of Serum TNA Titers in Surviving Monkeys Following Single Intravenous Administration of 20 mg/kg or 40 mg/kg Raxibacumab

		Serum Anti-PA Antibody Concentration (µg/mL)	
		Predose	28 Day Postdose
20 mg/kg	N	7	7
	Mean	0	302
	95% CI	(0,0)	(-39,643)
40 mg/kg	N	9	9
	Mean	0	775
	95% CI	(0,0)	(292,1257)

Source: Study Report AB50409-INF-0-040, Section 5.4

APPLICANT'S CONCLUSIONS:

- Serum raxibacumab concentrations in monkeys administered raxibacumab were best fit to a 2 compartment model with 1st-order elimination from the central compartment.
- V2 is dependent on body weight, increasing as body weight increases. V1 is about the same as the plasma volume; however, the data indicate that raxibacumab does distribute to tissues.
- V1 and CLD2 are dependent on sex, with females having smaller values for each parameter than males.

- In this study, raxibacumab PK appear to be independent of age, size of spore challenge, duration of spore challenge, survival time, survival status, time to 1st bacteremia by culture, and bacteremia outcome at each collection time. PK were linear over the dose range evaluated. Inter-individual variability in PK was low.
- Vss (245 to 285 mL) and CL (20 mL/day) were increased while t1/2,β (9.4 to 10.2 days) was decreased in monkeys with inhalation anthrax, relative to healthy monkeys in a prior study. These differences may reflect altered physiology in animals with inhalation anthrax, binding of raxibacumab to PA, or a combination of these or other factors.
- Serum total PA (free + bound) profiles in monkeys that died can be fit to a standard Gompertz model (rise-plateau). Monkeys that survived had profiles inconsistent with that model, in that serum PA concentrations tended to decrease at times coincident with negative bacteremia after raxibacumab dosing.
- For monkeys that died, λ (lag time for serum PA levels) is associated with time to 1st bacteremia by culture, while A (1st phase plateau serum PA concentration) is associated with raxibacumab dose. The latter likely reflects raxibacumab's protective mechanism against the lethal effects of anthrax toxins.
- Although there appears to be substantial inter-individual variability in serum PA concentrations, it was only possible to model the inter-individual variability of μ_m and N_0. Rather than revealing any deficiency in the model, this outcome may be a qualitative reflection of the data being modeled.
- In surviving monkeys, t1/2 for PA during the post raxibacumab dose bacteremia-free period averaged 54 hours with a median and range of 31 hours and 23 to 131 hours, respectively.
- The exposures attained for both raxibacumab doses in this study were sufficient for virtually complete binding of PA (based on observed exposures and in vitro binding kinetics).
- Although the increased extent of raxibacumab exposure at the higher dose was associated with increased median survival time, there was substantial overlap of survival times between the 2 raxibacumab doses. However, survival times were increased in raxibacumab-treated animals over those that were administered placebo.
- Although mean anti-PA antibodies concentrations and TNA titers for the surviving monkeys were higher for the 40 mg/kg dose than for the 20 mg/kg dose, the differences did not achieve statistical significance.

REVIEWER ASSESSMENT:
Results from Study AB50409.INF.0.040 adequately described the pharmacokinetics of raxibacumab following a single intravenous (IV) raxibacumab dose and the the kinetics of *Bacillus anthracis* protective antigen (PA) in monkeys with inhalation anthrax. The applicant's pharmacokinetic conclusions based on these findings are valid.

Based on in vitro binding kinetics, serum concentrations of approximately 40 μg/mL are required for 99% binding of PA. In the current study, serum concentrations of raxibacumab were greater than 40 μg/mL in all surviving animals in both dose groups through Day 14 of sampling. In the PK samples obtained from animals that did not survive, serum concentrations of raxibacumab were many fold greater than that required for 99.9% binding of PA. This supports the applicant's conclusion that the exposures attained for both raxibacumab doses in this study were sufficient for virtually complete binding of PA.

Results from the applicant's logistic regression analysis describing the relationship between the survival probability and exposure did not show a strong relationship for likelihood of survival to

Cmax or AUC0-∞ (p = 0.3125 for Cmax and p = 0.8122 for AUC0-∞). As suggested by the applicant, this is likely due to a lack of differentiation between surviving animals and those that died in the raxibacumab-treated groups, with regard to magnitude of raxibacumab exposure. Figures 11 and 12 below clearly illustrate the lack of differentiation in exposure between surviving animals and those that died between the two dose groups. As displayed in the graphs, concentration-time profiles and the ranges of exposure measures for the two dose groups demonstrated considerable overlap. Although definition of an exposure-response relationship for raxibacumab and survival would facilitate extrapolation of animal efficacy findings to humans, the application of the applicant's logistic regression findings in the current study is limited by the degree of overlapping exposure between surviving animals and animals that died.

Figure 11. Spaghetti Plot of Serum Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Monkeys

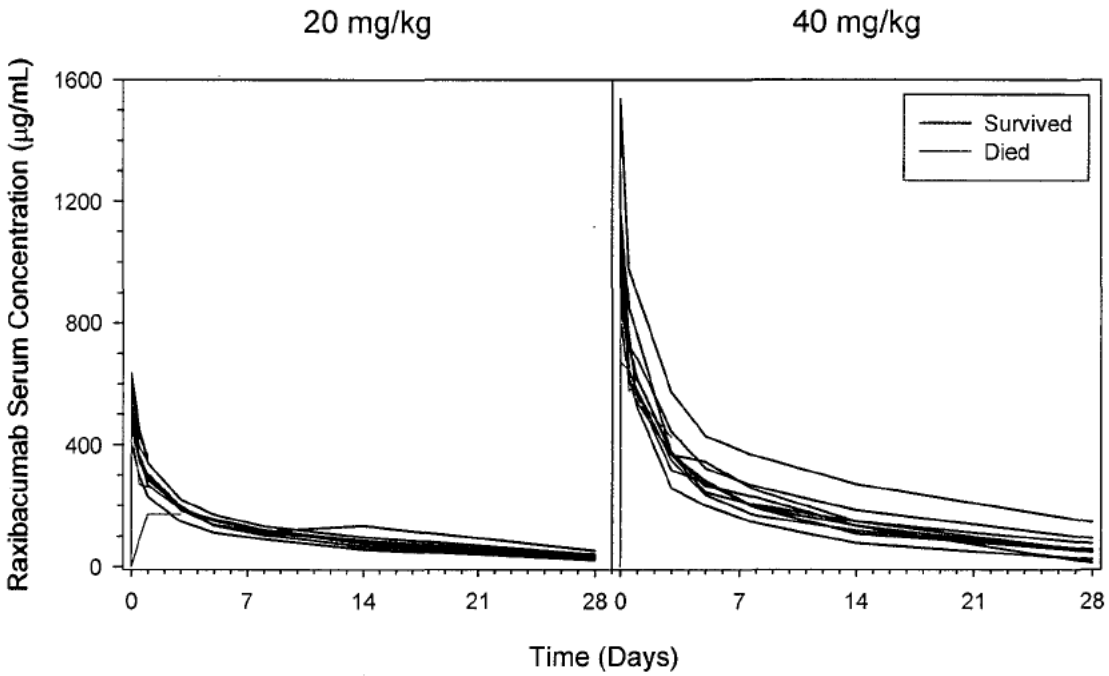
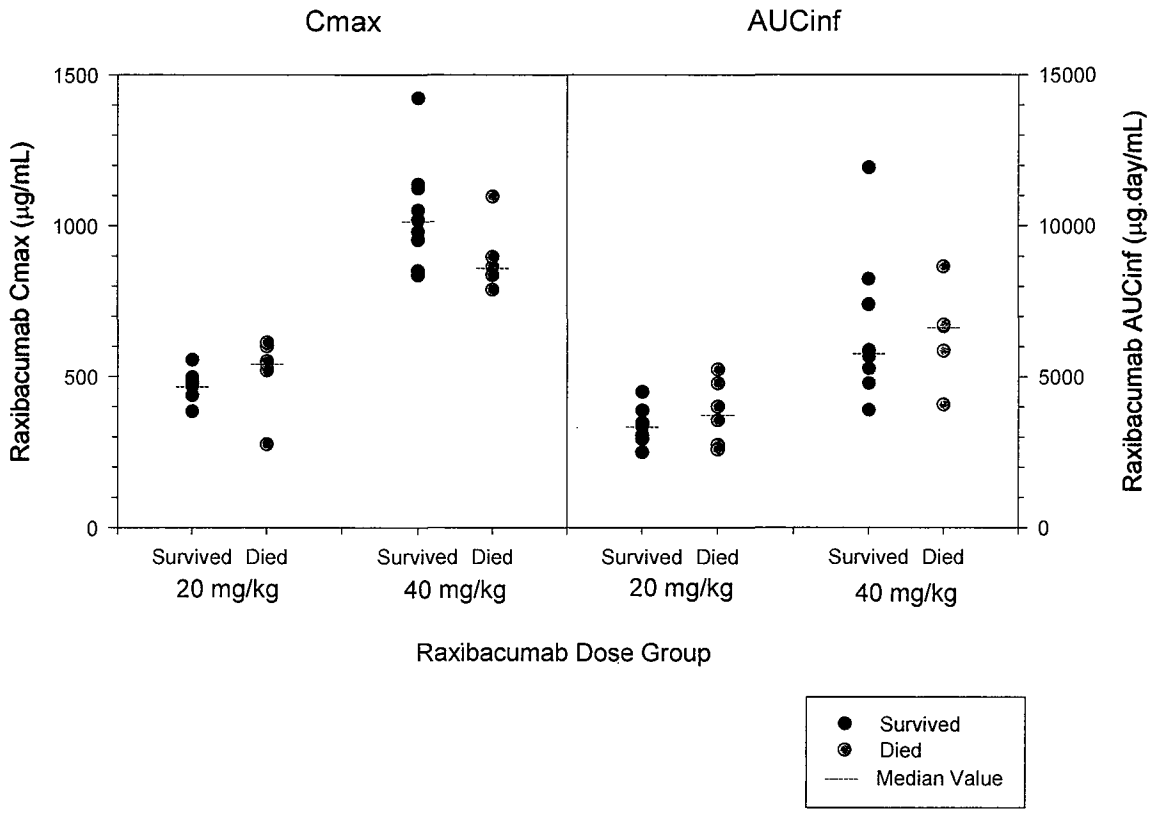


Figure 12. Comparison of Raxibacumab Cmax and AUCinf Between Animals that Survived and Animals that Died Following *Bacillus anthracis* Spore Challenge and Treatment with Single Intravenous Administration of 20 mg/kg or 40 mg/kg Raxibacumab



In addition, the applicant used the logistic regression model to predict exposures and doses that would achieve 60, 70, 80, and 90% survival probability. The applicant contends the logistic regression predictions are in reasonable agreement with the observed data for Cmax. In the current study, reported survival rates were 50% and 64% for 20 and 40 mg/kg, respectively. The logistic regression predictions for 50% and 64% survival are 509 and 908 µg/mL for Cmax, respectively. Following 20 and 40 mg/kg dose administration in monkeys, the mean Cmax were 491 and 990 µg/mL, in agreement with the model predictions. Using the model to predict Cmax values needed for a desired percent survival seems reasonable based on this comparison to observed data. But, further examination of the doses predicted to achieve specified survival rates underscore the model's limitations. The predictions suggest that the probability of achieving 60% survival corresponds with a Cmax of 765 µg/mL and a predicted dose of 61 mg/kg for a 3 kg male monkey. In the current study, the reported survival rate for the 40 mg/kg dose group was 64% and observed Cmax values in monkeys receiving this dose ranged between 789 to 1423 µg/mL. Thus, at a dose lower than the model predicted 61 mg/kg, a higher percent survival and higher Cmax was achieved in monkeys, suggesting the model over-predicts the dose necessary to achieve a desired outcome (survival).

Additionally, the predictions suggest that the probability of achieving 60% survival corresponds with an AUC0-∞ of 7655 µg·day/mL, which would be attained at a dose of 103 mg/kg. Following a 40 mg/kg single dose in monkeys, a survival rate of 64% was observed in animals with AUCinf values ranging between 3899 to 11934 µg·day/mL (mean of 6491 µg·day/mL). As

observed with Cmax, a dose significantly lower than the model predicted 103 mg/kg achieved a higher percent survival in monkeys. As emphasized by the applicant, these estimates of exposures and doses required to attain the tabulated survival probabilities must be interpreted with caution, as they are extrapolations outside the range of observed data.

The applicant conducted a cross-study comparison to examine the PK of raxibacumab in healthy monkeys compared to those with anthrax. In a previous study (HGS Report AB50409.INF.0.017, 2003), raxibacumab PK were determined following single IV bolus 1 and 10 mg/kg doses in healthy monkeys over a 42 day period post dose. In the current study, serum raxibacumab levels were measured for up to 28 days post dose. The cross-study comparison suggests that raxibacumab was cleared from the plasma more rapidly (shorter terminal half-life) in monkeys with anthrax than in healthy monkeys. This is confirmed by comparison of CL for healthy monkeys, which ranged from 4.1 to 5.0 mL/kg/day, with that for monkeys with anthrax, at 6.7 mL/kg/day. Similarly, t1/2,β in healthy monkeys ranged from 11.8 to 15.8 days, while in monkeys with anthrax, t1/2,β was shorter, at 9.4 to 10.2 days. However, t1/2,α ranged from 0.7 to 1.1 days in healthy monkeys, and was similar for the monkeys with anthrax disease in the current study (0.5 to 0.8 days), indicating that the differences in the profiles are related to elimination processes rather than initial distribution processes. V1 was slightly smaller in healthy monkeys (range: 36 to 39 mL/kg) than in monkeys with anthrax in the current study (40 to 53 mL/kg). Similarly, Vss in healthy monkeys (78 to 79 mL/kg) was slightly smaller than that in monkeys with anthrax (82 to 95 mL/kg). Based on the applicant's interpretation, the differences in CL, t1/2,β, V1, and Vss may be a reflection of altered physiology as a result of anthrax disease, binding of raxibacumab to PA, or a combination of those factors. Raxibacumab bound with PA could result in a complex that is more rapidly cleared than raxibacumab alone. Also of importance, the manufacturing processes used for raxibacumab differed between the two studies used for comparison which could also influence the disposition of raxibacumab. Thus, the applicant's conclusion that it is not possible to differentiate the contributions of these possible causes to the differences in PK between healthy monkeys and monkeys with anthrax infecton based on the results of this study is valid.

AB50409.INF.0.042

Ciprofloxacin and Raxibacumab Pharmacokinetics, with Protective Antigen Kinetics, During the Evaluation of the Efficacy of Raxibacumab in Combination with Ciprofloxacin for Therapeutic Treatment in the Cynomolgus Monkey Inhalation Anthrax Model (Battelle Study No. 789-G923702)

OBJECTIVES:

- To determine the pharmacokinetics (PK) of intragastric (IG) ciprofloxacin doses and of an intravenous (IV) raxibacumab dose when coadministered, as well as the kinetics of *Bacillus anthracis* protective antigen (PA), in monkeys with inhalation anthrax

STUDY DESIGN:

Forty juvenile (less than five years of age) cynomolgus monkeys (20 males and 20 females) that weighed 2.4 to 6.4 kg were randomized to treatment groups, challenge day, and challenge order. Animals were randomized by sex into two active treatment groups of 14 animals each and one control group of 12 animals, and into three aerosol challenge sets of animals. The three sets of monkeys were aerosol challenged on three staggered challenge days (Day 0). On Study Day 0, monkeys were anesthetized and individually aerosol challenged with a targeted 200 x lethal dose in 50% of the tested animals (LD_{50}, 1.24 x 10^7 spores) inhaled dose of *B. anthracis* (Ames strain) spores via a head-only inhalation exposure chamber. Following spore challenge, animals were frequently monitored for PA toxemia (every six hours from 24-54 hours post-spore challenge). Serum PA was measured using an electrochemiluminescence (ECL)-based screening assay. Immediately after the first detection of measurable serum PA for an individual monkey, the monkey was treated with one dose of intramuscular (IM) diphenhydramine (1 mg/kg), followed within five minutes by an IG 75 mg ciprofloxacin dose or an IG placebo (sterile water for injection) dose, according to treatment group assignment, which was in turn followed by one bolus IV injection of 40 mg/kg raxibacumab or raxibacumab vehicle (placebo, 1.0 mL/kg), according to treatment group assignment. IG 75 mg ciprofloxacin or placebo doses were administered every 12 hours (q12h) for a total of six doses (three days of treatment).

FORMULATIONS:

Raxibacumab (Lot 71044) and raxibacumab vehicle (Lot 71043) were supplied as ready-to-use sterile liquid formulations. Raxibacumab vehicle was raxibacumab formulation buffer (0.13 mg/mL citric acid, 2.8 mg/mL sodium citrate, 10 mg/mL sucrose, 18 mg/mL glycine, 0.2 mg/mL polysorbate 80, pH 6.5). Raxibacumab was provided as a solution (50 mg/mL) in the formulation buffer. During dosage periods, raxibacumab and vehicle were stored on ice. Prior to administration, raxibacumab was aseptically diluted to concentrations of 40 mg/mL in formulation buffer.

Ciprofloxacin (Cipro® IV, 10 mg/mL) was obtained from a commercial source (b) (4) (b) (4) Cipro I.V. was administered IG as supplied, without dilution. The vehicle control for Cipro I.V. was sterile water for injection purchased from a commercial supplier (b) (4)

PHARMACOKINETIC ASSESSMENTS:

Blood specimens for determination of ciprofloxacin serum concentrations were collected from all monkeys at seven days prior to spore challenge; at 1.5 hours after administration of the 1st dose; and at 12 hours after each dose (just prior to the subsequent dose). When feasible, a terminal blood sample was taken just prior to euthanasia for animals that were judged to be moribund.

Blood specimens for determination of raxibacumab serum concentrations were collected from all monkeys at seven days prior to spore challenge; just prior to ciprofloxacin, raxibacumab, or vehicle dosing; at 5 minutes after dosing; at 24, 48, 72, and 120 hours after dosing; and at 8, 14, 21, and 28 days after spore challenge. When feasible, a terminal blood sample was taken just prior to euthanasia for animals that were judged to be moribund.

Blood specimens for serum PA kinetics were collected from all monkeys at seven days prior to spore challenge as well as at 24, 30, 36, 42, 48, and 54 hours after spore challenge, unless treatment occurred prior to the collection time. Additional blood specimens were collected just prior to ciprofloxacin, raxibacumab, or vehicle dosing; at 5 minutes after dosing; at 24, 48, 72, and 120 hours after dosing; and at 8, 14, 21, and 28 days after spore challenge. When feasible, a terminal blood sample was taken just prior to euthanasia for animals that were judged to be moribund.

BIOANALYTICAL ANALYSIS:
Serum samples were analyzed for raxibacumab using an ECL-based assay. The raxibacumab in diluted serum samples binds to the biotinylated-PA on a streptavidin-coated plate and is detected by the addition of goat anti-human antibody labeled with MSD SULFOTAG™, an ECL label. The concentration of raxibacumab in serum samples is interpolated from a reference standard curve. The lower limit of quantitation (LLOQ) is 940 ng/mL of raxibacumab in 100% monkey serum.

Serum samples were analyzed for ciprofloxacin using a high performance liquid chromatography/ mass spectrometry/mass spectrometry (LC/MS/MS) assay. The calibration range for the assay was from 10 to 5000 ng/mL.

Total PA (free + raxibacumab-bound) concentrations in serum samples were determined using an ECL-based bridging assay. In brief, undiluted serum samples in duplicate wells are combined with monkey polyclonal antibody (pAb) anti-PA-biotin (capture) and monkey pAb anti-PA-SULFO-TAG (detector) and allowed to equilibrate in a streptavidin-coated assay plate. Following equilibration, plates are washed and read for ECL counts. The concentration of PA in serum samples and controls is interpolated from a reference standard curve. The LLOQ is 0.65 ng/mL of PA in 100% monkey serum.

Samples were also assayed for anti-PA antibody concentrations and toxin neutralization activity (TNA) titers. Anti-PA Ab concentrations in serum samples were determined using an enzyme-linked immunosorbent assay (ELISA). The LLOQ is < 600 ng/mL of anti-PA Ab in 100% monkey serum. TNA titers in serum samples were determined using a cell killing assay. In brief, inhibition of PA binding is correlated with increased cell viability and is used to determine the relative titer of serum samples compared to a known positive control. The LLOQ is < 52 titer in 100% monkey serum.

PHARMACOKINETIC/PHARMACODYNAMIC/STATISTICAL ANALYSIS:
PK analyses of raxibacumab concentration-time profiles for all raxibacumab-dosed monkeys were conducted using population analysis techniques (mixed effect modeling utilizing NONMEM). One (1)-, 2- and 3-compartment models with 1st order elimination from the central compartment were evaluated. Body weight, sex, age, size of spore challenge, duration of spore challenge, survival time, survival status, time to 1st bacteremia by culture, and bacteremia outcome at each collection time were evaluated as potential covariates for the PK parameters.

Kinetic analyses of total PA concentration-time profiles for all monkeys were conducted using population analysis techniques, with the NONMEM software. For individual animals that survived, the 1st order elimination rate constant and half-life of elimination (t1/2) for serum PA concentrations during the bacteremia-free post-treatment period were calculated, using actual collection time postchallenge. Body weight, sex, age, raxibacumab dose, size of spore challenge, survival status, time to 1st bacteremia by culture, and raxibacumab PK parameters were evaluated as potential covariates.

Serum ciprofloxacin concentration-time profiles for the subjects were analyzed individually. The maximum serum ciprofloxacin concentration after the 1st dose (Cmax,1) was defined as the concentration measured 1.5 hours after the 1st dose, while the minimum serum drug concentration after the nth dose (Cmin,n) was defined as the concentration measured just prior to the subsequent dose, or for the 6th dose, at 12 hours after that dose.

Serum ciprofloxacin concentration-time data, serum raxibacumab concentration-time data, serum PA concentration-time data, individual ciprofloxacin PK parameters, individual raxibacumab PK parameters, individual PA kinetic parameters, serum anti-PA Ab concentrations, and serum TNA titers were summarized using descriptive statistics (number of observations [N], mean, standard deviation [SD], standard error [SE], coefficient of variation [CV%], minimum, median, maximum, geometric mean, and 95% confidence interval [CI]). Unpaired t-tests were used to compare ciprofloxacin Cmax,1 and Cmax,n between Group 2 and Group 3. For animals that survived, an unpaired t-test was used to compare PA t1/2 between Group 2 and Group 3. Unpaired t-tests were used to compare serum anti-PA Ab or TNA concentrations at corresponding collection times between Group 2 and Group 3. All comparisons were carried out at the $\alpha = 0.05$ level of significance.

RESULTS:

A summary of sex, age and body weight for the monkeys is presented in Table 1.

Table 1. Baseline Characteristics of Monkeys in Study 789-G923702

Group	Number of Animals (N)	Age (yr)	Weight (kg)	Sex
1 (placebo/vehicle)	12	4.2 (3.6 – 4.7)	3.4 (2.6 – 4.8)	6 Females 6 Males
2 (ciprofloxacin/vehicle)	14	4.2 (2.9 – 5.0)	3.6 (2.6 – 6.5)	7 Females 7 Males
3 (ciprofloxacin/raxibacumab)	14	4.3 (3.3 – 5.1)	3.3 (2.5 – 5.3)	7 Females 7 Males

Data presented as mean (range).

Source: Study Report 789-G923702, Section 6.3

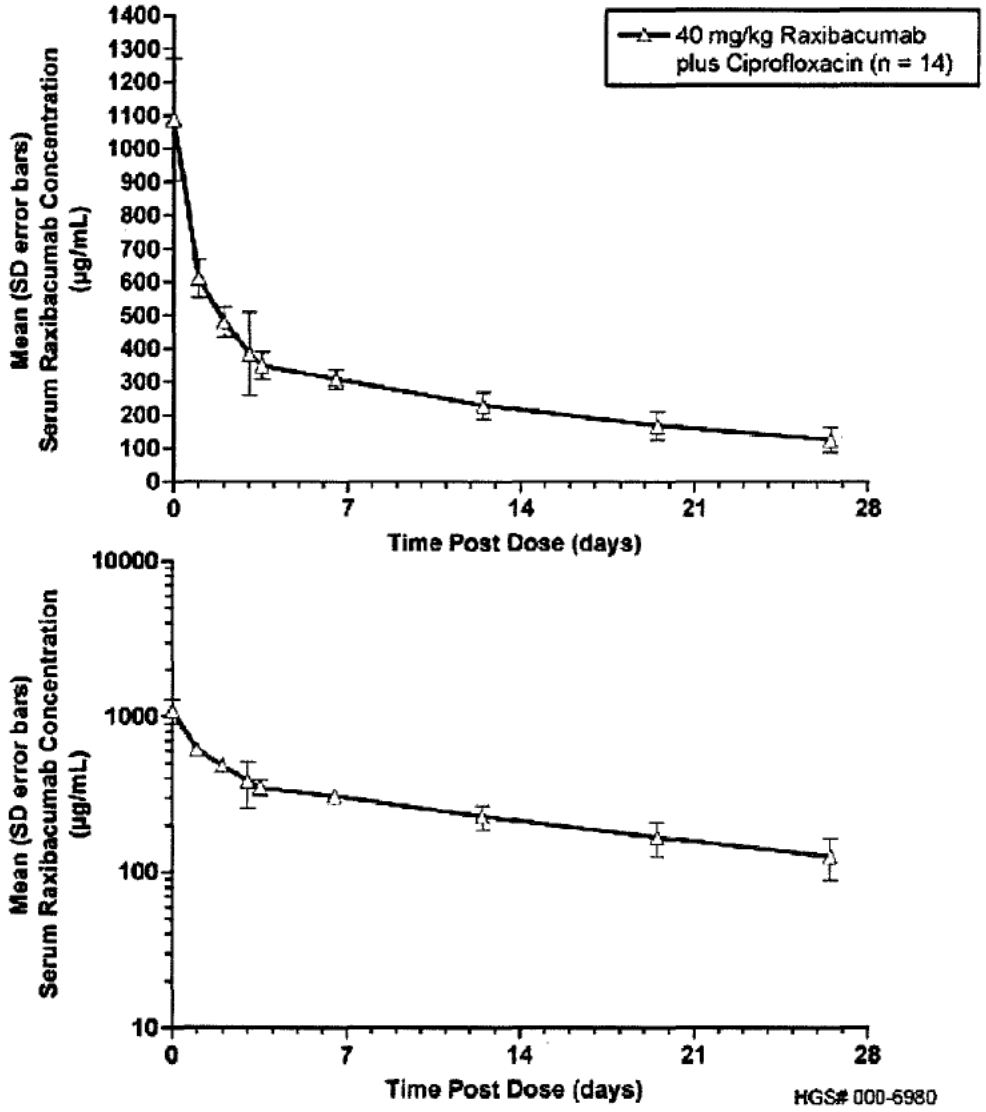
Raxibacumab Serum Pharmacokinetics

Mean (± SD) serum concentration-time profiles for raxibacumab following single intravenous administration 40 mg/kg raxibacumab in combination with ciprofloxacin q12h for 6 doses in cynomolgus monkeys are presented in Figure 1. Population predicted concentrations versus observed serum raxibacumab concentrations are presented in Figure 2. PK parameter estimates are summarized in Table 2. The serum raxibacumab concentrations best fit a 2-compartment open model with 1st-order elimination from the central compartment. Body weight was found to be a significant covariate for CL, V1, and V2. Other factors assessed (sex, age, duration of spore

challenge, size of spore challenge, bacteremia outcome at each collection time, and time to first bacteremia by culture) were not significant covariates accounting for inter-individual differences in PK. Survival time and survival status were not assessed as potential covariates due to the high survival rate for monkeys treated with ciprofloxacin plus raxibacumab.

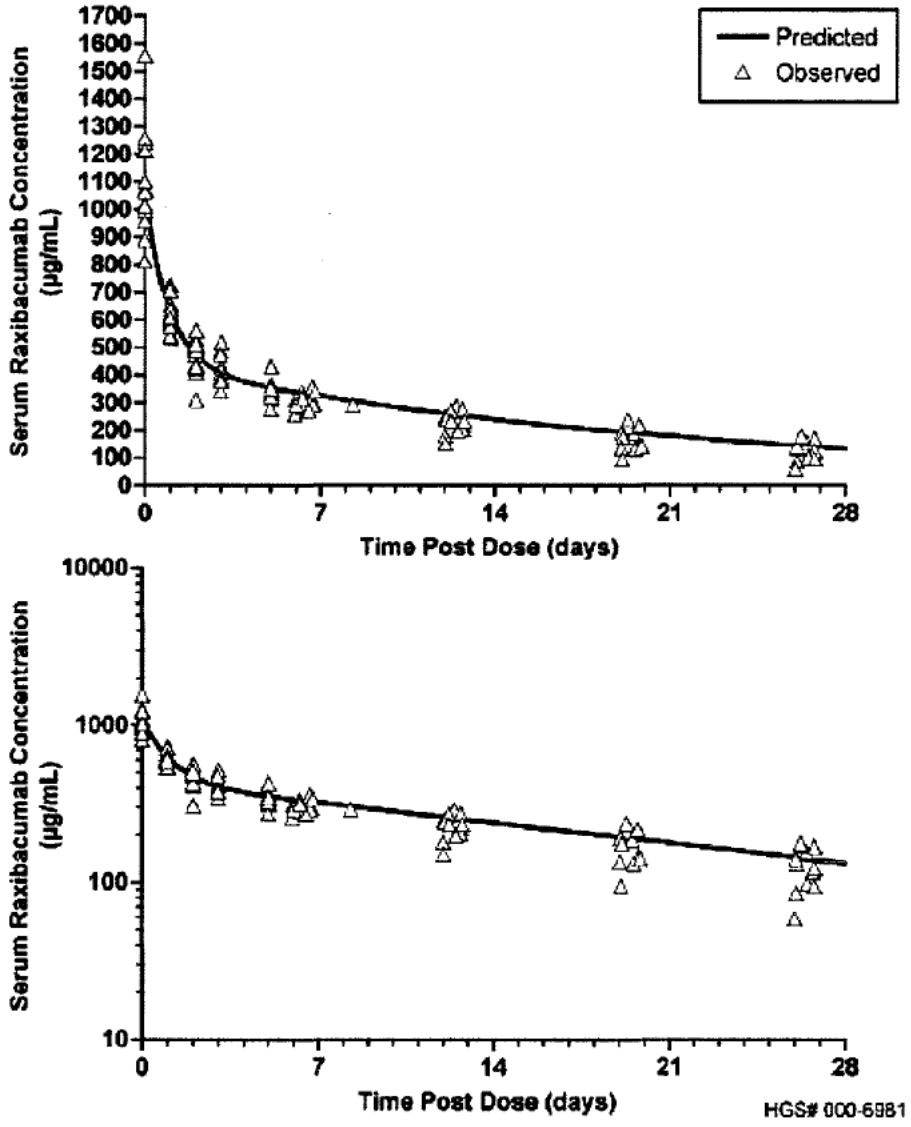
No measurable serum raxibacumab concentrations were encountered in any of the specimens collected from the monkeys in the control group. In the ciprofloxacin alone group, serum raxibacumab concentrations were encountered in some specimens (2/14 [14%] pretreatment specimens and 28/126 [22%] post-treatment specimens). These concentrations were all < 5 μg/mL and inconsistent with inadvertent administration of raxibacumab. The measurable posttreatment serum raxibacumab concentrations in the ciprofloxacin only treatment group occurred in 11 of the 14 animals, and occurred at collection times of 14 to 28 days postchallenge. This indicates that the measurable levels could represent endogenous monkey anti-PA antibodies (Ab) which are measurable in the raxibacumab assay. As noted above, the levels measured were extremely low, hence it is highly unlikely that estimation of serum raxibacumab PK was impacted by potential nonspecific measurement of monkey anti-PA Ab.

Figure 1. Mean (± SD) serum concentration-time profiles for raxibacumab following single intravenous administration 40 mg/kg raxibacumab in combination with ciprofloxacin q12h for 6 doses in cynomolgus monkeys (linear and log scale)



Source: Study Report AB50409-INF-0-042, Section 5.2

Figure 2. Predicted population average serum raxibacumab concentration-time profiles and individual observed serum raxibacumab concentrations following single intravenous administration of 40 mg/kg raxibacumab in combination with ciprofloxacin q12h for 6 doses in cynomolgus monkeys (linear and log scale)



Source: Study Report AB50409-INF-0-042, Section 5.2

Table 2. Summary of raxibacumab pharmacokinetic parameters following single intravenous administration of 40 mg/kg raxibacumab in combination with ciprofloxacin q12h for 6 doses in cynomolgus monkeys

Parameter	Units	Mean (CV%)
Primary Model-Derived Parameters		
V1	mL	117 (13.6)
CL	mL/day	11.472 (23.0)
V2	mL	139 (17.3)
CLD2	mL/day	64.8 (NA)
Secondary Parameters		
Cmax	µg/mL	1060
AUC0-∞	µg·day/mL	10782
t1/2,α	days	0.64
t1/2,β	days	16.27
MRT	days	22.26
Vss	mL	256

Volumes calculated assuming a typical monkey weighing 3.1 kg and a dose of 40 mg/kg.
Abbreviations: CV%, coefficient of variation; V1, volume of distribution for the central compartment; CL, clearance; V2, volume of distribution for the peripheral compartment; CLD2, intercompartmental clearance; NA, not applicable; Cmax, maximum serum drug concentration; AUC0-∞, area under the serum drug concentration-time curve from time 0 to infinite time; t1/2,α, elimination half-life for the 1st phase; t1/2,β, elimination half-life for the 2nd (terminal) phase; MRT, mean residence time; Vss, volume of distribution at steady-state.

Source: Study Report AB50409-INF-0-042, Section 5.2

Following IV raxibacumab administration, V1, at 117 mL (37.74 mL/kg for a 3.1 kg animal), is similar to the plasma volume (44.8 mL/kg). The steady-state volume of distribution (Vss) is nearly 2-fold greater than V1, at 256 mL (82.58 mL/kg for a 3.1 kg animal). These results suggest that although distribution of raxibacumab may initially be restricted to the plasma volume, raxibacumab does subsequently distribute to tissues. Both V1 and V2 increase with increasing weight.

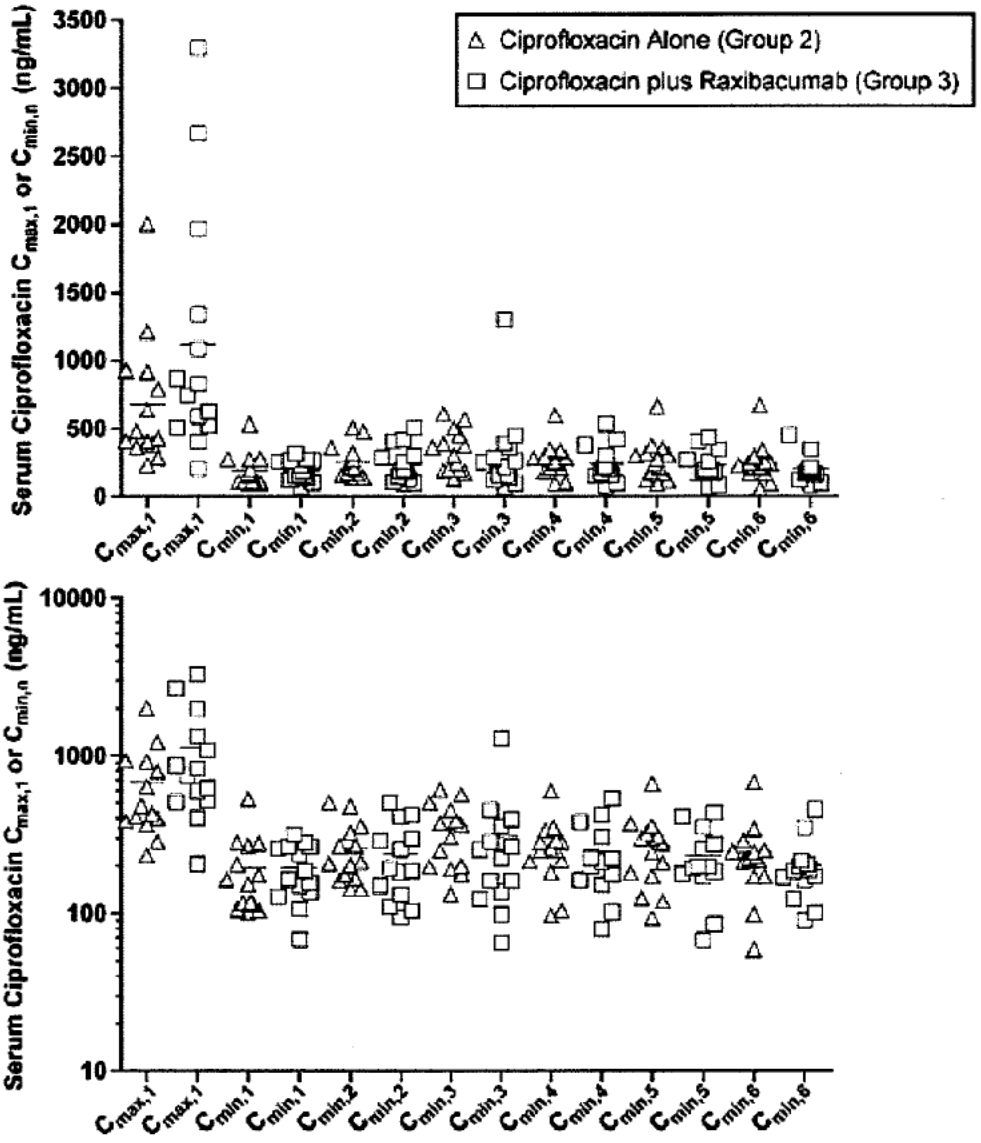
Ciprofloxacin Serum Pharmacokinetics

Mean and individual observed serum concentrations for ciprofloxacin in the ciprofloxacin alone and ciprofloxacin plus raxibacumab-treated groups (Groups 2 and 3, respectively) following multiple IG administration of 75 mg q12h for 6 doses in cynomolgus monkeys are presented in Figure 3. The Cmax,1 and Cmin,n results for Group 2 and Group 3 are summarized in Table 3. There was some accumulation of serum ciprofloxacin concentrations in both dose groups, with Cmin,n for the 2nd through 6th doses ranging from 193 to 335 ng/mL. While comparisons of Cmin,n across days within a dose group show some differences, there is no consistent pattern of increasing or decreasing values across days, suggesting that steady-state was attained prior to the 3rd ciprofloxacin dose in Group 2 and Group 3. In addition, unpaired t-tests of Cmax,n and Cmin,n between Group 2 and Group 3 show no differences that could be attributed to altered ciprofloxacin PK after raxibacumab administration.

Measurable serum ciprofloxacin concentrations were not encountered in the specimens collected from the monkeys in the control group, with the exception of Monkey C25576. Based on the observed concentrations, it appears that Monkey C25576 was administered ciprofloxacin prior to the 24 hour collection time, but since the concentrations for that animal declined throughout the

remainder of the study, it appears that ciprofloxacin was not administered at subsequent times. It should be noted that this animal did not survive; hence, the inadvertent administration of an apparent single ciprofloxacin dose had no impact on the survival outcome.

Figure 3. Mean and individual observed serum concentrations for ciprofloxacin in the ciprofloxacin alone and ciprofloxacin plus raxibacumab-treated groups (Groups 2 and 3, respectively) following multiple IG administration of ciprofloxacin 75 mg q12h for 6 doses in cynomolgus monkeys (linear and log scale)



Line represents mean at the specified time point.
Source: Study Report AB50409-INF-0-042, Section 5.1

Table 3. Summary of ciprofloxacin Cmax and Cmin concentrations following multiple IG administration of 75 mg q12h for 6 doses in cynomolgus monkeys

	Group 2 Ciprofloxacin Alone		Group 3 Ciprofloxacin plus Raxibacumab		
	N	Mean ± SD	N	Mean ± SD	P Value[1]
$C_{max,1}$ (ng/mL)	14	675.3 ±476.5	14	1119.3 ±912.2	0.1225
$C_{min,1}$ (ng/mL)	14	193.1± 117.1	14	194.5± 75.8	0.9699
$C_{min,2}$ (ng/mL)	14	257.1± 118.0	14	238.0± 130.9	0.6879
$C_{min,3}$ (ng/mL)	14	335.4± 153.4	13	229.6± 119.9	0.0580
$C_{min,4}$ (ng/mL)	14	264.9± 120.6	13	244.2± 132.2	0.6743
$C_{min,5}$ (ng/mL)	14	266.2± 144.6	13	230.7± 111.9	0.4846
$C_{min,6}$ (ng/mL)	14	243.0± 141.6	13	201.1± 99.5	0.3858

Abbreviations: $C_{max,n}$, maximum plasma ciprofloxacin concentration after the n^{th} dose, defined as the concentration measured 1.5 hour after the dose; $C_{min,n}$, minimum plasma ciprofloxacin concentration after the n^{th} dose, defined as the concentration measured just prior to the subsequent dose; NA, not applicable.

[1] From an unpaired t-test.

Source: Study Report AB50409-INF-0-042, Section 5.1

Protective Antigen (PA) Kinetics

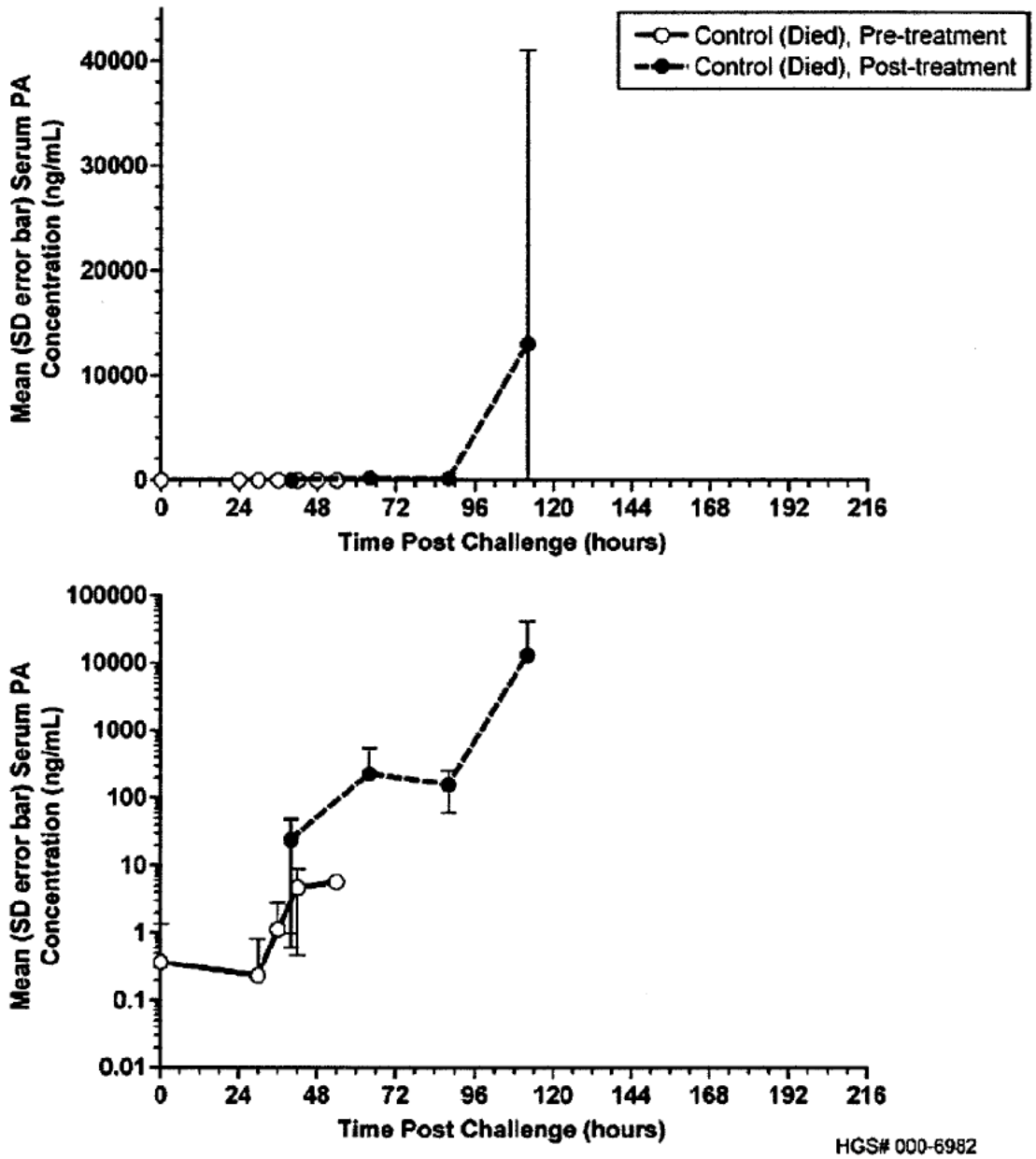
Mean (± SD) serum PA concentration-time profiles for control animals that died are illustrated in Figure 4. All animals in the control group died, whereas no animals died in the ciprofloxacin alone group. Two (2) animals died in the ciprofloxacin plus raxibacumab group and were excluded from analysis, as follows:

- Monkey C31142 in the ciprofloxacin plus raxibacumab group died, but that animal's serum PA concentration-time profile showed a decreasing pattern consistent with the effect of the drugs. Since that pattern is inconsistent with the Gompertz model, this animal was excluded from the modeling analysis.
- Monkey C24791 in the ciprofloxacin plus raxibacumab group died, but necropsy results for that animal indicated gavage error as the cause of death. Therefore, that animal was also excluded from the analysis.

In the control group, Monkey C24879 had a serum PA concentration that increased substantially after the 72 hour time point, which was excluded from the analysis. Monkey C20240 in the control group had a terminal sample that was almost 3 fold greater than mean of the terminal sample values for that group. This monkey was found dead in the cage and the time of death was unknown. Since substantial post-mortum increases in PA concentrations are possible, the reported terminal value may not represent the actual PA concentration at the time of death. Thus, that terminal value was excluded from the analysis. As noted previously, Monkey C25576 in the control group received at least 1 dose of ciprofloxacin, and was thus excluded from the analysis.

In animals that died, serum PA concentration-time profiles generally followed the pattern of rise-plateau-rise, with the exception that some animals died prior to attaining either the plateau phase or the 2nd rising phase of the serum PA concentration-time profile.

Figure 4. Mean (± SD) serum PA concentration-time profiles in cynomolgus monkeys that died following single intravenous administration of placebo (linear and log scale)

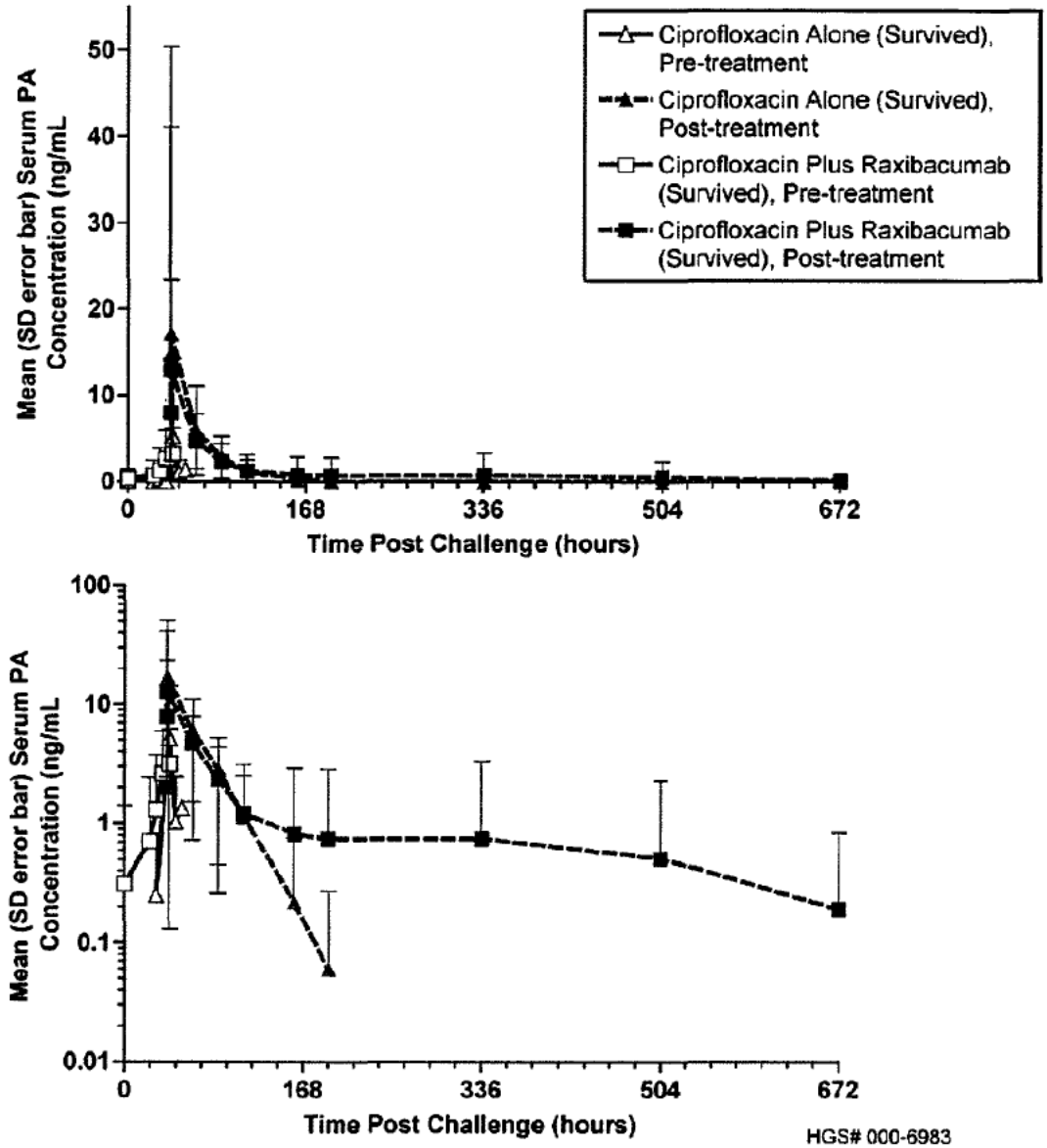


For plotting purposes, post-treatment collection times are expressed as times post challenge based on a typical treatment time of approximately 40 hours post challenge. Solid lines represent serum PA concentrations prior to treatment intervention, while dotted lines represent serum PA concentrations after treatment intervention.

Source: Study Report AB50409-INF-0-042, Section 5.3

Mean (± SD) serum PA concentration-time profiles for animals that survived within each treatment group are illustrated in Figure 5. Mean serum PA concentration-time profiles showed the initial rise, but before or shortly after the plateau phase was attained, serum PA concentrations began to decline. This increasing then decreasing type of PA profile is generally characteristic of surviving animals, and is generally not observed in those animals that died (see Study No. 685-G005762 and AB50409.INF.0.040, 2008). In previous studies, the decline of serum PA levels was observed to generally coincide with the sterilization of bacteremia. Similarly, for the surviving animals in this study, the onset of declining serum PA concentrations was generally associated with attainment of negative bacteremia results.

Figure 5. Mean (± SD) serum PA concentration-time profiles in cynomolgus monkeys that survived in the ciprofloxacin alone and ciprofloxacin plus raxibacumab-treated groups (linear and log scale)



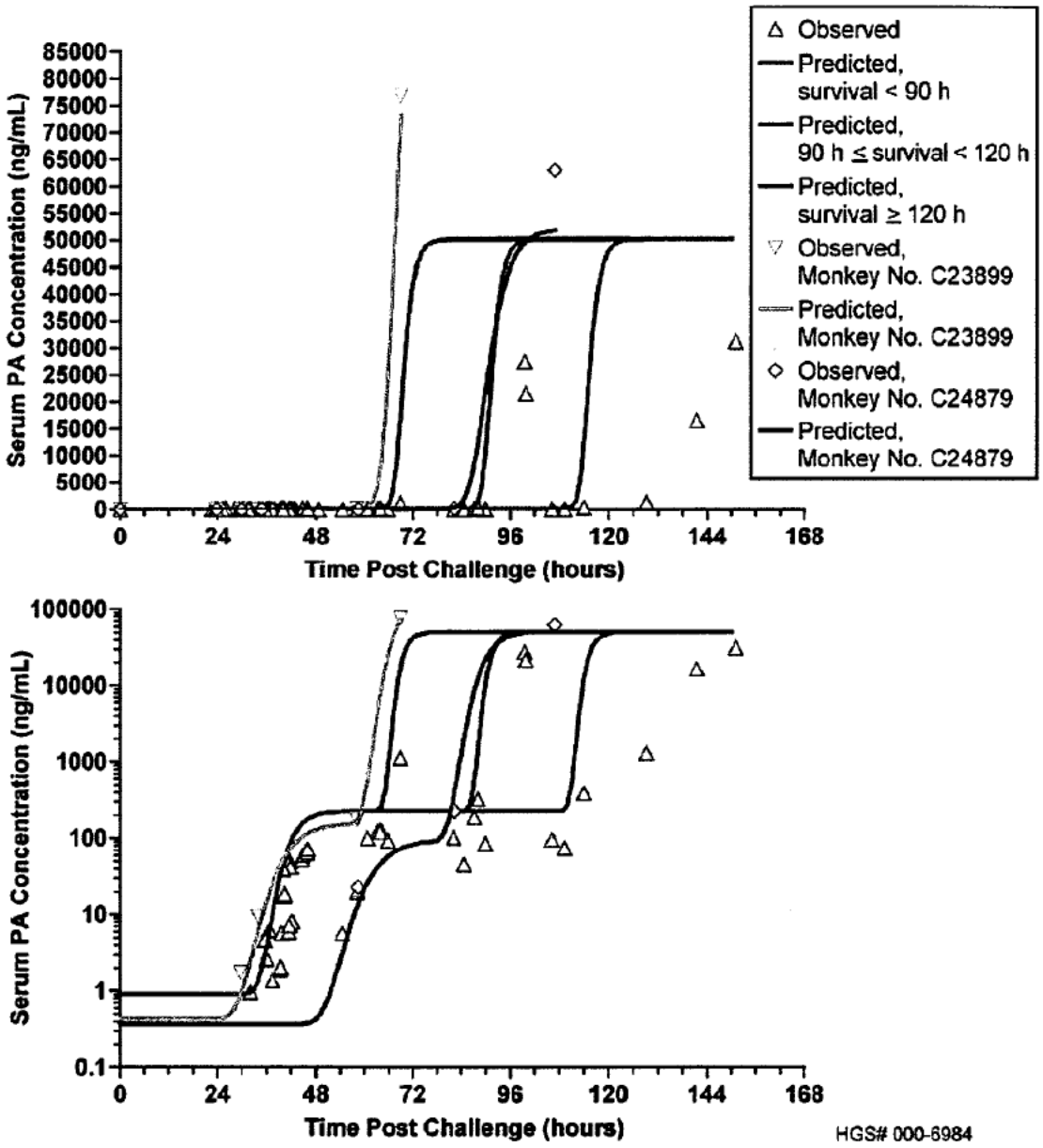
For plotting purposes, post-treatment collection times are expressed as times post challenge based on a typical treatment time of approximately 40 hours post challenge. Solid lines represent serum PA concentrations prior to treatment intervention, while dotted lines represent serum PA concentrations after treatment intervention.

Source: Study Report AB50409-INF-0-042, Section 5.3

Individual observed serum PA concentrations versus the predicted population average serum PA concentration-time profiles are displayed in Figure 6. Parameter estimates are summarized in Table 4. Despite several attempts, it was not possible to fit the data for all animals that died to the diauxic Gompertz model, likely due to the sparseness of the data and its large variability. A modified Gompertz model was employed, in which the weighting factors wα and

$w\beta$ were omitted to reduce the number of parameters to be estimated, and estimates of λ were stratified based on survival time. Monkeys C23899 and C24781 were modeled separately. Modeling of inter-individual variability for all of the model parameters was attempted, but was not successful, or did not result in an improved fit to the data. In the final model, interindividual variability could not be estimated for N0, TA, or $T\lambda$. No potential covariate effects could be evaluated, other than for sex. Sex was not found to be a significant covariate for PA kinetics.

Figure 6. Predicted population average serum PA concentration-time profiles and individual observed serum PA concentrations in cynomolgus monkeys that died which received ciprofloxacin placebo and raxibacumab vehicle (linear and log scale)



Source: Study Report AB50409-INF-0-042, Section 5.3

Table 3. Summary of model-derived PA kinetic parameters for cynomolgus monkeys that died which received ciprofloxacin placebo and raxibacumab vehicle

Parameters	Mean	CV%
N_0 (ng/mL)	0.922	NA
λ (h)	33.9	8.4
For Monkey No. C20240	49.7	NA
For Monkey No. C23899	28.5	NA
For Monkey No. C24879	49.4	NA
μ_m (h^{-1})	0.605	15.6
For Monkey No. C23899	0.430	NA
For Monkeys No. C20240 and C24789	0.373	NA
A (unitless)	4.93	4.25
T_λ (unitless)		
For survival times < 90 hours	30.8	NA
For survival times ≥ 90 hours and < 120 hours	52.6	NA
For survival times ≥ 120 hours	76.5	NA
$T_{\mu m}$ (unitless)	1.77	13.9
T_A (unitless)	0.975	NA
For Monkey No. C23899	1.13	NA
For Monkey No. C24879	1.13	NA
Residual variability (CV%), 1st phase	3.8	
Residual variability (CV%), 2nd phase	41.0	

Abbreviations: CV%, coefficient of variation; N_0, PA concentration at time 0; A, natural log of the ratio of the PA concentration in the asymptotic phase to N_0; μ_m, maximum specific growth rate; λ, lag time; T_A, the factor by which the value of A differs between the α and the β phases; $T_{\mu m}$, the factor by which the value of μ_m differs between the α and the β phases; T_λ, the factor by which the value of λ differs between the α and the β phases; NA, not applicable.

Source: Study Report AB50409-INF-0-042, Section 5.3

Longer lag times for the 2nd phase, resulting in a more prolonged plateau in the 1st phase, were related to longer times survival times. In surviving animals, the decline in serum PA concentrations coincided with the sterilization of bacteremia after treatment. Examination of the serum PA concentration-time profiles for the surviving monkeys in this study showed that for at least some monkeys, monotonically decreasing concentrations were present at post-ciprofloxacin/ raxibacumab treatment bacteremia-free collection times. Those profiles generally did not show increasing rates of PA clearance; hence, it seemed reasonable to assume that the clearance of PA from serum was not related to the development of anti-PA Ab in these animals. There was substantial inter-individual variability in serum PA levels, and there were relatively few animals in the population modeled. In addition, since each monkey survived for a different duration, each monkey's data contributed differing amounts of information regarding the different portions of the diauxic profile. The PA kinetic results should be interpreted with caution, due to the poor definition of the variability components and the relatively small sample size.

Half-life of PA was calculated for animals that had monotonically decreasing serum PA concentrations at times post raxibacumab dose. Estimation of t1/2 was only possible for 11 surviving monkeys in the ciprofloxacin alone dose group and 11 in the ciprofloxacin plus raxibacumab dose group. The mean t1/2 for serum PA was 22 hours for the ciprofloxacin alone dose group, and was 71 hours for the ciprofloxacin plus raxibacumab dose group; however, the mean for the latter group is biased by 1 monkey. Relative to this, the median t1/2 for serum PA

were similar at 23 and 27 hours for the ciprofloxacin alone and ciprofloxacin plus raxibacumab dose groups, respectively.

Exposure-Survival Analysis

Survival rates were very high in the ciprofloxacin alone and ciprofloxacin plus raxibacumab dose groups, with 0 and 2 monkeys, respectively, dying in those dose groups. Although it was intended to assess survival time as a function of raxibacumab or ciprofloxacin exposure, the minimal number of animals that died when administered those 2 drugs prevented any meaningful assessment from being made.

Anti-PA Antibodies

Serum anti-PA Ab concentration results for the ciprofloxacin alone and ciprofloxacin plus raxibacumab treatment groups are summarized in Table 4. There were no statistically significant differences in mean serum anti-PA Ab concentrations between the 2 treatment groups, indicating that co-administration of raxibacumab with ciprofloxacin did not affect the formation of anti-PA Ab by the surviving monkeys, relative to ciprofloxacin alone.

Table 4. Summary of serum anti-PA antibody concentrations in surviving monkeys administered q12h x 6 IG 75 mg ciprofloxacin doses alone or in combination with a single IV 40 mg/kg raxibacumab dose

		Serum Anti-PA Antibody Concentration (µg/mL)[1]		
		Prechallenge	21 Days Postchallenge	28 Days Postchallenge
Ciprofloxacin Alone	N	14	14	14
	Mean ± SD	0.068 ± 0.253	65.447 ± 37.687	89.126 ± 48.833
Ciprofloxacin Plus Raxibacumab	N	12	12	12
	Mean ± SD	0.295 ± 0.837	72.506 ± 33.779	100.899 ± 83.862
	p-Value[2]	0.3819	0.6222	0.6600

Source: Study Report AB50409-INF-0-042, Section 5.5

Toxin Neutralization Activity

Serum TNA titer results for the ciprofloxacin alone and ciprofloxacin plus raxibacumab treatment groups are summarized in Table 5. The mean serum TNA titers for the ciprofloxacin alone dose group were significantly higher (about 2-fold) than those for the ciprofloxacin plus raxibacumab dose group. It should be noted that these differences were not reflected in altered survival between the 2 groups, suggesting that the TNA response in monkeys administered both ciprofloxacin and raxibacumab was adequate for survival.

Table 5. Summary of serum TNA titers in surviving monkeys administered q12h x 6 IG 75 mg ciprofloxacin doses alone or in combination with a single IV 40 mg/kg raxibacumab dose

		Serum TNA Titer[1]		
		Prechallenge	21 Days Postchallenge	28 Days Postchallenge
Ciprofloxacin Alone	N	14	14	14
	Mean ± SD	0 ± 0	6369 ± 5288	7266 ± 4864
Ciprofloxacin Plus Raxibacumab	N	12	12	12
	Mean ± SD	0 ± 0	3020 ± 1802	3400 ± 1869
	p-Value[2]	-	0.0405	0.0136

Source: Study Report AB50409-INF-0-042, Section 5.5

APPLICANT'S CONCLUSIONS:

- Co-administration of IV raxibacumab had no effect on ciprofloxacin exposure for intragastic ciprofloxacin doses.
- Exposure to ciprofloxacin appears to have no impact on raxibacumab PK.
- Serum raxibacumab concentrations in monkeys were best fit to a two compartment model with 1st-order elimination from the central compartment. Inter-individual variability in PK was low. CL, V1, and V2 are dependent on body weight, increasing as body weight increases. V1 is about the same as the plasma volume; however, the data indicate that raxibacumab does distribute to tissues.
- In this study, raxibacumab PK appear to be independent of sex, age, size of spore challenge, duration of spore challenge, time to 1st bacteremia by culture, and bacteremia outcome at each collection time. Survival time and survival status could not be assessed for effects due to the high survival rate.
- Serum total PA (free + bound) profiles in monkeys that died can be fit to a diauxic Gompertz model (rise-plateau-rise). Monkeys that survived had profiles inconsistent with that model, in that serum PA concentrations tended to decrease at times coincident with negative bacteremia after raxibacumab dosing. For monkeys that died, lag time for the 2nd phase (longer plateau phase) is associated with longer survival time. There appears to be substantial inter-individual variability in serum PA concentrations.
- There were no significant differences in mean anti-PA antibody concentrations between the ciprofloxacin alone and ciprofloxacin plus raxibacumab treatment groups, whereas serum TNA titers were about 2-fold higher for ciprofloxacin alone, relative to ciprofloxacin plus raxibacumab. However, the high survival rates in these dose groups indicate that the difference in serum TNA titers is not meaningful.

REVIEWER ASSESSMENT:

Results from Study AB50409.INF.0.042 adequately determined the PK of IG ciprofloxacin doses and of an IV raxibacumab dose when coadministered, as well as the kinetics of *Bacillus anthracis* protective antigen (PA), in monkeys with inhalation anthrax. Overall, the applicant's pharmacokinetic conclusions based on these findings are valid.

Based on a cross-study comparison, PK results from Study 789-G923702 showed slight differences when compared to findings from the previous pivotal monkey efficacy study (Study 724-G005829). Mean (SD) raxibacumab serum concentration-time profiles and PK parameters following single IV administration of raxibacumab 40 mg/kg with and without

ciprofloxacin in monkeys are presented in Figure 7 and Table 6, respectively. Monkeys that received raxibacumab in combination with ciprofloxacin in Study 789-G923702 exhibited a slower clearance and higher AUC compared to monkeys in the pivotal animal efficacy study 724-G005829. Mean Cmax was comparable across studies. Since the current study did not include a raxibacumab alone control group, interpretation of these differences is limited and does not affect conclusions based on current study findings.

Figure 7. Mean (SD) raxibacumab serum concentration-time profiles following single IV administration of raxibacumab 40 mg/kg with (Study 789-G923702) and without (Study 724-G005829) ciprofloxacin in monkeys

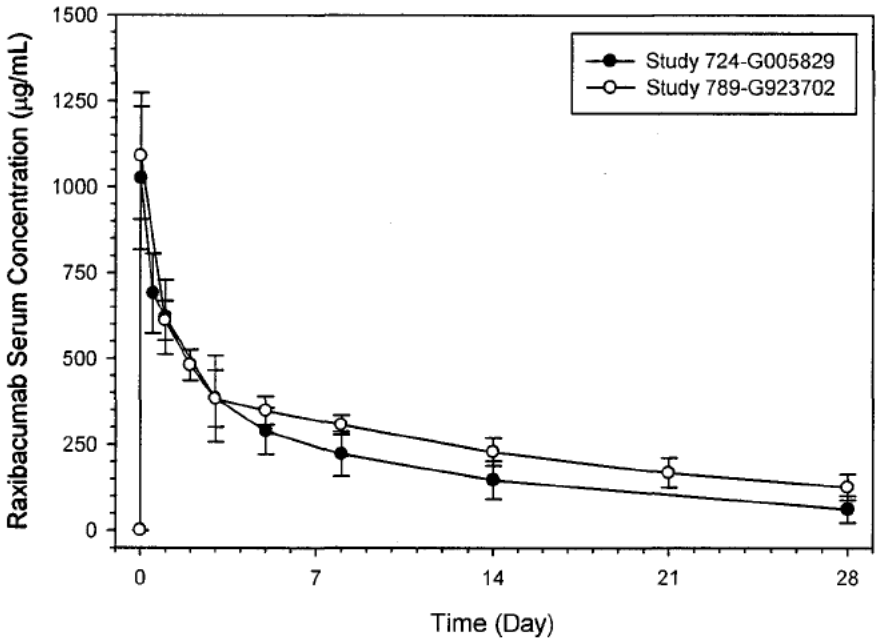


Table 6. PK parameters following single IV administration of raxibacumab 40 mg/kg with (Study 789-G923702) and without (Study 724-G005829) ciprofloxacin in monkeys

Parameter	Study	
	724-G005829	789-G923702
Cmax (μg/mL)	990	1060
AUC0-∞ (μg·day/mL)	6491	10782
T1/2β (day)	10.14	16.27
Vss (mL)	288	256
CL (mL/day)	20	11

In the current study, monkeys received intragastric ciprofloxacin at a dose of 75 mg or intragastric placebo (sterile water for injection), according to treatment group assignment. This was followed by intragastric ciprofloxacin or placebo doses administered every 12 hours (q12h) for a total of six doses (three days of treatment). The recommended oral ciprofloxacin dose in humans for post-exposure of inhalational anthrax is 500 mg q12h for 60 days. An oral dose of 500 mg ciprofloxacin in humans yields a Cmax of 2.4 μg/mL. In the current study, the mean ciprofloxacin Cmax values following the first intragastric dose of 75 mg in monkeys with and with out raxibacumab co-administration were 1.12 and 0.675 μg/mL, respectively. Thus, the ciprofloxacin dose of 75 mg in monkeys was not equivalent to the human oral dose of 500 mg approved for post-exposure inhalational anthrax. Ciprofloxacin administered at the labeled oral dose of 500 mg in humans is expected to achieve higher exposures than those observed in this study conducted in monkeys with inhalational anthrax.

AB50409.INF.0.036

Raxibacumab Pharmacokinetics and Protective Antigen Kinetics During the Evaluation of Raxibacumab Efficacy as Therapeutic Treatment Against Inhalation Anthrax in the Rabbit Model (Battelle Study No. 682-G005758)

OBJECTIVES:

- To determine the pharmacokinetics (PK) of raxibacumab following a single intravenous (IV) raxibacumab dose in rabbits with inhalation anthrax
- To determine the kinetics of *Bacillus anthracis* protective antigen (PA) in rabbits with inhalation anthrax

STUDY DESIGN:

Fifty-four (54) NZW rabbits (29 males and 25 females) weighing 2.90 to 3.97 kg prior to shipment were randomized to treatment groups, challenge day, and challenge order. On Study Day 0, rabbits were individually aerosol challenged with a targeted 200 LD50 (2.1×10^7 spores) inhaled dose of *B. anthracis* (Ames strain) spores. Following spore challenge, animals were frequently monitored for temperature increase and PA toxemia. Rabbits were treated with raxibacumab or vehicle (placebo) immediately after the 1st detection of measurable serum PA, using a screening assay, or after 2 consecutive temperature increases of $\geq$ 2°F above baseline average temperature, which ever occurred first. After 36 hours post challenge, initiation of treatment was based on detection of temperature increase only. Sixteen animals (8 males and 8 females) received a single bolus IV dose of the vehicle, while the remaining 37 animals received raxibacumab. Seventeen of the animals (10 males and 7 females) received a single 20 mg/kg IV bolus raxibacumab dose, while another 19 animals (11 males and 8 females) received a single 40 mg/kg IV bolus raxibacumab dose.

FORMULATIONS:

Raxibacumab (Lot No. 71044) and raxibacumab vehicle (Lot No. 71043) were produced at HGS, and were stored at 2-8°C prior to use. Raxibacumab and vehicle were supplied as ready-to-use sterile liquid formulations. Raxibacumab vehicle was raxibacumab formulation buffer (0.13 mg/mL citric acid, 2.8 mg/mL sodium citrate, 10 mg/mL sucrose, 18 mg/mL glycine, 0.2 mg/mL polysorbate 80, pH 6.5). Raxibacumab was provided as a solution (50 mg/mL) in the formulation buffer. During dosage periods, raxibacumab and vehicle were stored on ice.

PHARMACOKINETIC ASSESSMENTS:

Blood specimens for determination of serum raxibacumab concentrations were collected from all rabbits at 3 days prior to spore challenge; just prior to raxibacumab or vehicle dosing; and at 5 minutes as well as at 4, 10, 24, 36, 48, 72, 144, and 216 hours after dosing. The blood specimens for the 144 and 216 hours time points were not collected from the vascular access port (VAP). When feasible, a terminal blood sample was to be taken from any animal found dead or just prior to euthanasia for animals that were judged to be moribund.

Blood specimens for serum PA kinetics were collected from all rabbits at 3 days prior to spore challenge and at 12, 16, 20, 24, 28, 32, and 36 hours post challenge, unless treatment occurred prior to the collection time. Additional blood specimens were collected just prior to raxibacumab or vehicle dosing; and at 5 minutes as well as at 4, 10, 24, 36, 48, 72, 144, and 216 hours after dosing. The blood specimens for the 144 and 216 hours time points were not collected from the VAP. When feasible, a terminal blood sample was to be taken from any animal found dead or just prior to euthanasia for animals that were judged to be moribund.

BIOANALYTICAL ANALYSIS:
Serum samples were analyzed for raxibacumab using an ECL-based assay. The raxibacumab in diluted serum samples binds to the biotinylated-PA on a streptavidin-coated plate and is detected by the addition of goat anti-human antibody labeled with MSD SULFOTAG™, an ECL label. The concentration of raxibacumab in serum samples is interpolated from a reference standard curve. The lower limit of quantitation (LLOQ) is 750 ng/mL of raxibacumab in 100% rabbit serum.

Total PA (free + raxibacumab-bound) concentrations in serum samples were determined using an ECL-based bridging assay. In brief, undiluted serum samples in duplicate wells are combined with monkey polyclonal antibody (pAb) anti-PA-biotin (capture) and monkey pAb anti-PA-SULFO-TAG (detector) and allowed to equilibrate in a streptavidin-coated assay plate. Following equilibration, plates are washed and read for ECL counts. The concentration of PA in serum samples and controls is interpolated from a reference standard curve. The LLOQ is 0.34 ng/mL of PA in 100% rabbit serum and the LLOD is 0.228 ng/mL of PA in 100% rabbit serum. The upper limit of quantitation is 410000 ng/mL of PA in 100% rabbit serum.

PHARMACOKINETIC/PHARMACODYNAMIC/STATISTICAL ANALYSIS:
PK analyses of raxibacumab concentration-time profiles for all raxibacumab-dosed monkeys were conducted using population analysis techniques (mixed effect modeling utilizing NONMEM). Inspection of the individual serum raxibacumab concentration-time profiles following IV dosing revealed a profile that was multiphasic; hence, 2- and 3-compartment models with 1st order elimination from the central compartment were evaluated. Following identification of the most appropriate PK structural model and error models for the PK parameters, body weight, sex, age, treatment group, size of spore challenge, duration of spore challenge, survival time, survival status, time to 1st bacteremia by culture, and bacteremia outcome at each collection time were evaluated as potential covariates for the PK parameters. Selection between competing models was based on goodness of fit criteria (reduction in objective function [-2 log likelihood, or -2LL] value, diagnostic plots). PK parameter estimates for individual animals were derived from the final model.

Kinetic analyses of total PA concentration-time profiles for all monkeys were conducted using population analysis techniques, with the NONMEM software. Body weight, sex, treatment group, size of spore challenge, survival status, time to 1st bacteremia by culture, and raxibacumab PK parameters were evaluated as potential covariates.

In vitro, the dissociation equilibrium constant (Kd) for raxibacumab was estimated to be 2.78 nM. Using the in vitro estimate of Kd, the percentage of serum PA bound following raxibacumab dosing was calculated from the following equation:

$$\%\text{Bound} = 100\left(K_d^{-1}C_{raxi} \Big/ 1 + K_d^{-1}C_{raxi}\right)$$

where Craxi is the serum raxibacumab concentration, expressed as nM.

Serum raxibacumab concentration-time data, serum PA concentration-time data, individual raxibacumab PK parameters, and individual PA kinetic parameters were summarized using descriptive statistics. A logistic regression model was used to describe the relationship between the survival probability and maximum serum raxibacumab concentration (Cmax) or area under the serum raxibacumab concentration-time curve to infinite time (AUC0-∞).

RESULTS:
A summary of sex, age and body weight for the rabbits is presented in Table 1.

Table 1. Baseline Characteristics of Rabbits in Study 682-G005758

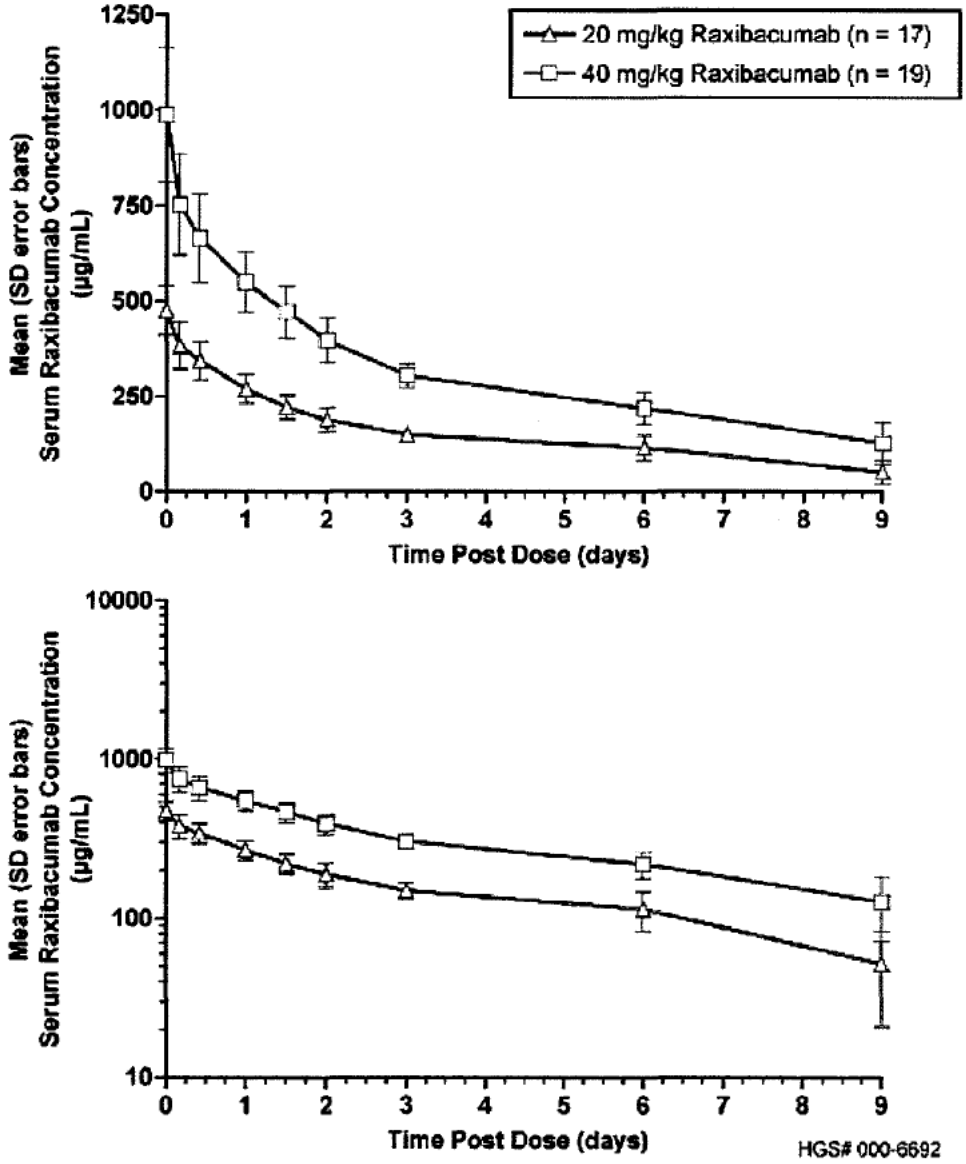
Group	Number of Animals (N)	Weight for Dosing (kg)	Sex
1 (Placebo)	16	3.0 (2.8 – 3.3)	8 Females 8 Males
2 (Raxibacumab 20 mg/kg)	18	3.1 (2.8 – 3.4)	8 Females 10 Males
3 (Raxibacumab 40 mg/kg)	19	3.1 (2.7 – 3.3)	8 Females 11 Males

Data presented as mean (range).

Source: Study Report AB50409-INF-0-036, Appendix 1

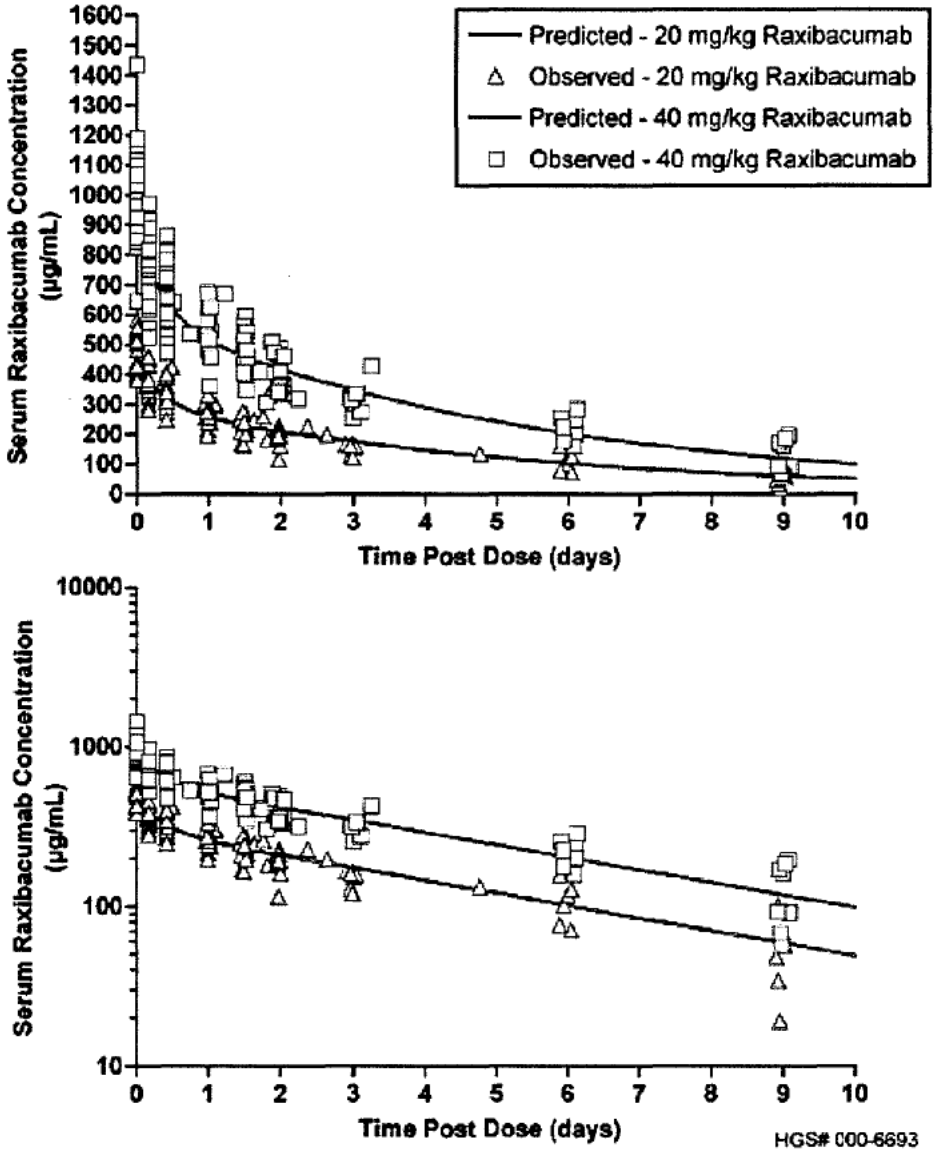
Raxibacumab Serum Pharmacokinetics
Serum concentration-time profiles for raxibacumab following single intravenous administration of 20 mg/kg and 40 mg/kg in spore-challenged rabbits are presented in Figure 1. Individual observed serum raxibacumab concentrations versus the population average serum raxibacumab concentration-time profiles are presented in Figure 2. Population predicted concentrations versus observed serum raxibacumab concentrations are presented in Figure 3. PK parameter estimates are summarized in Table 2. Serum raxibacumab concentrations were best fit a 2-compartment open model with 1st-order elimination from the central compartment. Body weight was found to be a significant covariate for V2. The lack of difference in PK between treatment groups is consistent with linear PK over the dose range studied. Following IV raxibacumab administration, V1, at 132 mL (44 mL/kg for a 3 kg animal), is the same as the plasma volume. The steady-state volume of distribution (Vss) is about 43% greater than V1, at 189 mL. These results suggest that although distribution of raxibacumab may initially be restricted to the plasma volume, raxibacumab does subsequently distribute to tissues. The disappearance of raxibacumab from serum is multiphasic, with an initial phase elimination half-life ($t1/2,\alpha$) of 0.24 days. The terminal phase elimination half-life ($t1/2,\beta$) is 3.84 days. The mean CL of raxibacumab was 35 mL/day (11.7 mL/day/kg for a 3 kg rabbit), which is much smaller than the glomerular filtration rate (4493 mL/day/kg indicating that, as expected, there is virtually no renal clearance of this monoclonal antibody. Inter-individual variability in raxibacumab PK was low, with CV% of 27% or less for the primary PK parameters.

Figure 1. Mean (± SD) Serum Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Rabbits (Linear and Log Scale)



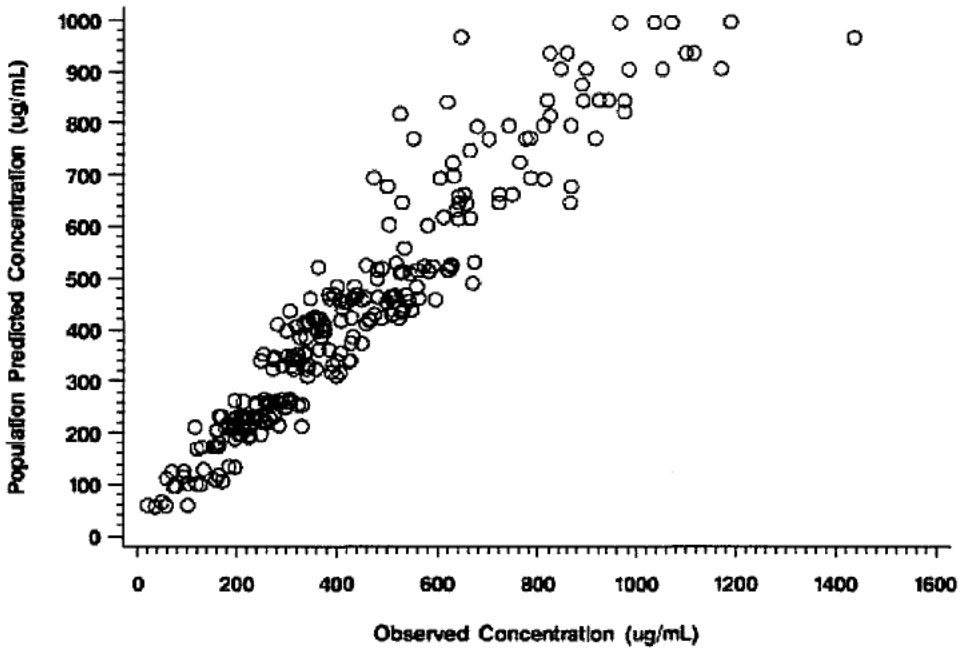
Source: Study Report AB50409-INF-0-036, Section 5.1

Figure 2. Predicted Population Average Serum Raxibacumab Concentration-Time Profiles and Individual Observed Serum Raxibacumab Concentrations Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Rabbits (Linear and Log Scale)



Source: Study Report AB50409-INF-0-036, Section 5.1

Figure 3. Population Predicted Serum Raxibacumab Concentrations Versus Individual Observed Serum Raxibacumab Concentrations Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Rabbits



Source: Study Report AB50409-INF-0-036, Appendix 8

Table 2. Summary of Model-Derived Raxiacumab Pharmacokinetic Parameters Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Rabbits

Primary Parameters	Mean	CV%
V_1 (mL)	132	15.1
CL (mL/day)	35	18.5
V_2 (mL)	57[1]	27.6
Effect of weight on V_2	$V_2 = 56.922 + (63.477 \times (\text{weight} - 3))$	
At 2.5 kg	25	
At 2.75 kg	41	
At 3.25 kg	73	
At 3.5 kg	89	
CLD_2 (mL/day)	112	NA

Secondary Parameters	Mean for 20 mg/kg	Mean for 40 mg/kg
C_{max} (µg/mL)[1]	455	909
$AUC_{0-\infty}$ (µg·day/mL)[1]	1706	3412
$t_{1/2,\alpha}$ (days)	0.24	0.24
$t_{1/2,\beta}$ (days)	3.84	3.84
MRT (days)	5.37	5.37
V_{ss} (mL)	189	189

1. Abbreviations: CV%, coefficient of variation; V_1, volume of distribution for the central compartment; CL, clearance; V_2, volume of distribution for the peripheral compartment; CLD_2, intercompartmental clearance; NA, not applicable; C_{max}, maximum serum drug concentration; $AUC_{0-\infty}$, area under the serum drug concentration-time curve from time 0 to infinite time; $t_{1/2,\alpha}$, elimination half-life for the 1st phase; $t_{1/2,\beta}$, elimination half-life for the 2nd (terminal) phase; MRT, mean residence time; V_{ss}, volume of distribution at steady-state.
2. Assuming a typical rabbit weighing 3 kg.

Source: Study Report AB50409-INF-0-036, Section 5.1

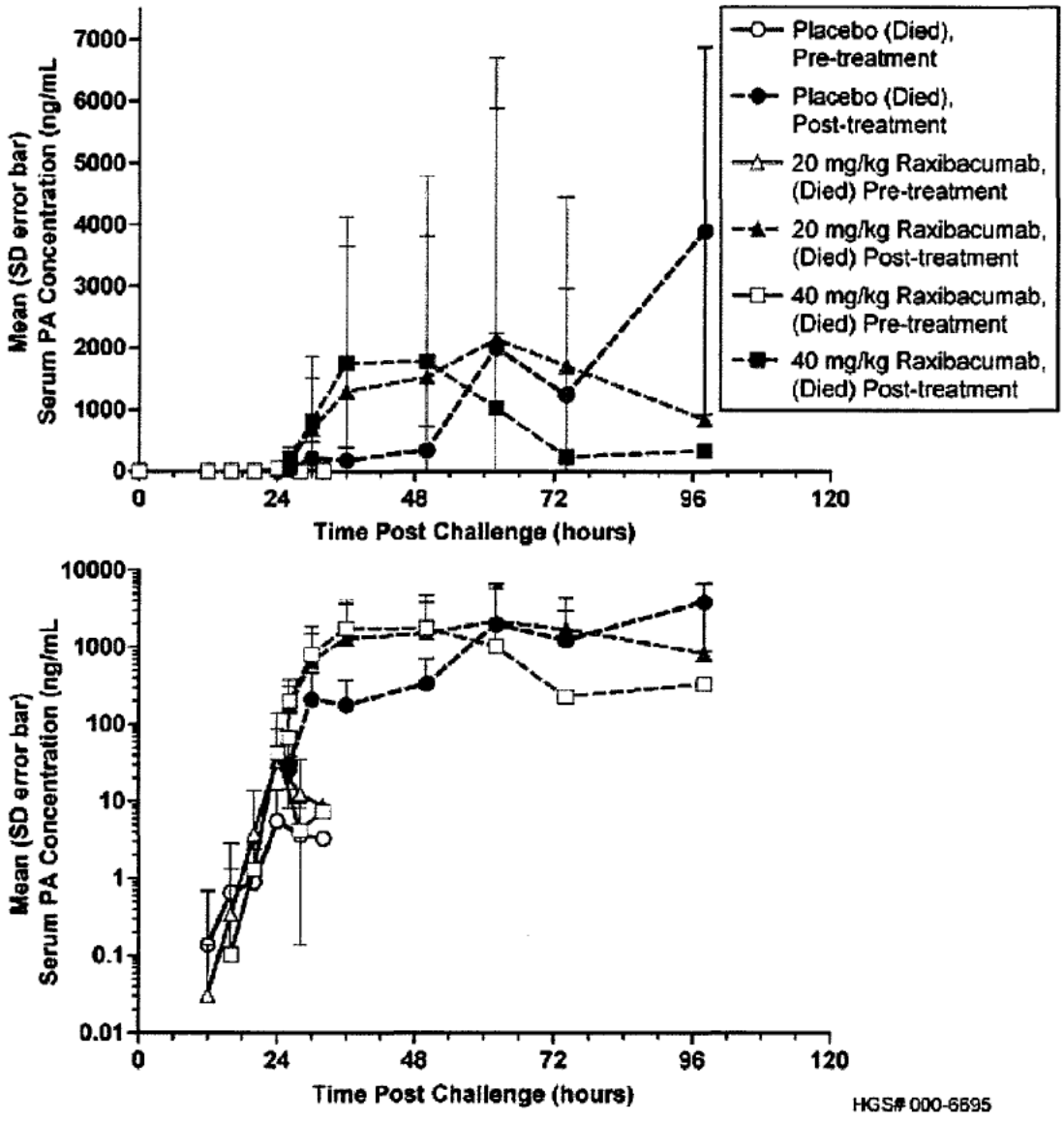
Protective Antigen (PA) Kinetics

Mean (± SD) serum PA concentration-time profiles for animals that died within each treatment group are illustrated in Figure 4. Examination of the profiles for the individual animals that died in all treatment groups revealed serum PA concentration-time profiles generally followed the pattern of rise-plateau-rise, with the exception that some animals died prior to attaining the plateau phase of the serum PA concentration-time profile. Although the mean serum PA concentrations for the rabbits that died in the 20 and 40 mg/kg dose groups are greater than those for the control group, there was substantial overlap of the SD bars among the 3 treatment groups. Relative to this, during qualification of the PA assay it was observed that the amount of PA measured increases in the presence of raxibacumab, which may, at least in part, explain the higher levels for the raxibacumab-treated groups.

Mean (± SD) serum PA concentration-time profiles for animals that survived within each treatment group are illustrated in Figure 5. Mean serum PA concentration-time profiles for the animals that survived in the 20 and 40 mg/kg raxibacumab groups showed the initial rise, but before the plateau phase was attained, serum PA concentrations began to decline. For the surviving animals in this study, the onset of declining serum PA concentrations was generally associated with attainment of negative bacteremia results. For all surviving animals in the

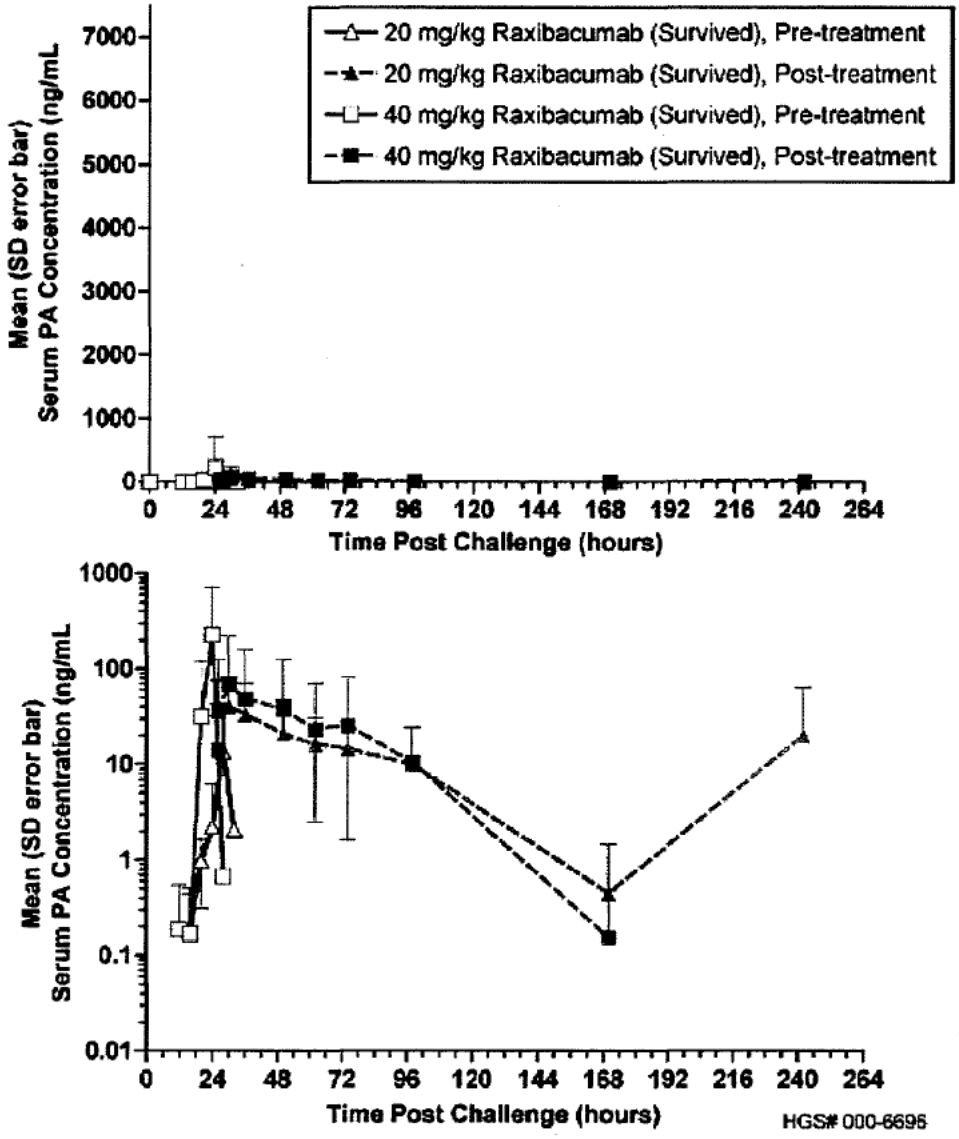
40 mg/kg dose group, serum PA concentrations declined to below the LLOQ by the end of the study. For the 20 mg/kg dose group, only 3 of the 5 surviving animals attained serum PA concentrations below the LLOQ by the end of the study.

Figure 4. Mean (± SD) Serum PA Concentration-Time Profiles in Rabbits That Died Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab and *Bacillus anthracis* Spore Challenge (Linear and Log Scale)



Source: Study Report AB50409-INF-0-036, Section 5.2

Figure 5. Mean (± SD) Serum PA Concentration-Time Profiles in Rabbits That Survived Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab and *Bacillus anthracis* Spore Challenge (Linear and Log Scale)

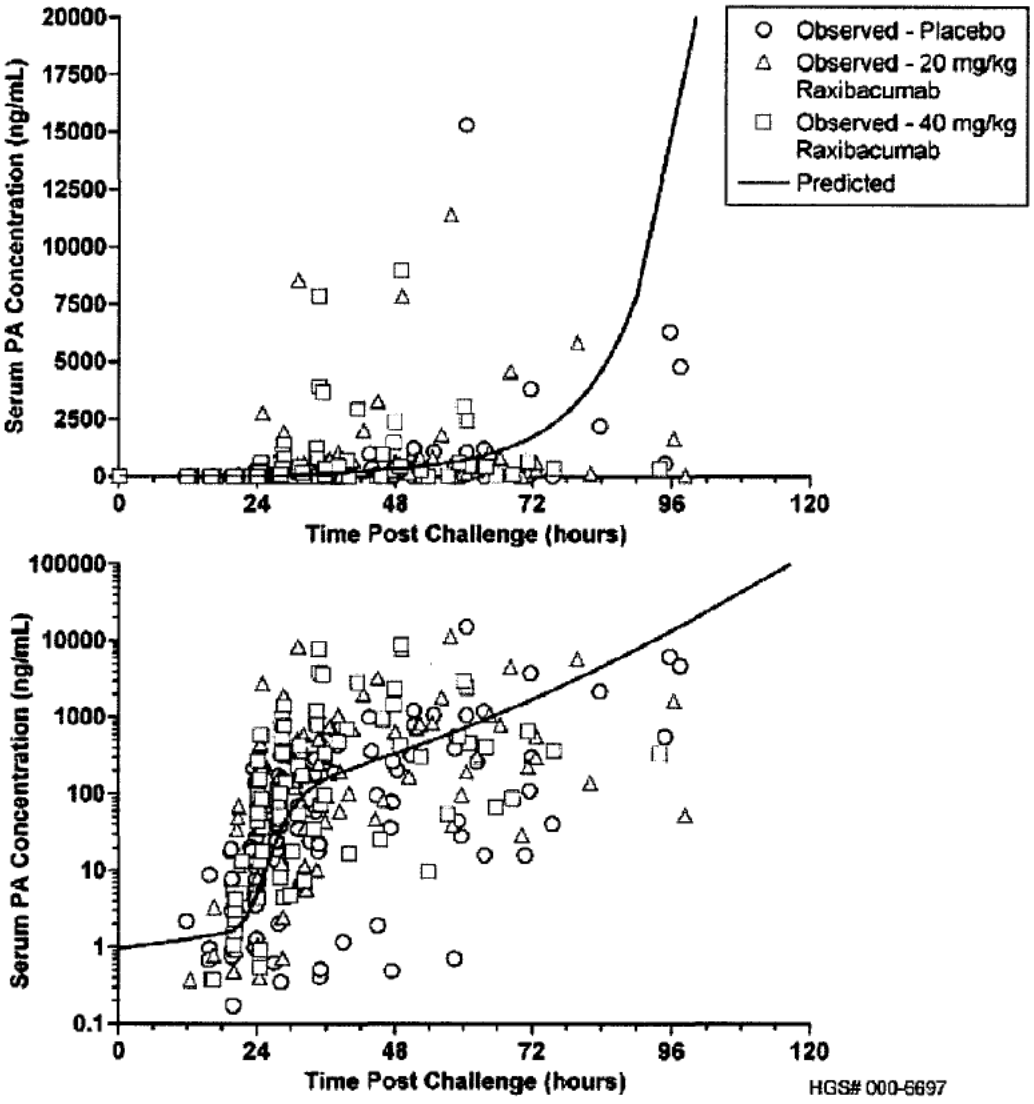


Source: Study Report AB50409-INF-0-036, Section 5.2

Individual observed serum PA concentrations versus the predicted population average serum PA concentration-time profile are displayed in Figure 6. Population predicted PA concentrations versus observed serum PA concentrations are presented in Figure 7. Parameter estimates are summarized in Table 3. For the PA kinetics modeling, data for all animals that died could be fit to the diauxic Gompertz model. The surviving animals in the 20 and 40 mg/kg dose groups had declining serum PA levels at times post dosing, and hence were inconsistent with and could not be fit to the diauxic model. The animals that died in the 20 and 40 mg/kg dose groups had serum PA concentration-time profiles that exhibited the initial rising phase and the 1st plateau phase, but some animals did not exhibit the 2nd rising or plateau phases of the diauxic model. For the

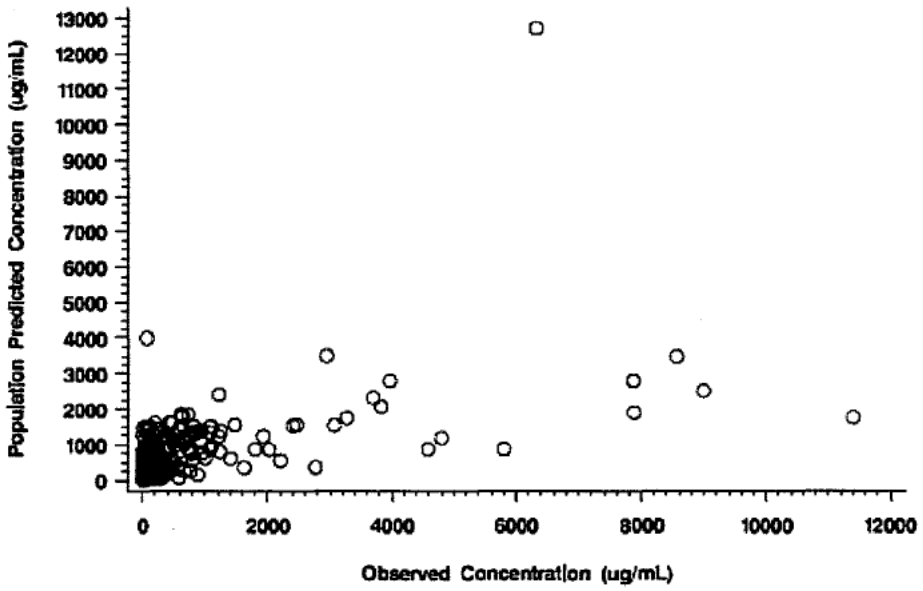
animals that died in the 20 and 40 mg/kg dose groups, it was necessary to fit a reduced model, which was essentially the standard Gompertz model (ie, only N_0, μ_m, A and λ were estimated; other parameters were fixed to zero).

Figure 6. Predicted Population Average Serum PA Concentration-Time Profiles and Individual Observed Serum PA Concentrations Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Rabbits (Linear and Log Scale)



Source: Study Report AB50409-INF-0-036, Section 5.2

Figure 7. Population Predicted Serum PA Concentrations Versus Individual Observed Serum PA Concentrations Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Rabbits



Source: Study Report AB50409-INF-0-036, Appendix 15

Table 3. Summary of Model-Derived PA Kinetic Parameters Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Rabbits

Parameters	Mean	CV%
N_0 (ng/mL)	0.625	NA
λ (h)	22	NA
Effect of time to 1st bacteremia by culture (t_B) on λ	$\lambda = 22 + (0.556 \times (t_B - 29.2))$	
At 12.2 h	13	
At 29.2 h	22	
At 94.9 h	58	
μ_m (h^{-1})	0.875	34.3
A (unitless)	7.64	72.5
Effect of survival time (t_S) on A	$A = 7.64 \times (t_S/76.3)^{-0.238}$	
At 32.7 h	9.35	
At 76.3 h	7.64	
At 166.0 h	6.35	
T_λ (unitless)	18	NA
$T_{\mu m}$ (unitless)	0.253	NA
T_A (unitless)	7.61	NA
w_α (unitless)	0.535	NA
w_β (unitless)	0.465	NA

Abbreviations: CV%, coefficient of variation; N_0, PA concentration at time 0; A, natural log of the ratio of the PA concentration in the asymptotic phase to N_0; μ_m, maximum specific growth rate; λ, lag time; T_A, the factor by which the value of A differs between the α and the β phases; $T_{\mu m}$, the factor by which the value of μ_m differs between the α and the β phases; T_λ, the factor by which the value of λ differs between the α and the β phases; w_α, weighting factor for the α phase; and, w_β, weighting factor for the β phase (1- w_α); NA, not applicable.

Source: Study Report AB50409-INF-0-036, Section 5.2

There was substantial inter-individual variability in serum PA levels. Further examination of the individual serum PA concentration-time profiles also suggests that a substantial intra-individual variability component may also exist. In addition, since each rabbit survived for a different duration (survival times range from 33 to 166 hours), each rabbit's data contributed differing amounts of information regarding the different portions of the diauxic profile. The PA kinetic results should be interpreted with caution, due to the poor definition of the variability components.

For A (relative magnitude of serum PA levels in the initial plateau phase), shorter survival times were associated with higher values for A, and longer survival times with lower values of A. This relationship would appear to be reasonable, since more morbidity could be expected to be associated with higher serum PA levels. As noted previously, for the surviving animals serum PA concentrations declined after raxibacumab treatment. The decline in serum PA concentrations coincided with the sterilization of bacteremia after raxibacumab treatment. Examination of the serum PA concentration-time profiles for the surviving rabbits in this study showed that for at least some rabbits, monotonically decreasing concentrations were present at post-raxibacumab treatment bacteremia-free collection times.

PA half-life was calculated for animals that had monotonically decreasing serum PA concentrations at times post raxibacumab dose, when the animal was bacteremia negative for

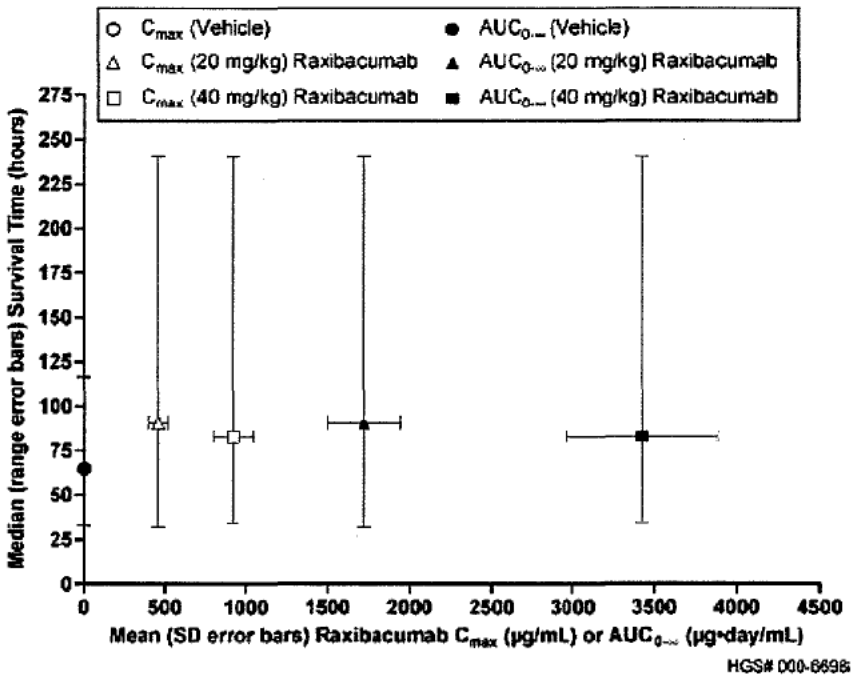
each successive collection time. Estimation of t1/2 was only possible for 4 surviving rabbits (2 in the 20 mg/kg dose group and 2 in the 40 mg/kg dose group). The overall mean t1/2 for serum PA was 30 hours, and ranged from 21 hours (2 rabbits) to 46 hours (1 rabbit).

Exposure-Survival Analysis

Based on the lowest measurable observed serum raxibacumab concentrations for any rabbit in either of the 2 raxibacumab treatment groups (20 mg/kg group - **Rabbit L08112, 19.128 µg/mL** [132 nM] at 9 days post dose [Appendix 5]; 40 mg/kg group - Rabbit L08138, **57.576 µg/mL** [397 nM] at 9 days post dose [Appendix 6]), the minimum % of serum PA bound was calculated to be 98.2% for the 20 mg/kg dose group and 99.4% for the 40 mg/kg dose group. Relative to this, the highest serum PA concentration observed in a rabbit from the 20 or 40 mg/kg dose groups prior to death was 11,400.5 ng/mL (181 nM; Rabbit No. L08132, 20 mg/kg dose group). On a nM basis, the lowest serum raxibacumab concentrations observed for either dose group, as detailed above, were similar to or ~2-fold greater in the 20 mg/kg and 40 mg/kg dose groups, respectively, than the highest serum PA concentration observed prior to death in any rabbit. Since serum raxibacumab concentrations at 5 minutes post dosing are many-fold higher than the lowest concentrations used to calculate the PA binding of 98.2 to 99.4%, it can be concluded that the exposures attained for both raxibacumab doses in this study were sufficient for virtually complete binding of PA throughout the duration of the study.

Plots of survival time as a function of raxibacumab Cmax and AUC0-∞ **are displayed in Figure 8.** Those plots show that although there was good separation of exposure for the 20 and 40 mg/kg dose groups, there was a minimal increase in mean survival time from 20 to 40 mg/kg, with substantial overlap of the survival time SD ranges between the 2 raxibacumab dose groups. Despite the lack of differentiation of survival time between the raxibacumab treatment groups, there was a marked increase in mean survival times between either raxibacumab treatment group and vehicle treatment group. This indicates that raxibacumab binding of PA is successful in alleviating mortality due to anthrax toxemia. Overall, these results indicate that there was not a well defined relationship between extent of raxibacumab exposure and survival time in the raxibacumab treated groups for the dose range studied, although survival times were increased in raxibacumab-treated animals.

Figure 8. Survival Time Versus Exposure Following Single Intravenous Administration of Vehicle, 20 mg/kg or 40 mg/kg Raxibacumab in Spore-Challenged Rabbits

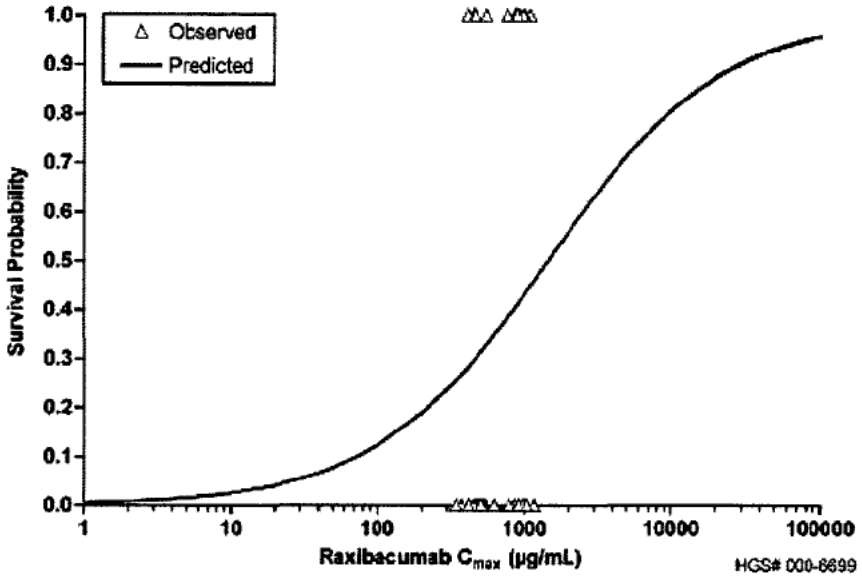


For purposes of this graph, animals in the vehicle treatment group were assigned a Cmax of 0 µg/mL and an AUC0-∞ of 0 µg·day/mL. Animals that survived were assigned a survival time of 240 hours.

Source: Study Report AB50409-INF-0-036, Section 5.3

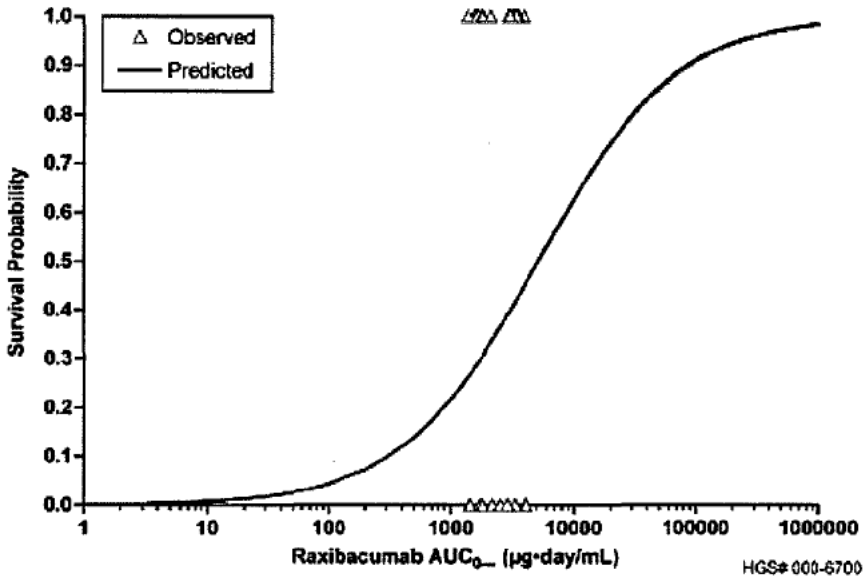
Results from the logistic regression model used to describe the relationship between the survival probability and Cmax or AUC0-∞ are presented in Figures 9 and 10, respectively. The results did not show a strong relationship for likelihood of survival to Cmax or AUC0-∞ ($p = 0.4378$ for Cmax and $p = 0.4143$ for AUC0-∞). This is likely due to a lack of differentiation between surviving animals and those that died in the raxibacumab-treated groups, with regard to magnitude of raxibacumab exposure. Exposure predictions for achieving 60, 70, 80, and 90% survival probability are shown in Table 4. The probability of achieving 60% survival corresponds to a Cmax of 2510 µg/mL, which would be attained at a dose of 110 mg/kg. The probability of achieving 60% survival corresponds to an AUC0-∞ of 8700 µg·day/mL, which would be attained at a dose of 101 mg/kg. These estimates of exposures and doses required to attain the tabulated survival probabilities must be interpreted with caution. All of the exposures predicted to result in ≥ 60% survival are at or above the ranges of observed exposures in this study. These extrapolations outside the range of observed data may not exhibit adequate accuracy, due to lack of definition for the exposure-survival relationship. Although caution is required, the predictions do seem to be consistent with the intent-to-treat survival data for this study; 28% and 44% survival at 20 and 40 mg/kg, respectively, with the numerical difference in survival at 40 mg/kg vs 20 mg/kg not attaining statistical significance. The logistic regression predictions for 28% and 44% survival are 400 and 1042 µg/mL for Cmax, respectively, and 1540 and 3796 µg·day/mL for AUC0-∞, respectively. For 20 and 40 mg/kg, the mean Cmax were 455 and 909 µg/mL, respectively, and the mean AUC0-∞ were 1706 and 3412 µg·day/mL, respectively, showing that the logistic regression predictions are in reasonable agreement with the observed data.

Figure 9. Logistic Regression Analysis Between Survival Probability and Cmax Following Single Intravenous Administration of Vehicle, 20 mg/kg or 40 mg/kg Raxibacumab in Spore-Challenged Rabbits



Source: Study Report AB50409-INF-0-036, Section 5.3

Figure 10. Logistic Regression Analysis Between Survival Probability and AUC0-∞ Following Single Intravenous Administration of Vehicle, 20 mg/kg or 40 mg/kg Raxibacumab in Spore-Challenged Rabbits



Source: Study Report AB50409-INF-0-036, Section 5.3

Table 4. Predicted Raxibacumab Cmax and AUC0-∞ Corresponding to 60, 70, 80, and 90% Survival Probability Based on Logistic Regression Analysis

	Survival Probability[1]			
	60%	70%	80%	90%
C_{max} (µg/mL)	2510	4575	9514	28640
Dose (mg/kg) to attain target C_{max}[2]	110	201	419	1260
$AUC_{0-\infty}$ (µg·day/mL)	8700	15333	30612	86624
Dose (mg/kg) to attain target $AUC_{0-\infty}$[2]	101	179	357	1011

1. Results were obtained from the logistic regression modeling.
2. Assuming linear PK, for a 3 kg rabbit.

Source: Study Report AB50409-INF-0-036, Section 5.3

APPLICANT'S CONCLUSIONS:

- Serum raxibacumab concentrations in rabbits administered raxibacumab were best fit to a two compartment model with 1st-order elimination from the central compartment.
- V2 is dependent on body weight, with V2 increasing as body weight increases. V1 is about the same as the plasma volume; however, the data indicate that raxibacumab does distribute to tissues.
- In this study, raxibacumab PK appear to be independent of sex, magnitude of spore challenge, and survival status. PK were linear over the dose range evaluated. Inter-individual variability in PK was low.
- Serum total PA (free + bound) profiles in rabbits that died can be fit to a diauxic Gompertz model (rise-plateau-rise). Rabbits that survived had profiles inconsistent with

that model, in that serum PA concentrations tended to decrease at times coincident with negative bacteremia after raxibacumab dosing.

- For **rabbits that died, λ (lag time for serum PA levels) is associated with time to 1st** bacteremia by culture, while A (1st phase plateau serum PA concentration) is associated with survival time.
- In surviving rabbits, t1/2 for PA during the post raxibacumab dose bacteremia-free period averaged 30 hours and ranged from 21 to 46 hours.
- The exposures attained for both raxibacumab doses in this study were sufficient for virtually complete binding of PA (based on observed exposures and in vitro binding kinetics).
- There was not a well-defined relationship between extent of raxibacumab exposure and survival time in the raxibacumab-treated groups for the dose range studied, although survival times were increased in raxibacumab-treated animals over those that were administered placebo.

REVIEWER ASSESSMENT:
Results from Study AB50409.INF.0.036 adequately described the pharmacokinetics of raxibacumab following a single intravenous (IV) raxibacumab dose and the the kinetics of *Bacillus anthracis* protective antigen (PA) in rabbits with inhalation anthrax. The applicant's pharmacokinetic conclusions based on these findings are valid.

Based on in vitro binding kinetics, serum concentrations of approximately 40 μg/mL are required for 99% binding of PA. In the current study, serum concentrations of raxibacumab were greater **than 40 μg/mL in all surviving animals in both dose groups through** Day 6 of sampling. In the PK samples obtained from animals that did not survive, serum concentrations of raxibacumab were many fold greater that that required for 99.9% binding of PA. This supports the applicant's conclusion that the exposures attained for both raxibacumab doses in this study were sufficient for virtually complete binding of PA.

Results from the applicant's logistic regression analysis describing the relationship between the survival probability and exposure did not show a strong relationship for likelihood of survival to Cmax or AUC0-∞ ($p = 0.4378$ for Cmax and $p = 0.4143$ for AUC0-∞). As suggested by the applicant, this is likely due to a lack of differentiation between surviving animals and those that died in the raxibacumab-treated groups, with regard to magnitude of raxibacumab exposure. Figures 11 and 12 below clearly illustrate the lack of differentiation in exposure between surviving animals and those that died between the two dose groups. As displayed in the graphs, concentration-time profiles and the ranges of exposure measures for the two dose groups demonstrated considerable overlap. Although definition of an exposure-response relationship for raxibacumab and survival would facilitate extrapolation of animal efficacy findings to humans, the application of the applicant's logistic regression findings in the current is limited by the degree of overlapping exposure between surviving animals and animals that died.

Figure 11. Spaghetti Plot of Serum Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Rabbits

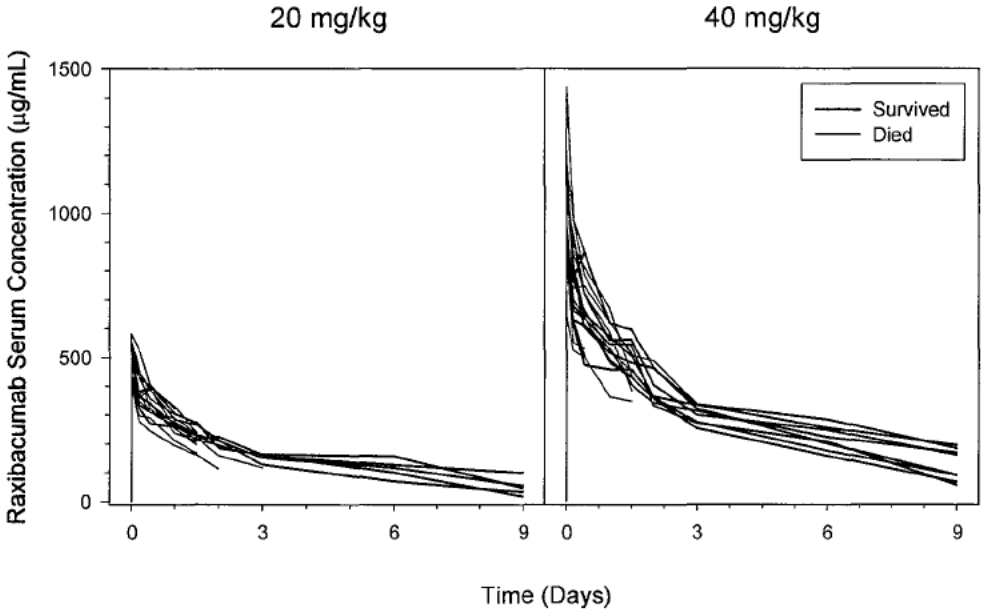
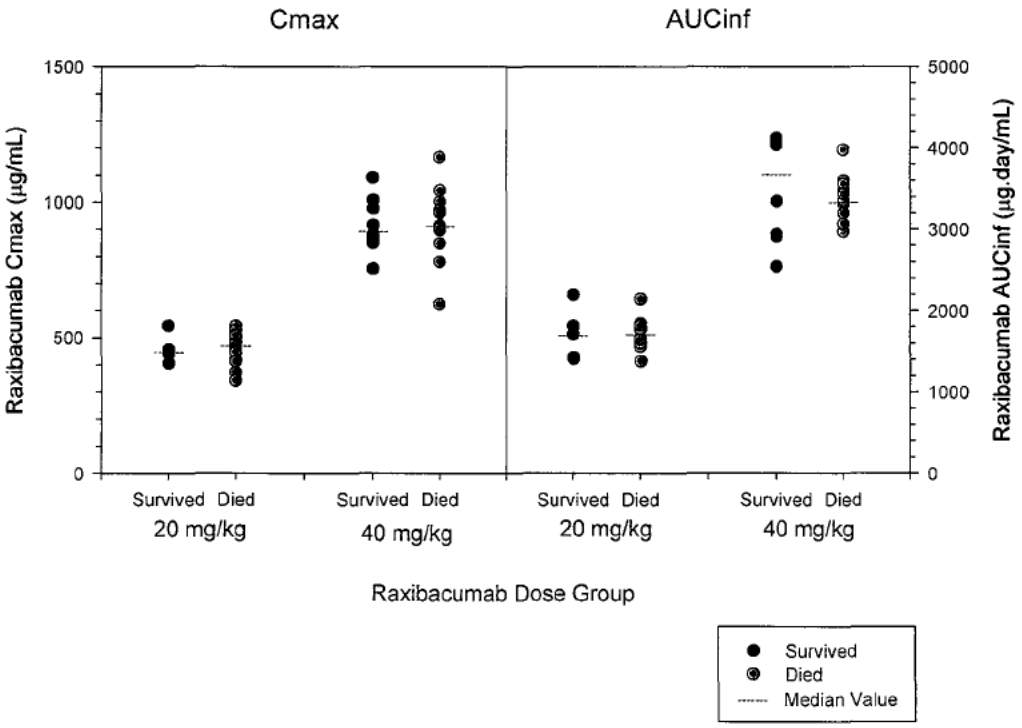


Figure 12. Comparison of Raxibacumab Cmax and AUCinf Between Animals that Survived and Animals that Died Following *Bacillus anthracis* Spore Challenge and Treatment with Single Intravenous Administration of 20 mg/kg or 40 mg/kg Raxibacumab



In addition, the applicant used the logistic regression model to predict exposures and doses that would achieve 60, 70, 80, and 90% survival probability. As the applicant suggests, the logistic regression predictions are in reasonable agreement with the observed data for Cmax and AUC0-∞. In the current study, reported survival rates were 28% and 44% for 20 and 40 mg/kg, respectively. The logistic regression predictions for 28% and 44% survival are 400 and 1042 μg/mL for Cmax, respectively, and 1540 and 3796 μg·day/mL for AUC0-∞, respectively. For 20 and 40 mg/kg, the mean Cmax were 455 and 909 μg/mL, respectively, and the mean AUC0-∞ were 1706 and 3412 μg·day/mL, respectively, showing that the logistic regression predictions are in reasonable agreement with the observed data. As the applicant emphasizes, the estimates of exposures and doses required to attain survival probabilities greater than those observed in the rabbit study must be interpreted with caution, as they are extrapolations outside the range of observed data.

The applicant conducted a cross-study comparison to examine the PK of raxibacumab in healthy rabbits compared to those with anthrax. In a previous study (HGS Report AB50409.INF.0.016, 2003), raxibacumab PK were determined following single IV bolus 1 and 10 mg/kg doses in healthy rabbits, over a 28 day period post dose. In the current study, serum raxibacumab levels were measured for up to 9 days post dose. The cross-study comparison suggests that raxibacumab was cleared from the plasma more rapidly (shorter terminal half-life) in rabbits with anthrax than in healthy rabbits. This is confirmed by comparison of CL for healthy rabbits, which ranged from 6.2 to 6.9 mL/kg/day, with that for rabbits with anthrax, at 11.7 mL/kg/day. Similarly, t1/2,β in healthy rabbits ranged from 6.9 to 8.7 days, while in rabbits with anthrax, t1/2,β was shorter, at 3.8 days. However, t1/2,α ranged from 0.24 to 0.40 days in healthy rabbits, and was similar for the rabbits with anthrax disease in the current study (0.24 days), indicating that the differences in the profiles are related to elimination processes rather than initial distribution processes. This is corroborated by the similarity of V1 in healthy rabbits (range: 36 to 39 mL/kg) and for the rabbits with anthrax in the current study (44 mL/kg). Vss in healthy rabbits (63 to 67 mL/kg) was similar to that in rabbits with anthrax (189 mL, or 63 mL/kg for a 3 kg rabbit). Based on the applicant's interpretation, the differences in CL and t1/2,β may be a reflection of altered physiology in anthrax disease, binding of raxibacumab to PA, or a combination of those factors. Altered physiology in anthrax disease could result in greater distribution of raxibacumab, resulting in faster apparent clearance from plasma. Raxibacumab bound with PA could result in a complex that is more rapidly cleared than raxibacumab alone. Also of importance, the manufacturing processes used for raxibacumab differed between the two studies used for comparison which could also influence the disposition of raxibacumab. Thus, the applicant's conclusion that it is not possible to differentiate the contributions of these possible causes to the differences in PK between healthy monkeys and monkeys with anthrax infecton based on the results of this study is valid.

AB50409.INF.0.043

Levofloxacin and Raxibacumab Pharmacokinetics, with Protective Antigen Kinetics, During the Evaluation of the Efficacy of Raxibacumab in Combination with Levofloxacin for Post-Exposure Treatment in the New Zealand White Rabbit Inhalation Anthrax Model (Battelle Study No. 781-G923701)

OBJECTIVES:

- To determine the pharmacokinetics (PK) of intragastric (IG) levofloxacin doses and of an intravenous (IV) raxibacumab dose when coadministered, as well as the kinetics of *Bacillus anthracis* protective antigen (PA), in rabbits with inhalation anthrax

STUDY DESIGN:

Fifty-two (52) New Zealand white rabbits that weighed 2.97 to 4.02 kg were randomized by gender (50% male and 50% female in each group) and body weight into one of three treatment groups, challenge day, and challenge order. Animals were randomized into two raxibacumab dosing groups of 20 animals each and one control group of 12 animals and into three aerosol challenge sets of animals. The 3 sets of monkeys were aerosol challenged on 3 staggered challenge days (Day 0). On Study Day 0, monkeys were anesthetized and individually aerosol challenged with a targeted 200 x lethal dose in 50% of the tested animals (LD_{50}, 1.24 x 105,000 CFU) inhaled dose of *B. anthracis* (Ames strain). Following spore challenge, animals were frequently monitored for increased temperature and PA toxemia (every 4 hours from 16 to 72 hours post-spore challenge). Plasma PA was measured using an electrochemiluminescence (ECL)-based screening assay. Immediately after onset of either the first 2 consecutive hourly body temperature measurements 2 or more degrees Fahrenheit higher than the individual rabbits' baseline average temperature, or the 1st detection of measurable plasma PA for an individual rabbit, the rabbit was treated with an IG 50 mg/kg levofloxacin dose or an IG placebo (sterile water for injection) dose, according to treatment group assignment, which was in turn followed by 1 bolus IV injection of 40 mg/kg raxibacumab or raxibacumab vehicle (placebo, 1.0 mL/kg), according to treatment group assignment. Any animal that had not been treated up to the 72 hour post-median challenge time point was treated as soon as the last hourly temperature measurement was obtained. IG 50 mg/kg levofloxacin or placebo doses were administered every 24 hours (qd) for a total of 3 doses (3 days of treatment). In summary, Group assignments were as follows: Group 1, levofloxacin placebo plus raxibacumab vehicle (n = 12); Group 2, levofloxacin 50 mg/kg plus raxibacumb vehicle (n = 20); and Group 3, levofloxacin 50 mg/kg plus raxibacumab 40 mg/kg (n = 20).

FORMULATIONS:

Raxibacumab (Lot 71044) and raxibacumab vehicle (Lot 71043) were supplied as ready-to-use sterile liquid formulations. Raxibacumab vehicle was raxibacumab formulation buffer (0.13 mg/mL citric acid, 2.8 mg/mL sodium citrate, 10 mg/mL sucrose, 18 mg/mL glycine, 0.2 mg/mL polysorbate 80, pH 6.5). Raxibacumab was provided as a solution (50 mg/mL) in the formulation buffer. During dosage periods, raxibacumab and vehicle were stored on ice. Prior to administration, raxibacumab was aseptically diluted to concentrations of 40 mg/mL in formulation buffer.

Levaquin® Oral Solution (levofloxacin 25 mg/mL) was used. Levaquin Oral Solution is a multi-use self-preserving aqueous solution of levofloxacin with pH ranging from 5.0–6.0. Its appearance may range from clear yellow to clear greenish-yellow. It contains the following inactive ingredients: sucrose, glycerin, sucralose, hydrochloric acid, purified water, propylene glycol, artificial and natural flavors, benzyl alcohol, ascorbic acid, and caramel color. It may also contain a solution of sodium hydroxide for pH adjustment. Levaquin Oral Solution was

administered IG as supplied, without dilution. The vehicle control for Levaquin Oral Solution was sterile water for injection, which was administered IG as supplied. Levaquin Oral Solution (levofloxacin 25 mg/mL; (b) (4) lot #7DB4J00) was obtained from a commercial source. The vehicle control for Levaquin Oral Solution was sterile water for injection purchased from a commercial supplier (b) (4)

PHARMACOKINETIC ASSESSMENTS:
Blood specimens for determination of levofloxacin plasma concentrations were collected from all rabbits at 7 days prior to spore challenge and just prior to the 1st dose; at 2 hours after administration of each dose; at 24 hours after each dose (just prior to the subsequent dose); and at 48 hours after the 3rd dose. When feasible, a terminal blood sample was taken just prior to euthanasia for animals that were judged to be moribund.

Blood specimens for determination of raxibacumab plasma concentrations were collected from all rabbits at 7 days prior to spore challenge; just prior to levofloxacin, raxibacumab, or vehicle dosing; at 5 minutes after dosing; at 8, 24, 48, and 96 hours after dosing; and at 7, 14, 21, and 28 days after spore challenge. When feasible, a terminal blood sample was taken just prior to euthanasia for animals that were judged to be moribund.

Blood specimens for plasma PA kinetics were collected from all rabbits at 7 days prior to spore challenge as well as at 16, 20, 24, 28, 32, and 36 hours after spore challenge, unless treatment occurred prior to the collection time. Additional blood specimens were collected just prior to levofloxacin, raxibacumab, or vehicle dosing; at 5 minutes after dosing; at 8, 24, 48, and 96 hours after dosing; and at 7, 14, 21, and 28 days after spore challenge. When feasible, a terminal blood sample was taken just prior to euthanasia for animals that were judged to be moribund.

BIOANALYTICAL ANALYSIS:
Plasma samples were analyzed for raxibacumab using an ECL-based assay. The raxibacumab in diluted serum samples binds to the biotinylated-PA on a streptavidin-coated plate and is detected by the addition of goat anti-human antibody labeled with MSD SULFOTAG™, an ECL label. The concentration of raxibacumab in serum samples is interpolated from a reference standard curve. The lower limit of quantitation (LLOQ) is 750 ng/mL of raxibacumab in 100% rabbit serum.

Plasma samples were analyzed for levofloxacin using a high performance liquid chromatography/ mass spectroscopy/mass spectroscopy (LC/MS/MS) assay. The calibration range for the assay was from 40.40 to 16159.00 ng/mL.

PA concentrations in plasma samples were determined using an ECL-based bridging assay. In brief, diluted plasma samples in duplicate wells are combined with rabbit polyclonal antibody (pAb) anti-PA-biotin (capture) and rabbit pAb anti-PA-SULFO-TAG (detector) and allowed to equilibrate in a streptavidin-coated assay plate. Following equilibration, plates are washed and read for ECL counts. The concentration of PA in plasma samples and controls is interpolated from a reference standard curve. The assay must meet system suitability criteria, as outlined in the SOP, to be valid. The LLOQ is 0.56 ng/mL of PA in 100% rabbit plasma.

Samples were also assayed for anti-PA antibody concentrations and toxin neutralization activity (TNA) titers. Anti-PA Ab concentrations in serum samples were determined using an enzyme-linked immunosorbent assay (ELISA). The LLOQ is 125 ng/mL of anti-PA Ab in 100% rabbit serum. TNA titers in serum samples were determined using a cell killing assay. In brief,

inhibition of PA binding is correlated with increased cell viability and is used to determine the relative titer of serum samples compared to a known positive control. The LLOQ is < 52 titer in 100% rabbit serum.

Plasma samples were assessed for anti-raxibacumab activity (immunogenicity) using an ECL-based assay. If a plasma sample for the screening assay (Assay A) wzas identified as a potential positive, it was further tested in Assay B, the inhibition of binding assay, to confirm the specificity of binding. If the specificity was confirmed, the sample was considered anti-raxibacumab positive.

PHARMACOKINETIC AND KINETIC ANALYSIS:
PK analyses of raxibacumab concentration-time profiles for all raxibacumab-dosed rabbits were conducted using population analysis techniques (mixed effect modeling utilizing NONMEM). One (1)-, 2- and 3-compartment models with 1st order elimination from the central compartment were evaluated. Body weight, sex, age, size of spore challenge, duration of spore challenge, survival time, survival status, time to 1st bacteremia by culture, bacteremia outcome at each collection time, and immunogenicity status were evaluated as potential covariates for the PK parameters.

Kinetic analyses of total PA concentration-time profiles for control group rabbits were conducted using population analysis techniques, with the NONMEM software. For individual animals that survived, the 1st order elimination rate constant and half-life of elimination (t1/2) for serum PA concentrations during the bacteremia-free post-treatment period were calculated, using actual collection time postchallenge. Body weight, sex, age, size of spore challenge, duration of spore challenge, survival status, time to 1st bacteremia by culture, and immunogenicity status were evaluated as potential covariates.

Plasma levofloxacin concentration-time profiles for the subjects were analyzed individually. The maximum plasma levofloxacin concentration after each dose (Cmax,n) was defined as the concentration measured 2 hours after the nth dose, while the minimum plasma drug concentration after the nth dose (Cmin,n) was defined as the concentration measured just prior to the subsequent dose, or for the 3rd dose, at 24 hours after that dose.

Plasma levofloxacin concentration-time data, plasma raxibacumab concentration-time data, plasma PA concentration-time data, individual levofloxacin PK parameters, individual raxibacumab PK parameters, individual PA kinetic parameters, plasma anti-PA Ab concentrations, and plasma TNA titers were summarized using descriptive statistics (number of observations [N], mean, standard deviation [SD], standard error [SE], coefficient of variation [CV%], minimum, median, maximum, geometric mean, and 95% confidence interval [CI]).

Unpaired t-tests were used to compare levofloxacin Cmax,n and Cmax,n between Group 2 and Group 3. For animals that survived, an unpaired t-test was used to compare PA t1/2 between Group 2 and Group 3. Unpaired t-tests were used to compare plasma anti-PA Ab or TNA concentrations at corresponding collection times between Group 2 and Group 3. All comparisons were carried out at the $\alpha = 0.05$ **level of significance.**

RESULTS:
A summary of sex, age and body weight for the rabbits is presented in Table 1.

Table 1. Baseline Characteristics of Rabbits in Study 781-G923701

Group	Number of Animals (N)	Age (yr)	Weight (kg)	Sex
1 (placebo/vehicle)	12	0.82 (0.75 – 0.89)	3.5 (3.27 – 3.80)	6 Females 6 Males
2 (levofloxacin/vehicle)	20	0.82 (0.75 – 0.89)	3.45 (3.07 – 3.87)	10 Females 10 Males
3 (levofloxacin/raxibacumab)	20	0.82 (0.75 – 0.89)	3.52 (2.97 – 4.02)	10 Females 10 Males

Data presented as mean (range).

Source: Study Report AB50409-INF-0-043, ST-2

For three rabbits (Rabbits K99218, K99220, and K99228 [control group]), a terminal blood specimen was not collected. For Rabbit K99261 (control group), a terminal blood specimen was collected, but a low volume was obtained; hence, there was only sufficient plasma available for levofloxacin analysis. There were no other missing specimens or results for any of the other animals.

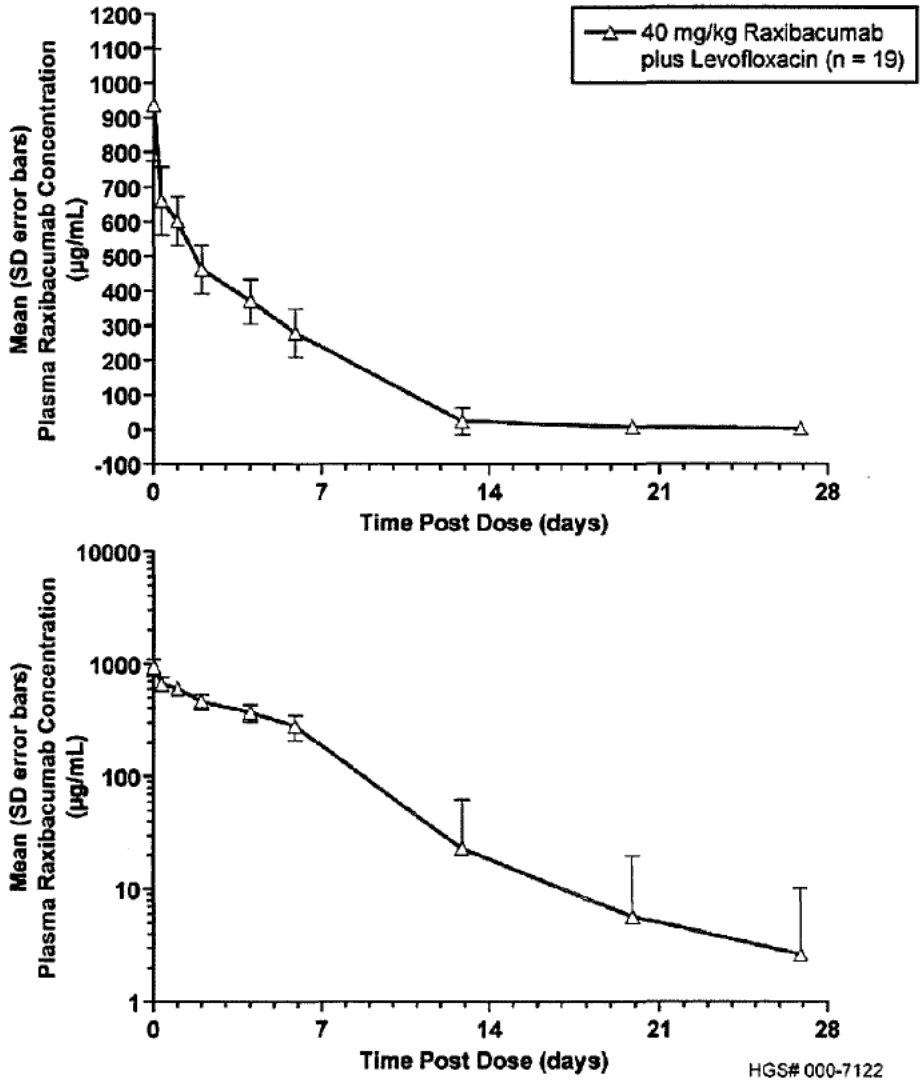
Raxibacumab Plasma Pharmacokinetics

Mean (± SD) plasma concentration-time profiles for raxibacumab following single intravenous administration 40 mg/kg raxibacumab in combination with levofloxacin 50 mg/kg qd for 3 doses in NZW rabbits are presented in Figure 1. Population predicted concentrations versus observed serum raxibacumab concentrations are presented in Figure 2. PK parameter estimates are summarized in Table 2. The plasma raxibacumab concentrations best fit a 2-compartment open model with 1st-order elimination from the central compartment. A biphasic apparent elimination profile was noted for several rabbits, with apparent accelerated elimination in the latter portion of the profile. Immunogenicity results indicated 18 out of 19 rabbits included in the analysis had anti-raxibacumab Ab titers that were positive in the immunogenicity screening assay on Day 14 post-challenge. The apparent increase in raxibacumab clearance in the later portion of the profile is associated with the presence of anti-raxibacumab antibodies in many of the individual rabbits at those times, and immunogenicity outcome was found to be a significant covariate for CL. Body weight was found to be a significant covariate for V1. Other factors assessed (sex, age, size of spore challenge, duration of spore challenge, time to 1st bacteremia by culture, and bacteremia outcome at each collection time) were not significant covariates accounting for inter-individual differences in PK. Survival time and survival status were not assessed as potential covariates due to the high survival rate for rabbits treated with levofloxacin plus raxibacumab.

In the raxibacumab treatment group, one rabbit (Rabbit K99251) had no measurable plasma raxibacumab concentrations at any time post dose. Since all other raxibacumab-treated animals had substantial measurable plasma raxibacumab concentrations for up to at least the 7 days post-challenge collection time, it appeared that Rabbit K99251 was misdosed with placebo, rather than raxibacumab. Although no such dosing error was documented, the results for Rabbit K99251 were excluded from the analyses. In the levofloxacin alone group, a plasma raxibacumab concentration was encountered in 1 specimen (1/178 [0.6%] post treatment specimens). The measured raxibacumab concentration was 1.224 µg/mL and occurred at the 7 days post-challenge collection time. For comparison, for animals treated with raxibacumab, the lowest plasma raxibacumab concentration for any animal at 7 days post-challenge was 119.16 µg/mL, more than 100-fold higher than that detected in the animal from the levofloxacin only treatment group.

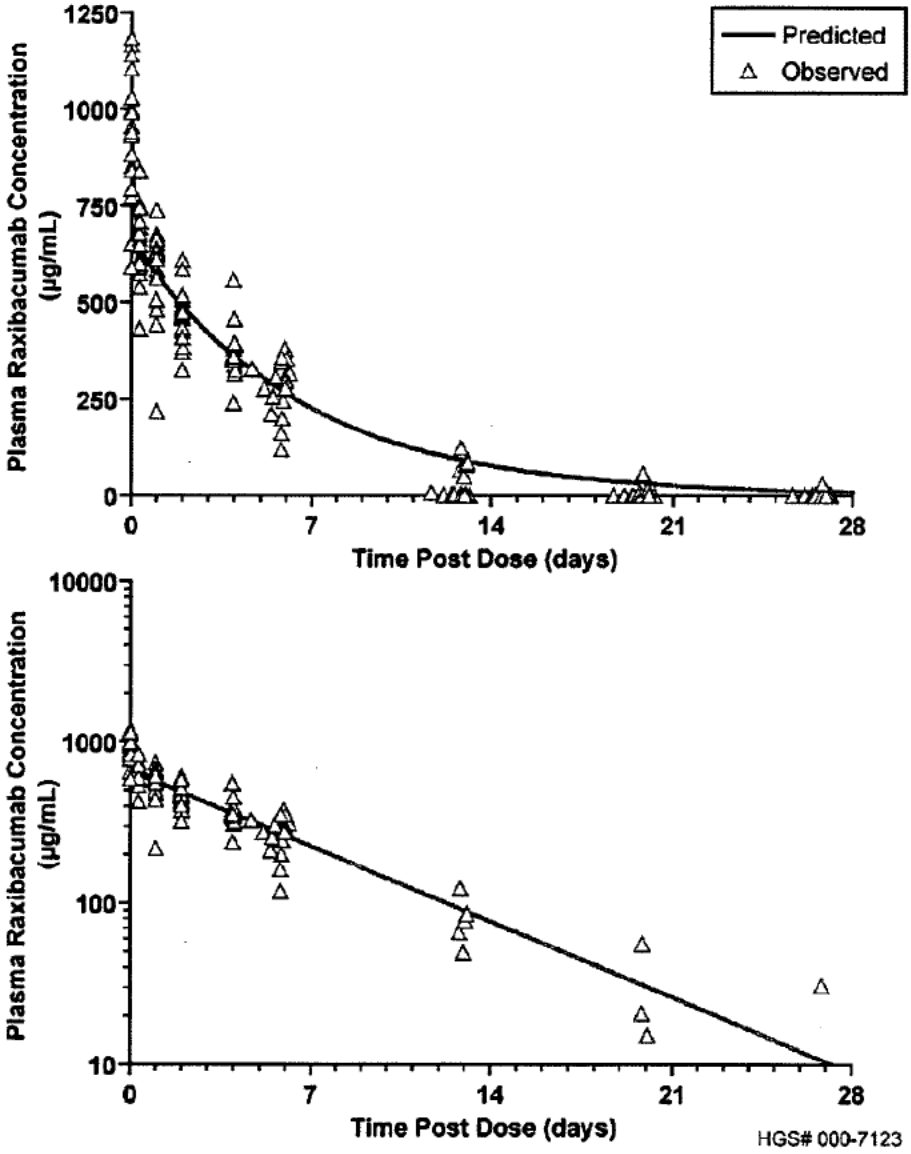
Hence, the plasma raxibacumab concentration for the animal in the levofloxacin only group is inconsistent with inadvertent administration of raxibacumab and was excluded.

Figure 1. Mean (± SD) serum concentration-time profiles for raxibacumab following single intravenous administration 40 mg/kg raxibacumab in combination with levofloxacin 50 mg/kg qd for 3 doses in NZW rabbits (linear and log scale)



Source: Study Report AB50409-INF-0-043, Section 5.2

Figure 2. Predicted population average serum raxibacumab concentration-time profiles and individual observed serum raxibacumab concentrations following single intravenous administration 40 mg/kg raxibacumab in combination with levofloxacin 50 mg/kg qd for 3 doses in NZW rabbits (linear and log scale)



Source: Study Report AB50409-INF-0-043, Section 5.2

Table 2. Summary of raxibacumab pharmacokinetic parameters following single intravenous administration 40 mg/kg raxibacumab in combination with levofloxacin 50 mg/kg qd for 3 doses in NZW rabbits

Parameter	Units	Mean (CV%)
Primary Model-Derived Parameters		
V1	mL	150 (13.3)
CL Anti-raxibacumab Ab negative	mL/day	31.92 (23.4)
CL Anti-raxibacumab Ab positive	mL/day	64.08 (23.4)
V2	mL	55.3 (29.1)
CLD2	mL/day	302.4 (NA)
Secondary Parameters		
Cmax	µg/mL	928
AUC0-∞	µg·day/mL	4361
t1/2,α	days	0.09
t1/2,β	days	4.49
MRT	days	6.43
Vss	mL	58.99

Volumes calculated assuming a typical rabbit weighing 3.48 kg and a dose of 40 mg/kg.
Abbreviations: CV%, coefficient of variation; V1, volume of distribution for the central compartment; CL, clearance; V2, volume of distribution for the peripheral compartment; CLD2, intercompartmental clearance; NA, not applicable; Cmax, maximum serum drug concentration; AUC0-∞, area under the serum drug concentration-time curve from time 0 to infinite time; t1/2,α, elimination half-life for the 1st phase; t1/2,β, elimination half-life for the 2nd (terminal) phase; MRT, mean residence time; Vss, volume of distribution at steady-state.

Source: Study Report AB50409-INF-0-043, Section 5.2

Following IV raxibacumab administration, V1, at 150 mL (43 mL/kg for a 3.48 kg animal), is similar to the plasma volume (44 mL/kg). The steady-state volume of distribution (Vss) is 37% greater than V1, at 59 mL/kg for a 3.48 kg animal. These results suggest that although distribution of raxibacumab may initially be restricted to the plasma volume, raxibacumab does subsequently distribute to tissues. V1 increases with increasing weight

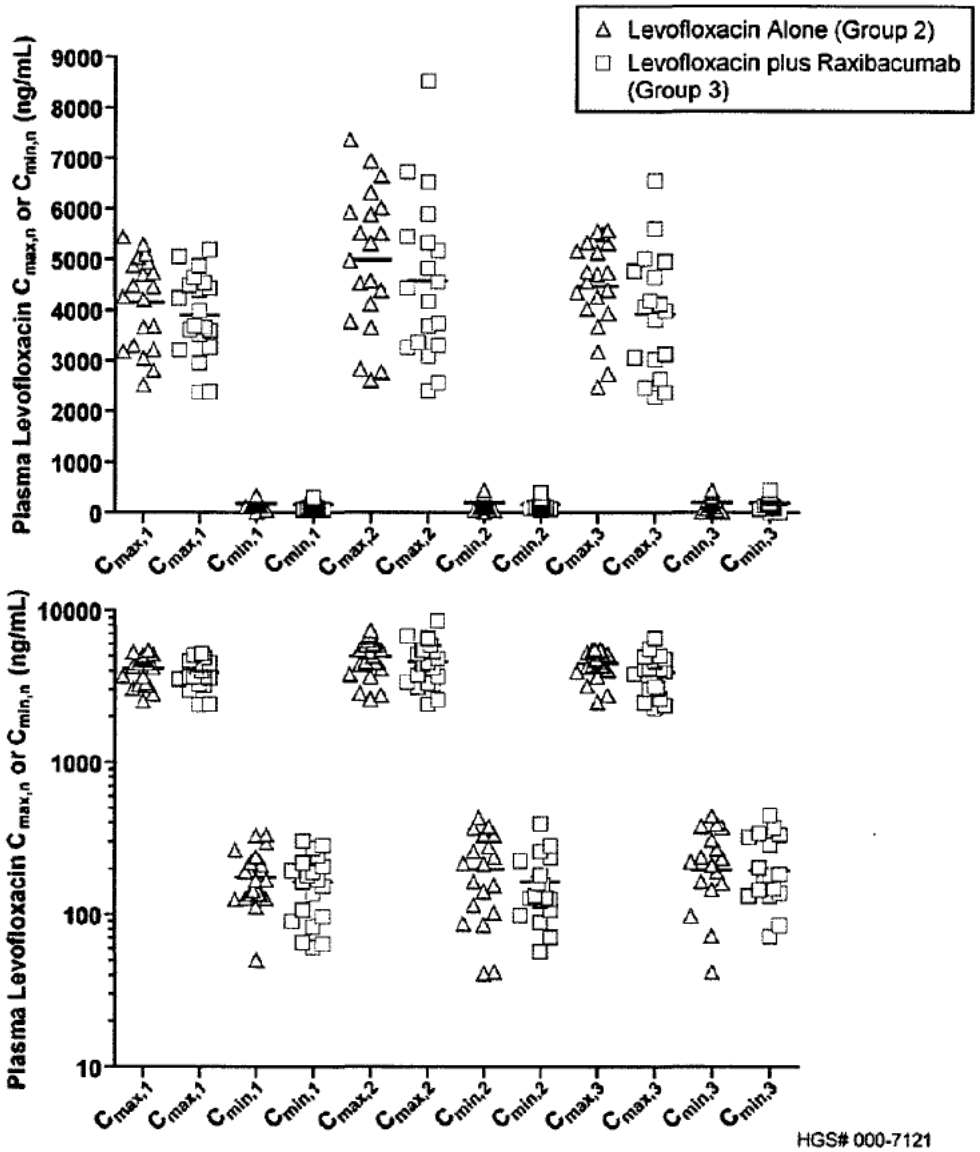
The disappearance of raxibacumab from plasma is multiphasic, with an initial phase elimination half-life (t1/2,α) of 0.09 days. The terminal phase elimination half-life (t1/2,β) is 4.49 days for an anti-raxibacumab Ab negative rabbit. In the absence of anti-raxibacumab Ab, the mean CL of raxibacumab was 31.92 mL/day (9.2 mL/day/kg for a 3.48 kg rabbit), whereas CL was 64.08 mL/day (18.4 mL/day/kg for a 3.48 kg rabbit) in the presence of anti-raxibacumab Ab. For either anti-raxibacumab Ab status, CL is much less than the glomerular filtration rate (4493 mL/day/kg), indicating there is virtually no renal clearance of this monoclonal antibody. Inter-individual variability in raxibacumab PK was low, with CV% of 29% or less for the primary PK parameters. Overall, these results indicate that there is minimal variability in raxibacumab disposition, once the impact of body weight on V1 and anti-raxibacumab Ab status on CL are taken into account, even in rabbits exhibiting symptoms of inhalation anthrax.

Levofloxacin Plasma Pharmacokinetics

Mean and individual observed plasma concentrations for levofloxacin in the levofloxacin alone and levofloxacin plus raxibacumab-treated groups (Groups 2 and 3, respectively) following multiple IG administration of 50 mg qd for 3 doses in NZW rabbits are presented in Figure 3. For

one rabbit (Rabbit K99222; levofloxacin plus raxibacumab group), the results for the 48 hour and 50 hour collection times appeared to have been switched (the peak and trough values appeared to be reversed); hence, those results were excluded. The Cmax,n and Cmin,n results for Group 2 and Group 3 are summarized in Table 3. While comparisons of Cmax,n and Cmin,n across days within a dose group show some differences, there is no consistent pattern of increasing or decreasing values across days, suggesting that steady-state was attained prior to the 2nd levofloxacin dose in Group 2 and Group 3. In addition, unpaired t-tests of Cmax,n and Cmin,n between Group 2 and Group 3 show no differences that could be attributed to altered levofloxacin PK after raxibacumab administration.

Figure 3. Mean and individual observed plasma concentrations for levofloxacin in the levofloxacin alone and levofloxacin plus raxibacumab-treated groups (Groups 2 and 3, respectively) following multiple IG administration of 50 mg qd for 3 doses in NZW rabbits (linear and log scale)



Line represents mean at the specified time point.
Source: Study Report AB50409-INF-0-043, Section 5.1

Table 3. Summary of levofloxacin Cmax and Cmin concentrations in the levofloxacin alone and levofloxacin plus raxibacumab-treated groups (Groups 2 and 3, respectively) following multiple IG administration of 50 mg qd for 3 doses in NZW rabbits

	Group 2 Levofloxacin Alone		Group 3 Levofloxacin Plus Raxibacumab		
	N	Mean ± SD	N	Mean ± SD	P-Value[1]
$C_{max,1}$ (ng/mL)	20	4148.6 ± 899.6	20	3907.5 ± 817.8	0.3808
$C_{min,1}$ (ng/mL)	20	176.4 ± 86.4	20	165.0 ± 76.1	0.6599
$C_{max,2}$ (ng/mL)	20	4983.2 ± 1402.3	19	4583.7 ± 1581.0	0.4086
$C_{min,2}$ (ng/mL)	20	199.5 ± 125.8	18	165.2 ± 86.3	0.3396
$C_{max,3}$ (ng/mL)	20	4470.2 ± 913.7	18	3934.9 ± 1200.9	0.1285
$C_{min,3}$ (ng/mL)	20	197.5 ± 137.0	19	195.7 ± 124.7	0.9667

Abbreviations: $C_{max,n}$, maximum plasma levofloxacin concentration after the n^{th} dose, defined as the concentration measured 2 hour after the dose; $C_{min,n}$, minimum plasma levofloxacin concentration after the n^{th} dose, defined as the concentration measured just prior to the subsequent dose, or at 24 hours after the 3rd dose; NA, not applicable.

[1] From an unpaired t-test.

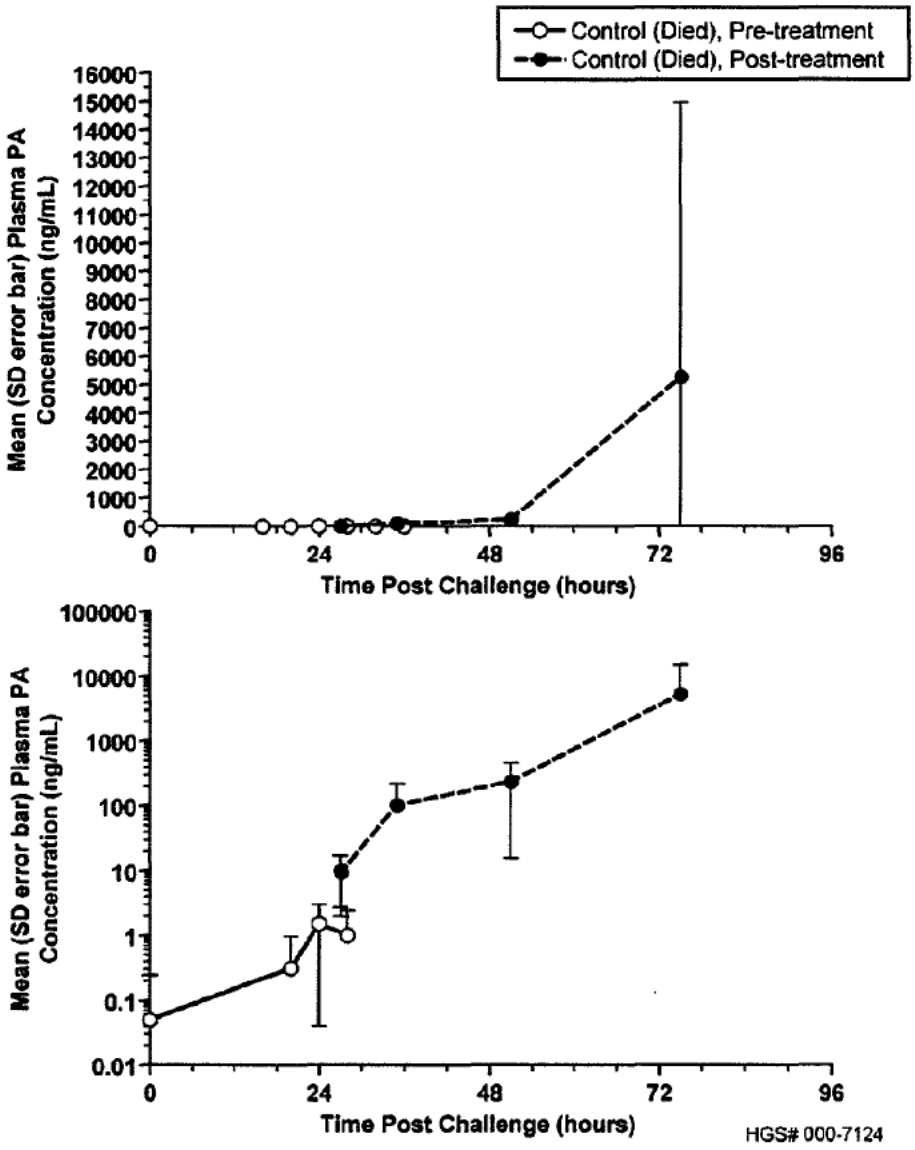
Source: Study Report AB50409-INF-0-043, Section 5.1

Protective Antigen (PA) Kinetics

Mean (± SD) serum PA concentration-time profiles for control animals included in the analysis are illustrated in Figure 4. All animals in the control group died, whereas 1 animal (Rabbit K99203) died in the levofloxacin alone group and 1 animal (Rabbit K99246) died in the levofloxacin plus raxibacumab group. Rabbit K99203 in the levofloxacin alone group died, but that animal's plasma PA concentration-time profile showed a decreasing pattern consistent with the effect of the drug. Since that pattern is inconsistent with the kinetic model, this animal was excluded from the modeling analysis. Rabbit K99246 in the levofloxacin plus raxibacumab group died, but necropsy results for that animal indicated gavage error as the cause of death. Therefore, that animal was also excluded from the analysis. As a result, the PA kinetic analysis for animals that died was based on animals in the control group.

In animals that died, plasma PA concentration-time profiles generally followed the pattern of rise-plateau-rise, with the exception that some animals died prior to attaining either the plateau phase or the 2nd rising phase of the plasma PA concentration-time profile. These findings are consistent with previous studies in rabbits.

Figure 4. Mean (± SD) serum PA concentration-time profiles in rabbits that died following single intravenous administration of placebo (linear and log scale)



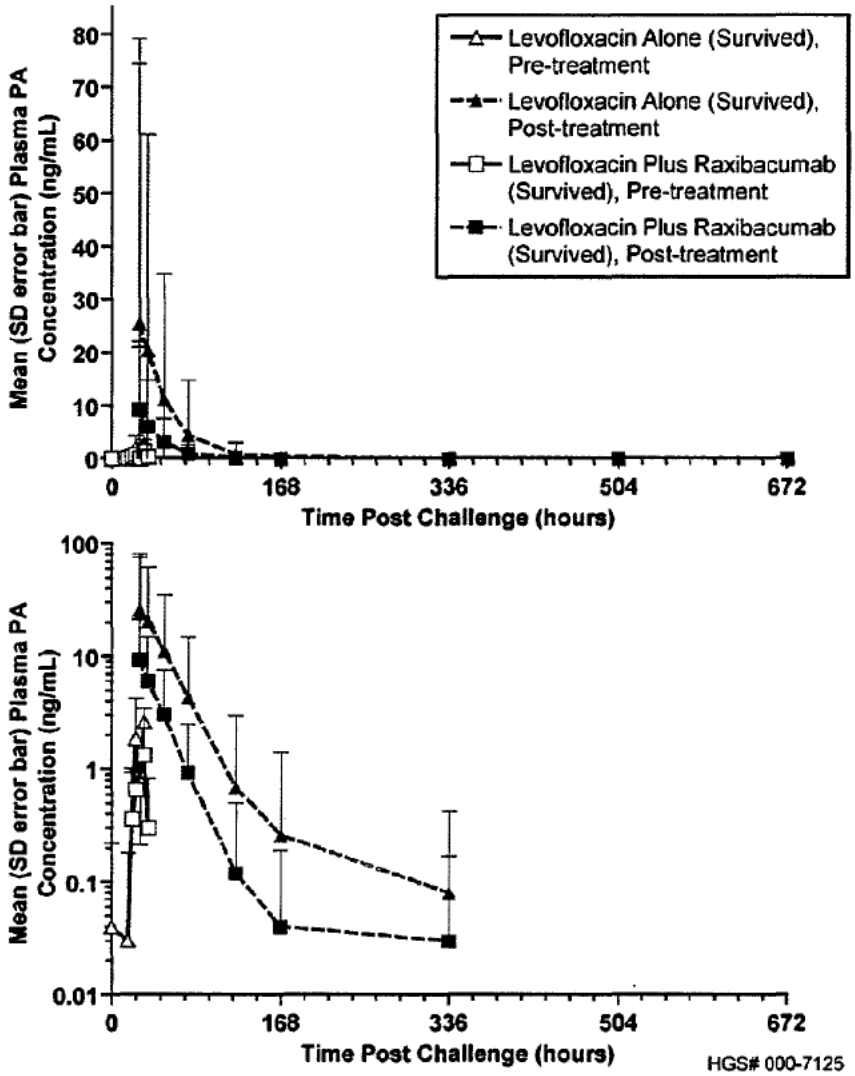
For plotting purposes, post-treatment collection times are expressed as times post challenge based on a typical treatment time of approximately 27 hours post challenge. Solid lines represent serum PA concentrations prior to treatment intervention, while dotted lines represent serum PA concentrations after treatment intervention.

Source: Study Report AB50409-INF-0-043, Section 5.3

Mean (± SD) plasma PA concentration-time profiles for animals that survived within each treatment group are illustrated in Figure 5. Mean plasma PA concentration-time profiles showed the initial rise, but before or shortly after the plateau phase was attained, serum PA concentrations began to decline. This increasing then decreasing type of PA profile is generally characteristic of surviving animals, and is generally not observed in those animals that died. In previous studies, the decline of serum PA levels was observed to generally coincide with the sterilization of bacteremia. Similarly, for the surviving animals in this study, the onset of declining serum PA concentrations was generally associated with attainment of negative bacteremia results.

For all but two of the surviving animals, plasma PA concentrations declined to below the LLOQ by 14 days post challenge. Rabbit K99254 (levofloxacin alone group) survived, and had a low but measurable plasma PA concentration of 1.468 ng/mL at 14 days post-challenge. However, at collections times from 24 hours post treatment to 7 days post-challenge, plasma PA concentrations were undetectable for that animal, as well as at the 21 and 28 days post-challenge collection times. Similarly, Rabbit K99214 (levofloxacin + raxibacumab group) survived, and had a low but measurable plasma PA concentration of 0.616 ng/mL at 14 days post-challenge. For this rabbit also, the measurable 14 days post-challenge result was preceded by nondetectable PA concentrations from 48 hours post treatment through 7 days post-challenge, and was followed by nondetectable concentrations at the 21 and 28 days post-challenge collection times. Hence the measurable concentrations for those 2 animals at 14 days post-challenge were deemed by the applicant to be spurious results.

Figure 5. Mean (± SD) plasma PA concentration-time profiles in rabbits that survived in the levofloxacin alone and levofloxacin plus raxibacumab-treated groups (linear and log scale)



For plotting purposes, post-treatment collection times are expressed as times post challenge based on a typical treatment time of approximately 27 hours post challenge. Solid lines represent serum PA concentrations prior to treatment intervention, while dotted lines represent serum PA concentrations after treatment intervention.

Source: Study Report AB50409-INF-0-043, Section 5.3

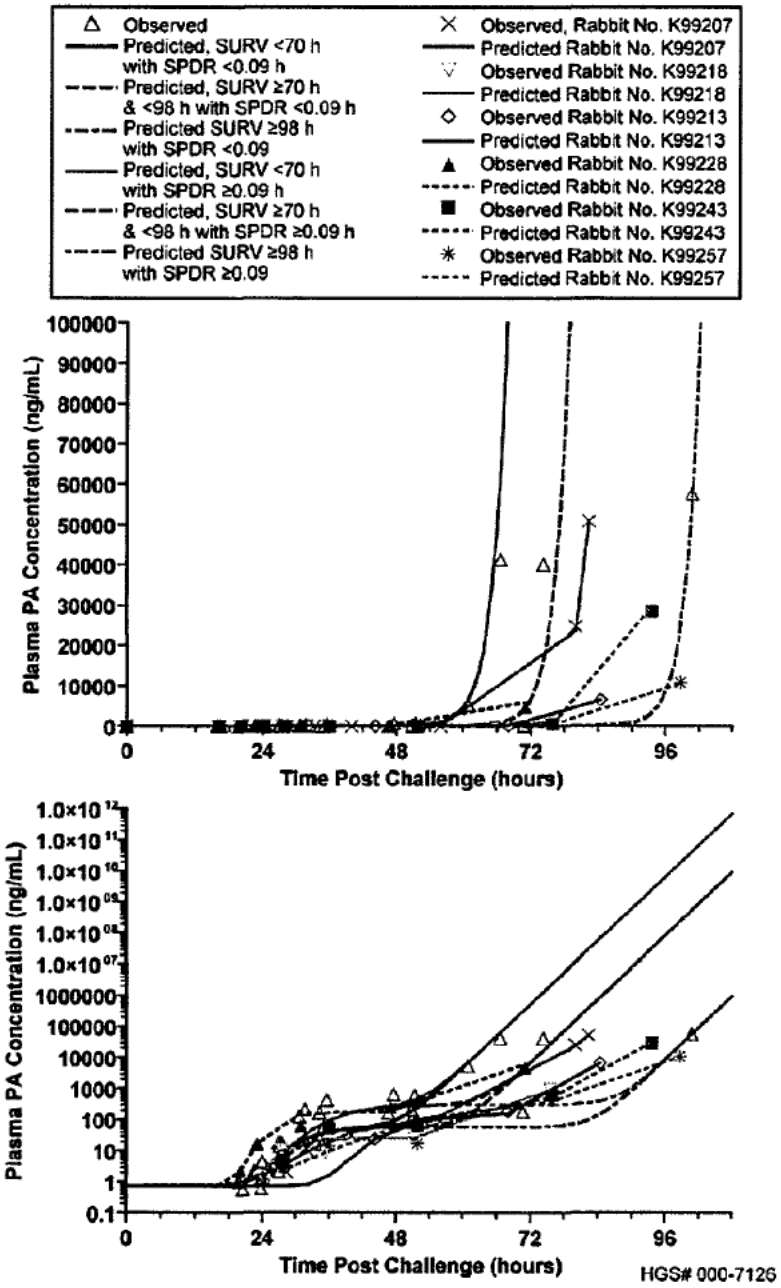
Individual observed plasma PA concentrations versus the predicted population average plasma PA concentration-time profiles are displayed in Figure 6. Parameter estimates are summarized in Table 4. There was substantial inter-individual variability in plasma PA levels, and there were relatively few animals in the population modeled. Despite several attempts, it was not possible to fit the data for all animals that died to the diauxic Gompertz model, likely due to the sparseness of the data and its large variability. This was addressed by first obtaining estimates for the standard Gompertz model (rise–plateau), using a subset of the data for timepoints consistent with that model. The parameter estimates from that run were then applied to a simplified diauxic model. In

addition, estimates of λ were stratified based on survival time. Estimates of μ_m, $\mu_{m,2}$, λ, or T_λ were obtained separately for some rabbits. Modeling of inter-individual variability for all of the model parameters was attempted, but was not successful, or did not result in an improved fit to the data. In the final model, interindividual variability could not be estimated for N_0, T_A, or T_λ. T_λ was stratified by survival time, and duration of spore challenge was a significant covariate for A. No other significant covariate effects were found. The PA kinetic results should be interpreted with caution, due to the poor definition of the variability components and the relatively small sample size.

Some variability in $T\lambda$ was associated with survival time. Longer lag times for the 2nd phase, resulting in a more prolonged plateau in the 1st phase, were related to longer survival times. In addition, some variability in A was related to duration of spore challenge, with shorter spore challenge related to lower PA concentrations in the plateau phase. Review of the plasma PA concentration-time results for the surviving animals, in comparison with the results of bacteremia by culture tests, revealed that the decline in plasma PA concentrations coincided with the sterilization of bacteremia after treatment, as noted in a previous study. It is unknown if that reflects an anti-bacterial effect of raxibacumab, or if it reflects the ability of the animals' immune response to sterilize bacteremia once the mortality-inducing effects of toxemia were neutralized by raxibacumab. Examination of the plasma PA concentration-time profiles for the surviving rabbits in this study showed that for at least some rabbits, monotonically decreasing concentrations were present at post-levofloxacin/raxibacumab treatment bacteremia-free collection times. Those profiles generally did not show increasing rates of PA clearance; hence, it seemed reasonable to assume that the clearance of PA from plasma was not related to the development of anti-PA Ab in these animals.

Half-life of PA was calculated for animals that had monotonically decreasing serum PA concentrations at times post raxibacumab dose, when the animal was bacteremia negative for each successive collection time. Estimation of t1/2 was only possible for 13 surviving rabbits in the levofloxacin alone dose group and 9 in the levofloxacin plus raxibacumab dose group. The mean t1/2 for plasma PA was 16.80 hours for the levofloxacin alone dose group, and was 45.51 hours for the levofloxacin plus raxibacumab dose group, while the median t1/2 for plasma PA were similar at 16.63 (range: 12.27 to 25.21) and 15.96 (range: 11.35 to 269.56) hours for the levofloxacin alone and levofloxacin plus raxibacumab dose groups, respectively. The higher mean t1/2 for the levofloxacin plus raxibacumab dose group is due to one individual animal with a very large value for t1/2. Therefore, comparison of the median t1/2 between the dose groups is more appropriate, and shows no meaningful difference between the two dose groups.

Figure 6. Predicted population average plasma PA concentration-time profiles and individual observed plasma PA concentrations in NZW rabbits that died which received levofloxacin placebo and raxibacumab vehicle (linear and log scale)



SURV, survived
SPDR, spore challenge duration

Source: Study Report AB50409-INF-0-043, Section 5.3

Table 3. Summary of model-derived PA kinetic parameters for rabbits that died which received levofloxacin placebo and raxibacumab vehicle

Parameters	Mean	CV%
N_0 (ng/mL)	0.738	NA
λ (h)	21.4	7.82
For Rabbit No. K99213	37.1	
μ_m (h^{-1})	0.378	
For Rabbit No. K99207	0.268	53.0
For Rabbit No. K99228	1.02	
A (unitless), for spore challenge duration < 0.09 h	4.36	13.9
A (unitless), for spore challenge duration ≥ 0.09 h	6.02	
T_λ (h)		
For survival times <70 hours	17.0	
For survival times ≥70 hours and <98 hours	28.1	NA
For survival times ≥98 hours	51.5	
For Rabbit No. K99218	36.5	
$\mu_{m,2}$ (h^{-1})	0.392	
For Rabbit No. K99257	0.362	5.43
For Rabbit No. K99243	0.233	
For Rabbit No. K99207	0.317	
Residual variability (CV%)	28.0	

Abbreviations: CV%, coefficient of variation; N_0, PA concentration at time 0; A, natural log of the ratio of the PA concentration in the asymptotic phase to N_0; μ_m, maximum specific growth rate; λ, lag time; $\mu_{m,2}$, maximum specific growth rate for the 2nd phase; T_λ, the factor by which the value of λ differs between the 1st and the 2nd phases; NA, not applicable.

Source: Study Report AB50409-INF-0-043, Section 5.3

Exposure-Survival Analysis

Survival rates were very high in the levofloxacin alone and levofloxacin plus raxibacumab dose groups, with only one rabbit in the levofloxacin alone group dying of anthrax. Although it was intended to assess survival time as a function of raxibacumab or ciprofloxacin exposure, the minimal number of animals that died when administered the two drugs prevented any meaningful assessment from being made.

Anti-PA Antibodies

Plasma anti-PA Ab concentration results for the levofloxacin alone and levofloxacin plus raxibacumab treatment groups are summarized in Table 4. Since two of the rabbits in the levofloxacin alone dose group appeared to be outliers, the analysis was also performed excluding the values for those two animals. The mean plasma anti-PA Ab concentration at 28 days post challenge for the levofloxacin alone dose group was numerically higher (48%) than that for the levofloxacin plus raxibacumab dose group, although the difference did not attain statistical significance. For the analysis which included the two outlier animals, the difference between dose groups was numerically larger (about 2-fold), but did not attain statistical significance. It should be noted that difference in Day 28 anti-PA Ab concentrations was not reflected in altered survival between the 2 groups, suggesting that the anti-PA Ab response in rabbits administered both levofloxacin and raxibacumab was adequate for survival.

Table 4. Summary of plasma anti-PA antibody concentrations in surviving rabbits administered following multiple IG administration of 50 mg qd levofloxacin for 3 doses alone or in combination with a single IV 40 mg/kg raxibacumab dose

		Plasma Anti-PA Antibody Concentration (µg/mL)[1]	
		Prechallenge	28 Day Postchallenge
Levofloxacin Alone	N	17	17
	Mean ± SD	0.0148 ± 0.0609	78.237 ± 64.875
Levofloxacin Plus Raxibacumab	N	19	19
	Mean ± SD	0.000 ± 0.000	52.851 ± 40.799
	p-Value[2]	0.3322	0.1643

Source: Study Report AB50409-INF-0-043, Section 5.5

Toxin Neutralization Activity

Plasma TNA concentrations for the levofloxacin alone and levofloxacin plus raxibacumab treatment groups are summarized in Table 5. The mean plasma TNA titer at 28 days post-challenge for the levofloxacin alone dose group was significantly higher (about 3-fold, $p = 0.0050$) than that for the levofloxacin plus raxibacumab dose group. It should be noted that these differences were not reflected in altered survival between the 2 groups, suggesting that the TNA response in rabbits administered both ciprofloxacin and raxibacumab was adequate for survival.

Table 5. Summary of plasma TNA titers in surviving rabbits administered qd x 3 IG 50 mg/kg levofloxacin doses alone or in combination with a single IV 40 mg/kg raxibacumab dose

		Plasma TNA Titer[1]	
		Prechallenge	28 Day Postchallenge
Levofloxacin Alone	N	19	19
	Mean ± SD	0 ± 0	2099.8 ± 1944
Levofloxacin Plus Raxibacumab	N	19	19
	Mean ± SD	0 ± 0	648.84 ± 505.98
	p-Value[2]	-	0.0050

Source: Study Report AB50409-INF-0-043, Section 5.6

APPLICANT'S CONCLUSIONS:

- Co-administration of IV raxibacumab had no effect on levofloxacin exposure following IG levofloxacin dosing.
- Plasma raxibacumab concentrations in rabbits were best fit to a 2 compartment model with 1st-order elimination from the central compartment. CL is dependent on anti-raxibacumab Ab status, increasing when positive for anti-raxibacumab Ab. V1 is dependent on body weight, increasing as body weight increases. In this study,

raxibacumab PK appear to be independent of sex, age, size of spore challenge, duration of spore challenge, time to 1st bacteremia by culture, and bacteremia outcome at each collection time.

- Despite the presence of anthrax disease at the time of raxibacumab administration, inter-individual variability in PK was low.
- Plasma total PA (free + bound) profiles in rabbits that died can be fit to a diauxic model (rise-plateau-rise). Rabbits that survived had profiles inconsistent with that model, in that plasma PA concentrations tended to decrease at times coincident with negative bacteremia. For rabbits that died, lag time for the 2nd phase (longer plateau phase) is associated with longer survival time. Higher plasma PA levels at plateau are associated with longer duration of the spore challenge. In surviving rabbits, the median t1/2 for PA during the post raxibacumab dose bacteremia-free period were 16 to 17 hours.
- The exposures attained for the 40 mg/kg IV raxibacumab dose in this study were sufficient for binding of the majority of PA (based on observed exposures and in vitro binding kinetics).
- The mean anti-PA antibody concentrations and plasma TNA titers were 48% and about 3-fold higher, respectively, for levofloxacin alone, relative to levofloxacin plus raxibacumab. However, the high survival rates in these dose groups indicate that these differences were not meaningful.

REVIEWER ASSESSMENT:
Results from Study AB50409.INF.0.043 adequately determined the PK of IG levofloxacin doses and of an IV raxibacumab dose when coadministered, as well as the kinetics of *Bacillus anthracis* protective antigen (PA), in rabbits with inhalation anthrax. Overall, the applicant's pharmacokinetic conclusions based on these findings are valid.

Based on a cross-study comparison, PK results from Study 781-G923701 showed slight differences when compared to findings from the previous pivotal rabbit efficacy study (Study 682-G005758). Mean (SD) raxibacumab serum concentration-time profiles and PK parameters following single IV administration of raxibacumab 40 mg/kg with and without levofloxacin in rabbits are presented in Figure 7 and Table 6, respectively. Rabbits that received raxibacumab in combination with levofloxacin in Study 781-G923701 exhibited a higher AUC compared to rabbits in the pivotal animal efficacy Study 682-G005758. Mean Cmax was comparable across studies. In the current study, raxibacumab PK differed based on the presence of anti-raxibacumab Ab. In the absence of anti-raxibacumab Ab, the mean CL of raxibacumab was 31.92 mL/day (9.2 mL/day/kg for a 3.48 kg rabbit), whereas CL was 64.08 mL/day (18.4 mL/day/kg for a 3.48 kg rabbit) in the presence of anti-raxibacumab Ab. This finding could have contributed to the variability in PK observed across studies. Since the current study did not include a raxibacumab alone control group, interpretation of these differences is limited and does not affect conclusions based on current study findings. In addition, the current study measured ***plasma*** concentrations of raxibacumab; raxibacumab concentrations obtained in Study 682-G005758 were determined in ***serum***. Since mean Cmax values were comparable across studies, a matrix effect accounting for PK differences between the two studies is unlikely.

Figure 7. Mean (SD) raxibacumab concentration-time profiles following single IV administration of raxibacumab 40 mg/kg with (Study 781-G923701) and without (Study 682-G005758) levofloxacin in rabbits

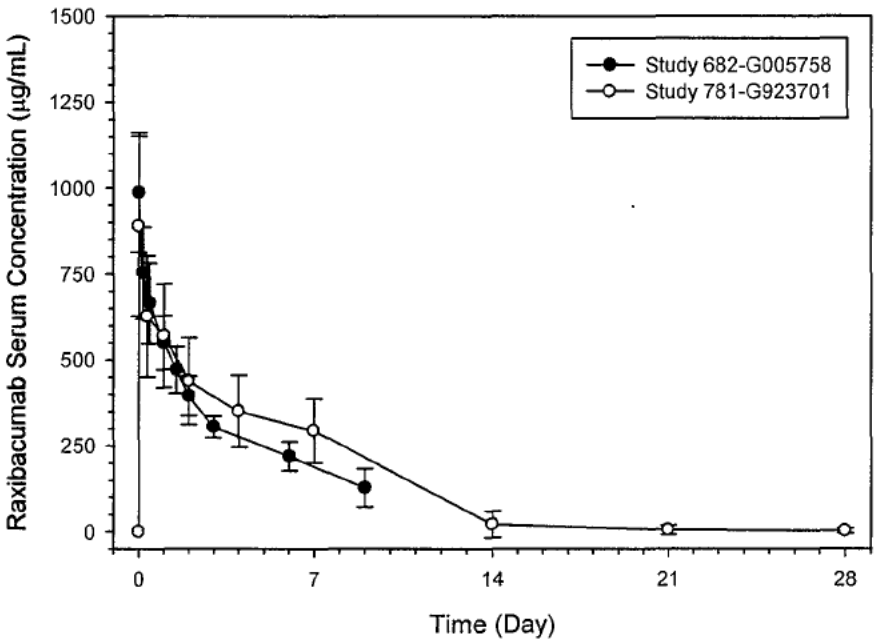


Table 6. PK parameters following single IV administration of raxibacumab 40 mg/kg with (Study 781-G923701) and without (Study 682-G005758) levofloxacin in rabbits

Parameter	Study	
	682-G005758	781-G923701
Cmax (µg/mL)	919	928
AUC0-∞ (µg·day/mL)	3424	4361
T1/2β (day)	4.01	4.49
Vss (mL)	196	59
CL (mL/day)	36	Ab negative: 32 Ab positive: 64

In the current study, rabbits received intragastric levofloxacin 50 mg/kg or placebo (sterile water for injection), according to treatment group assignment. Intragastric 50 mg/kg levofloxacin or placebo doses were then administered every 24 hours (qd) for a total of 3 doses (3 days of treatment). The recommended oral levofloxacin dose in humans for post-exposure of inhalational anthrax is 500 mg q24h for 60 days. An oral dose of 500 mg levofloxacin in humans yields a Cmax of 5.1 µg/mL. **In the current study, mean** levofloxacin Cmax values following intragastric dosing of 50 mg/kg rabbits with and with out raxibacumab co-administration were slightly lower than the human Cmax with a 500 mg oral dose (mean Cmax range in rabbits was approximately 3.94 to 4.98 µg/mL). Thus, levofloxacin administered at the labeled oral dose of 500 mg in humans is expected to achieve slightly higher exposures than those observed in this study conducted in rabbits with inhalational anthrax.

APPEARS THIS WAY ON ORIGINAL

HGS1021-C1063:
A Randomized, Single-Blind, Placebo-Controlled Study to Evaluate the Safety and Tolerability of Raxibacumab (Human Monoclonal Antibody to *B. anthracis* Protective Antigen) in Healthy Subjects

Date(s): 12MAR2008 to 22JUL2008

Investigator and Clinical Site: multicenter (6 US sites)

OBJECTIVES:

Primary: To evaluate the safety and tolerability of IV administered raxibacumab in healthy subjects, and

Secondary: To determine serum raxibacumab concentrations for use in a population PK analysis.

STUDY DESIGN:

HGS1021-C1063 was a randomized, single-blind, placebo-controlled study of raxibacumab in healthy subjects. The study was designed to evaluate the safety and tolerability of raxibacumab administered IV in healthy subjects. Subjects were randomized to 1 of 2 raxibacumab groups (40 mg/kg double-dose or 40 mg/kg single dose) or to 1 of 2 matching placebo groups. Subjects were randomized to a treatment group at a ratio of 3:1 (raxibacumab:placebo). Subjects were first randomized to the double-dose cohorts. When randomization was completed for the double-dose cohorts, subjects were randomized into the single-dose cohorts. The planned treatment groups were as follows:

Table 1. Treatment Groups in Study HGS1021-C1063

Group	# of Subjects	Treatment	Dose	Frequency
1	24	Double-dose raxibacumab	40 mg/kg IV	Dose 1 - Day 0 Dose 2 - Day 14
3	8	Double-dose placebo	Placebo IV	Dose 1 - Day 0 Dose 2 - Day 14
2	216	Single-dose raxibacumab	40 mg/kg IV	Single-dose - Day 0
4	72	Single-dose placebo	Placebo IV	Single-dose - Day 0

Source: Study Report HGS1021-C1063, Section 5.1

Subjects were stratified at randomization by age (< age 65 or ≥ age 65). Enrollment was to target a population that was approximately 35% female, 8% Hispanic, and 12% non-white and 15% ≥ age 65 years (in the single dose cohorts). Approximately 320 subjects were to be enrolled.

Subjects who were randomized to receive a single dose of study agent were to complete visits through Day 56/End of Study visit. Subjects who were randomized to receive a double dose of study agent and received both infusions were to complete visits through Day 70/End of Study visit. Double-dose subjects who discontinued treatment prior to receiving the second infusion were to return for follow up visits according to the single-dose study (through Day 56/End of Study).

FORMULATIONS:
Raxibacumab was supplied in 50 mL sterile, single-use vials containing 34 mL of liquid formulation per vial. Each vial contained 50 mg/mL raxibacumab in 0.12 mg/mL citric acid, 2.8 mg/mL sodium citrate, 10 mg/mL sucrose, 18 mg/mL glycine, 0.2 mg/mL polysorbate 80, pH 6.5. Lots 71044 and 71051 (both process code/formulation M11/21-A) were administered to the subjects as a 40 mg/kg IV infusion.

Placebo was raxibacumab formulation buffer, and was also supplied in 50 mL sterile, single-use vials containing a minimum of 34 mL of liquid formulation per vial (Lot # 71043).

PHARMACOKINETIC ASSESSMENTS:
Blood samples for serum raxibacumab concentration measurement were collected from all subjects in the single-dose cohorts prior to administration of the raxibacumab and diphenhydramine doses on Day 0, at 30 minutes and 2 to 6 hours after completion of the raxibacumab infusion, and at 14, 28, and 56 days after the raxibacumab dose. For the double-dose cohorts, blood samples for serum raxibacumab concentration measurement were to be collected from all subjects prior to administration of the raxibacumab and diphenhydramine doses on Days 0 and 14, at 30 minutes and 2 to 6 hours after completion of each raxibacumab infusion, and at 28, 42, 56, and 70 days after the first raxibacumab dose.

IMMUNOGENICITY ASSESSMENTS:
The following blood samples were obtained for immunogenicity testing in HGS1021-C1063:

- Double-dose groups (Group 1 and Group 3): blood samples were collected prior to dosing on Day 0 and Day 14, and on Days 42 and 70/End of Study.
- Single-dose groups (Group 2 and Group 4): blood samples were collected prior to dosing on Day 0, and on Days 28 and 56/End of Study.

Subjects with a positive anti-raxibacumab antibody response after raxibacumab administration were to return 6 months after their last dose for a follow up assessment of immunogenicity.

BIOANALYTICAL ANALYSIS:
Serum samples were analyzed for raxibacumab using an electrochemiluminescence (ECL)-based assay. Biotinylated PA is bound to a streptavidin coated Meso Scale Discovery (MSD) 96-well assay plate for raxibacumab capture. The raxibacumab in diluted serum samples binds to the biotinylated PA and is detected by the addition of rabbit anti-raxibacumab followed by goat anti-human antibody labeled with MSD SULFOTAG™, an ECL label. MSD Read Buffer is added to the plate and the plate is inserted into an MSD plate reader, where voltage applied to the plate electrodes causes the MSD SULFO-TAG™ to emit light proportional to the amount of raxibacumab present in the serum. The concentration of raxibacumab in serum samples is interpolated from an 8-point reference standard curve. The lower limit of quantitation (LLOQ) is 800 ng/mL of raxibacumab in 100% serum.

Immunogenicity testing was performed to determine if raxibacumab induced an anti-raxibacumab immune response. Testing comprised 2 assays (screening and confirmatory). The screening assay (direct binding) is an electrochemiluminescence (ECL)-based bridging assay. A rabbit polyclonal antibody was used as a positive control. Samples above the assay cut point were considered positive. Samples identified as positive in the screening assay were to be confirmed positive in a confirmatory assay. Samples must have demonstrated a significant percent drop in the confirmatory inhibition of binding assay to be considered positive. The inhibition of binding confirmatory assay was performed identically to the direct binding screening assay with the

exception that the samples were tested in parallel with excess unlabeled raxibacumab. The assay limit of detection is 62.5 ng/mL.

PHARMACOKINETIC/PHARMACODYNAMIC/STATISTICAL ANALYSIS:
The primary purpose of this study was to evaluate the safety and tolerability of raxibacumab. The study was not prospectively designed to power statistical comparisons of raxibacumab PK between subgroups of subjects. Serum raxibacumab concentration data obtained from this study were pooled with data obtained from other studies for use in a population PK analysis (reported separately, study report HGS1021-POP01). No calculations of PK parameters were performed for this study report. Serum raxibacumab concentration results from this study were summarized in graphs and tables. Subjects were considered evaluable for raxibacumab PK if they received a raxibacumab dose and had at least 1 measurable post dose serum raxibacumab concentration.

Immunogenicity screening involved an initial screening assay, and if any positives were identified, these were analyzed by a confirmation assay. Immunogenicity data were summarized in tabular form if there were significant numbers of subjects with antibody formation. A listing was provided to show immunogenicity responses for all subjects by visit.

RESULTS:
Study Population
A total of 700 subjects were screened to provide 322 randomized subjects for the study. Of the 322 randomized subjects, 320 were treated (72 in the placebo single-dose group, 8 in the placebo double-dose group, 216 in the raxibacumab single-dose group, and 24 in the raxibacumab double-dose group). One subject (US002-004) randomized to the placebo single-dose group had a positive urine drug screen (UDS) and was not dosed per investigator decision. One subject (US005-014) in the raxibacumab single-dose group refused treatment following an unsuccessful initial attempt at venipuncture for IV dosing.

Demographics

A summary of demographic and baseline characteristics for the study population is presented in Table 2. Demographic characteristics were similar among treatment groups.

Table 2. Demographics and Baseline Characteristics – Study HGS1021-C1063

	Placebo - Single-Dose N = 74	Placebo - Double-Dose N = 6	All Placebo N = 80	Raxibacumab - Single-Dose N = 217	Raxibacumab - Double-Dose N = 23	All Raxibacumab N = 240
Sex						
Male	44 (59.5%)	2 (33.3%)	46 (57.5%)	100 (46.1%)	10 (43.5%)	110 (45.8%)
Female	30 (40.5%)	4 (66.7%)	34 (42.5%)	117 (53.9%)	13 (56.5%)	130 (54.2%)
Race[1]						
White	65 (87.8%)	4 (66.7%)	69 (86.3%)	169 (77.9%)	21 (91.3%)	190 (79.2%)
Asian	6 (8.1%)	-	6 (7.5%)	18 (8.3%)	-	18 (7.5%)
Black or African American	4 (5.4%)	2 (33.3%)	6 (7.5%)	23 (10.6%)	1 (4.3%)	24 (10.0%)
Native Hawaiian or Other Pacific Islander	-	-	-	5 (2.3%)	-	5 (2.1%)
Not Listed	1 (1.4%)	-	1 (1.3%)	11 (5.1%)	1 (4.3%)	12 (5.0%)
Multiracial	2 (2.7%)	-	2 (2.5%)	8 (3.7%)	-	8 (3.3%)
Hispanic or Latino origin	7 (9.5%)	-	7 (8.8%)	28 (12.9%)	1 (4.3%)	29 (12.1%)
Age (years)[2]						
n	74	6	80	217	23	240
Mean ± SD	39.8 ± 16.8	52.2 ± 13.8	40.7 ± 16.8	40.3 ± 16.3	48.5 ± 14.6	41.1 ± 16.3
Median	39.3	48.0	41.5	37.5	50.5	39.4
Range	(18.1, 77.9)	(38.7, 78.0)	(18.1, 78.0)	(18.1, 87.9)	(22.1, 76.2)	(18.1, 87.9)
Age group						
< 65 years	67 (90.5%)	5 (83.3%)	72 (90.0%)	198 (91.2%)	21 (91.3%)	219 (91.3%)
≥ 65 years	7 (9.5%)	1 (16.7%)	8 (10.0%)	19 (8.8%)	2 (8.7%)	21 (8.8%)

Source: Study Report HGS1021-C1063, Section 6.4.1

Raxibacumab Serum Pharmacokinetics

In total, 432 specimens were collected after placebo doses. Of note, in four of the 80 subjects (5%) administered placebo (Subjects US003-004, US003-020, US003-044, and US006-046, all in the single-dose group) each had a single measurable serum raxibacumab concentration, ranging from 12.192 to 339.312 µg/mL, all of which occurred at times post dose. These results were confirmed by retesting. These measurable concentrations were less than 37% of the mean 30 minute post dose serum raxibacumab concentration for the raxibacumab single-dose group. None of the placebo-dosed subjects had measurable raxibacumab concentrations at more than one collection time post dose, and only one of these subjects had a measurable concentration at the first collection time of 30 minutes post dose. Overall, the relatively low levels of raxibacumab detected in some placebo-dosed subjects, along with the lack of measurable concentrations at more than one timepoint for a subject, are not consistent with inadvertent administration of raxibacumab to any of the placebo-dosed subjects.

A total of 1258 specimens were collected after raxibacumab doses for the evaluable subjects. On average, nine post dose specimens were collected from a subject in the raxibacumab double-dose group, with a range of eight to nine specimens per subject, while on average, five post dose specimens were collected from a subject in the raxibacumab single-dose group, with a range of two to six specimens per subject. Of note, Subjects US003-003 and US003-042 had serum raxibacumab concentrations that were not measurable at the Day 14 collection time, while Subject US006-044 had a nonmeasurable serum raxibacumab concentration at the Day 56 collection time. These results were confirmed by retesting. These collection times coincide with those at which placebo-dosed Subjects US003-004, US003-044, and US006-046 had measurable serum raxibacumab concentrations. Thus, it appears likely that there was a mishandling of these specimens during processing. However, it is not possible to retrospectively and unequivocally determine which specimens were interchanged between which subjects.

Only one of the 238 evaluable subjects (0.4%) administered raxibacumab in this study (Subject US004-024) had a measurable serum raxibacumab concentration (1.152 µg/mL) prior to administration of the first raxibacumab dose. That concentration is less than 1% of the mean 30 minute post dose concentration, and less than 2% of the mean concentration measured at 56 days post dose, for the raxibacumab single-dose group.

For Subject US003-026, the serum raxibacumab concentration-time profile was inconsistent with IV administration, in that the peak serum raxibacumab concentration did not occur until 14 days post dose. The atypical profile was confirmed by retesting. That subject's profile appeared to be more consistent with extravascular administration. However, the dosing information reported for that subject indicated that a dose volume of 279 mL was administered over 2.53 hours, a volume and duration that is incompatible with subcutaneous or intramuscular administration. It is possible that the specimens for that subject were misidentified, but no documentation of such an error exists.

Data Excluded From Analysis

In the raxibacumab single-dose group, two subjects (US003-016, US003-017) were to have been administered raxibacumab, but no measurable raxibacumab concentrations were obtained for any post dose collection time for those subjects. Since no other raxibacumab-dosed subject in this study had a complete absence of measurable serum raxibacumab concentrations, it was judged that the two subjects in question had inadvertently not been administered raxibacumab and were hence considered as nonevaluable for raxibacumab PK. A review of study documentation did not reveal any evidence that would suggest these subjects had inadvertently been administered placebo in lieu of raxibacumab. However, given that high serum raxibacumab levels were

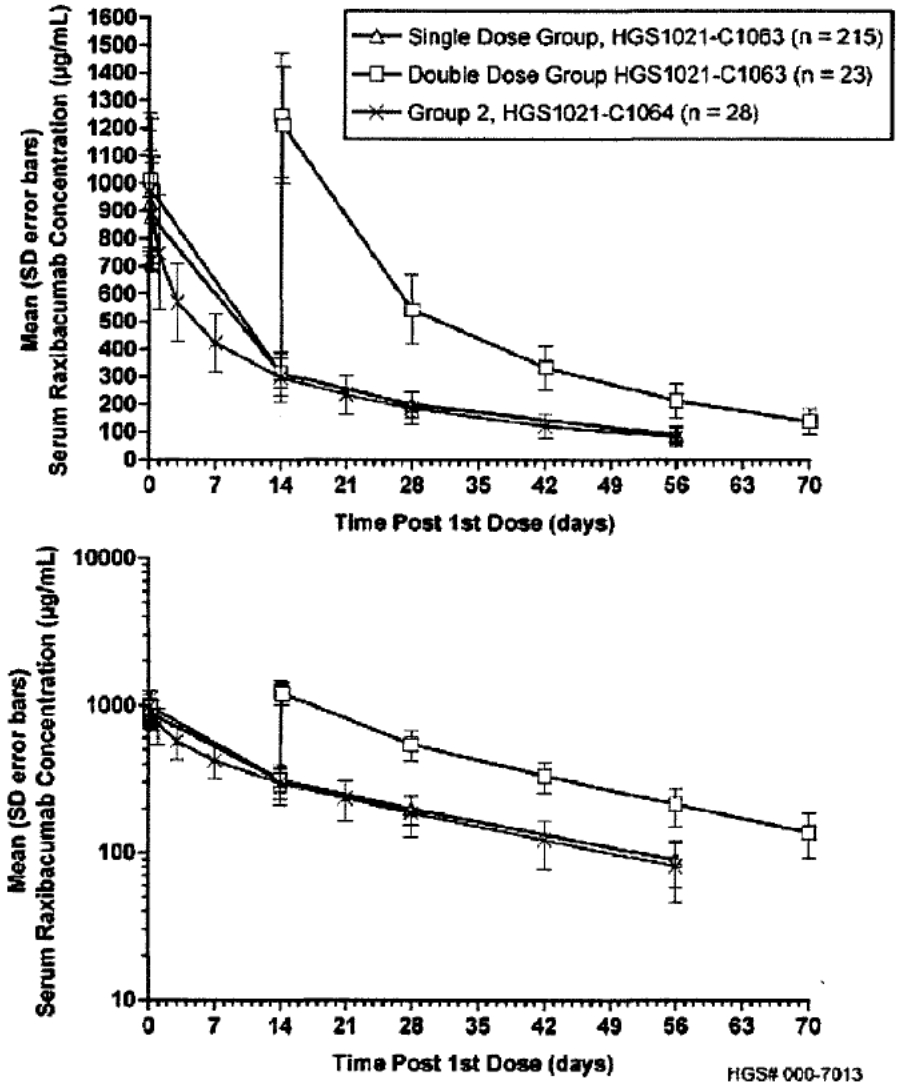
attained in other subjects in that dose group, the complete absence of measurable serum raxibacumab concentrations implies that it is very unlikely that these two nonevaluable subjects were actually administered a raxibacumab dose. All other raxibacumab-dosed subjects were considered evaluable (total number of evaluable subjects, 238; double-dose raxibacumab: 23 subjects, single-dose raxibacumab: 215 subjects).

Raxibacumab Serum Concentrations and PK Parameters

Serum concentration-time profiles for raxibacumab following single (single-dose group) and multiple (double-dose group) intravenous administration of 40 mg/kg in healthy, male and female subjects are presented in Figure 1. For purposes of comparison, the mean serum raxibacumab concentration-time profile following a single 40 mg/kg IV infusion raxibacumab dose for Group 2 (raxibacumab 40 mg/kg alone; n = 28) in a previous study (Clinical Study Report HGS1021-C1064) is also shown in Figure 1. The two concentration-time profiles are in agreement, with overlapping SD error bars, for the single dose in this study and the single dose in Study HGS1021-C1064. Similarly, during the first 14 days for the double-dose group (prior to the second dose), the SD error bars overlap with those for the single-dose groups in this study and the prior study. Overall, these observations suggest that raxibacumab PK in this study population were similar to those for the prior study conducted with the M11 formulation. Since the second dose was administered to the double-dose group while serum raxibacumab concentrations were still measurable, there was accumulation, as could be expected.

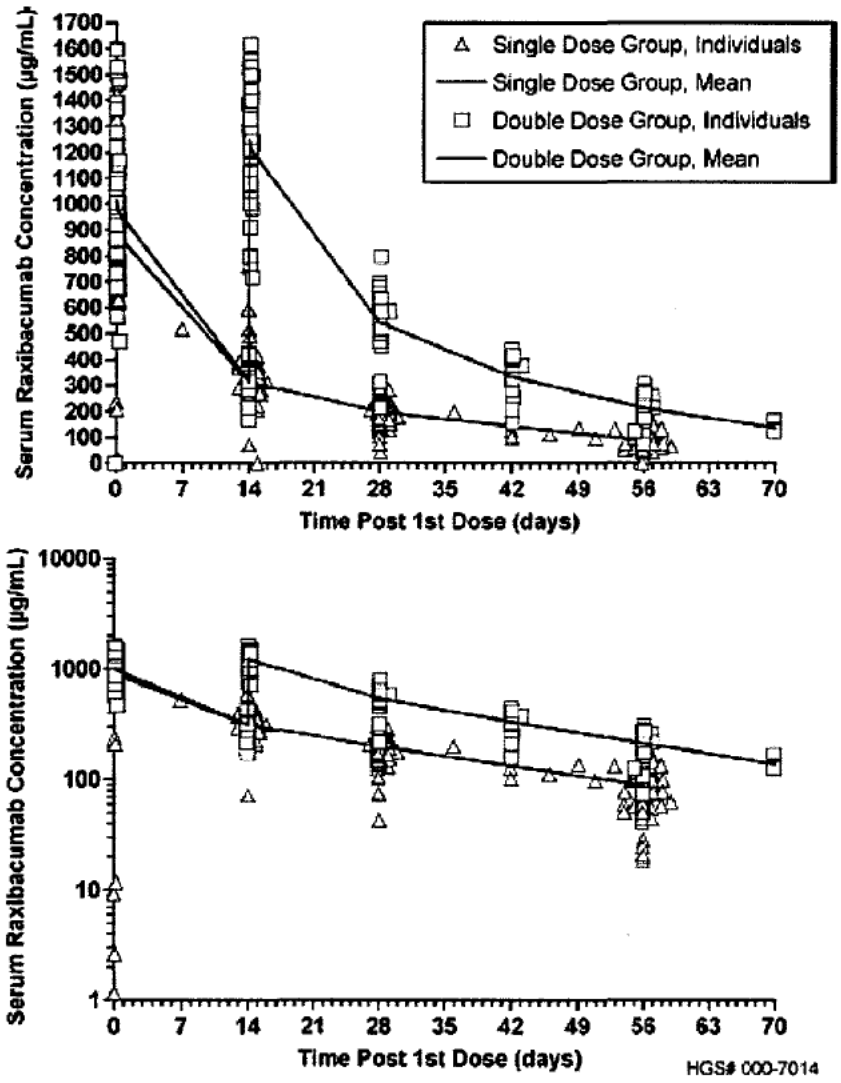
Individual serum raxibacumab concentration-time results for the raxibacumab single-dose and double-dose groups are illustrated in Figure 2, with the mean concentration-time profile for each group for comparison. Serum raxibacumab levels among individuals displayed low variability and consistency between the single- and double-dose groups up to the time of the second dose.

Figure 1. Mean (± SD) serum concentration-time profiles for raxibacumab following single (single-dose group) and multiple (double-dose group) intravenous administration of 40 mg/kg in healthy, male and female subjects (linear and log scale)



Source: Study Report HGS1021-C1063, Section 10.4.4

Figure 2. Individual serum concentrations and mean (± SD) serum concentration-time profiles for raxibacumab following single (single-dose group) and multiple (double-dose group) intravenous administration of 40 mg/kg in healthy, male and female subjects (linear and log scale)



Source: Study Report HGS1021-C1063, Section 10.4.4

Immunogenicity
There were no subjects who developed an anti-raxibacumab antibody response during the study.

APPLICANT'S DISCUSSION:
The objective of this study was to determine serum raxibacumab concentrations for use in a population PK analysis, to be reported separately. In this study, subjects were administered either a single IV infusion dose of placebo or 40 mg/kg raxibacumab, or 2 IV infusions doses given 14 days apart of placebo or 40 mg/kg raxibacumab. No immunogenicity was detected in these subjects following raxibacumab administration.

To approximate a cross-section of the United States population, enrollment in the study targeted approximately 15% of the subjects in the single dose cohorts ≥ 65 years of age, and also targeted a population of approximately 35% female, 8% Hispanic, and 12% non-white. The actual enrollment included 9% of subjects ≥ 65 years of age (range: 18 to 88 years), 54% female, 12% Hispanic, and 21% non-white. Overall, the study population was sufficiently diverse to support evaluation of subject demographics as potential covariates in the population PK analyses.

In total 1258 specimens were collected after raxibacumab doses for the evaluable subjects. On average, nine postdose specimens were collected from a subject in the raxibacumab double-dose group, while on average, five postdose specimens were collected from a subject in the raxibacumab single-dose group. Essentially all raxibacumab-dosed subjects had measurable serum raxibacumab concentrations for up to 56 days post dose. The mean serum raxibacumab concentration-time profiles for this study are consistent with those for subjects administered a single 40 mg/kg IV infusion raxibacumab dose in a prior study.

APPLICANT'S CONCLUSIONS:
- The population of subjects enrolled in this study is representative of the United States population, and is suitable for the planned population PK analyses.
- The serum raxibacumab concentrations observed for this study are in agreement with the serum raxibacumab concentration-time profile in a previous study, and should be adequate to support the planned population PK analyses.
- In essentially all raxibacumab-dosed subjects, serum raxibacumab concentrations remained measurable for up to 56 days post dose.

REVIEWER ASSESSMENT:
Results from Study HGS1021-C1063 adequately achieved the secondary objective of determining serum raxibacumab concentrations for use in a population PK analysis. The applicant's pharmacokinetic conclusions based on these findings are valid. In addition, an anti-raxibacumab immune response did not develop following two doses of raxibacumab 40 mg/kg IV dosed 14 days apart.

For further evalauation of the safety and tolerability results for Study HGS1021-C1063, refer to the Medical Officers' review for BLA 125349.

HGS1021-C1064:
An Open-Label Study to Evaluate the Pharmacokinetics and Safety of Raxibacumab (Human Monoclonal Antibody to *B. anthracis* Protective Antigen) Administered in Combination with Ciprofloxacin in Healthy Subjects

Date(s): 26JAN2007 to 28AUG2007

Investigator and Clinical Site: multicenter

OBJECTIVES:
- To determine the effect of co-administration of raxibacumab on PO ciprofloxacin pharmacokinetics (PK).
- To evaluate the safety of raxibacumab alone and in combination with PO or IV administered ciprofloxacin.

STUDY DESIGN:
HGS1021-C1064 was an open-label study to evaluate the effect of raxibacumab on ciprofloxacin PK as well as the safety and PK of raxibacumab in combination with ciprofloxacin in healthy adult male and female subjects. Three treatment groups were evaluated. Group 1 received PO ciprofloxacin (500 mg Q12h, Days 0 to 7), with a single raxibacumab (40 mg/kg) dose IV on Day 5. Group 2 received a single raxibacumab (40 mg/kg) dose IV on Day 0. Group 3 received a single IV ciprofloxacin (400 mg) dose on Day 0 immediately followed by a single IV raxibacumab (40 mg/kg) dose, a 2nd IV ciprofloxacin (400 mg) dose 12 hours later, and then PO ciprofloxacin (500 mg Q12h, Days 1 to 7) for a total of 13 doses.Subjects were randomized in a 1:1 ratio to Group 1 or Group 2. Enrollment in Groups 1 and 2 (28 evaluable subjects per group) was completed, and then an additional 28 evaluable subjects were enrolled into Group 3. A minimum of 3 evaluable female subjects were to be enrolled into each group.

FORMULATIONS:
Raxibacumab was supplied in 50 mL sterile, single-use vials containing 35.1 mL of liquid formulation per vial. Each vial contained 50 mg/mL raxibacumab in 0.13 mg/mL citric acid, 2.8 mg/mL sodium citrate, 10 mg/mL sucrose, 18 mg/mL glycine, 0.2 mg/mL polysorbate 80, pH 6.5. Lot 71044 was administered to the subjects as a 40 mg/kg IV infusion.

Ciprofloxacin was supplied as tablets (500 mg, (b) (4) and as a 2 mg/mL stock solution in 5% dextrose (b) (4). IV ciprofloxacin was to be administered over 60 minutes at a constant rate. Lot 2500LXV was used for IV ciprofloxacin. Lots C07187 and 157547A were used for PO ciprofloxacin.

PHARMACOKINETIC ASSESSMENTS:
Blood samples for plasma ciprofloxacin concentration measurement were collected from Group 1 subjects at the following times: Day 0 (prior to dose #1); Day 2 (prior to dose #5, at 1 hour post dose #5, prior to dose #6, and at 1 hour post dose #6); Day 3 (prior to dose #7, at 1 hour post dose #7, prior to dose #8, and at 1 hour post dose #8); Day 4 (prior to dose #9, at 0.5, 1, 1.5, 2, 4, 6, 8, and 12 hours post dose #9, and at 1 hour post dose #10); Day 5 (prior to raxibacumab infusion, at 0.5, 1, 1.5, 2, 4, 6, 8, and 12 hours post dose #11, and at 1 hour post dose #12); Day 6 (prior to dose #13, at 1 hour post dose #13, prior to dose #14, and at 1 hour post dose #14); and, Day 7 (prior to dose #15, at 1 hour post dose #15, and 12 hours post dose 15).

Blood samples for plasma ciprofloxacin concentration measurement were collected from Group 3 subjects at the following times: Day 0 (prior to IV dose #1, at 0.5, 1, 1.5, 2, 4, 6, 8, and 12 hours post-dose #1 just before IV dose #2); Day 5 (prior to dose #11 [9th PO dose], at 1 hour post-dose #11, prior to dose #12 [10th PO dose], and at 1 hour post-dose #12); Day 6 (prior to dose #13 [11th PO dose], 1 hour post-dose #13, prior to dose #14 [12th PO dose], and at 1 hour post-dose #14); and, Day 7 (prior to dose #15 [13th PO dose], at 0.5, 1, 1.5, 2, 4, 6, 8, and 12 hours post-dose #15).

Blood samples for serum raxibacumab concentration measurement were collected from all subjects just prior to administration of the raxibacumab dose, at 5 minutes and 8 hours after completion of the raxibacumab infusion, and at 1, 3, 7, 14, 21, 28, 42, and 56 days after the raxibacumab dose.

IMMUNOGENICITY ASSESSMENTS:
Blood samples for assessment of anti-raxibacumab antibodies were collected at the following times:

- Group 1: Blood samples were collected on Days 0 (prior to raxibacumab dose), 33, and 61/End of Study.
- Group 2 and Group 3: Blood samples were collected on Days 0 (prior to prior to raxibacumab dose), 28, and 56/End of Study.

Subjects found to have a positive anti-raxibacumab antibody titer following raxibacumab administration were to have serum collected approximately 6 months after the dose for follow-up immunogenicity assessment.

BIOANALYTICAL ANALYSIS:
Serum samples were analyzed for raxibacumab using an electrochemiluminescence (ECL)-based assay. Biotinylated PA is bound to a streptavidin coated Meso Scale Discovery (MSD) 96-well assay plate for raxibacumab capture. The raxibacumab in diluted serum samples binds to the biotinylated PA and is detected by the addition of rabbit anti-raxibacumab followed by goat anti-human antibody labeled with MSD SULFOTAG™, an ECL label. MSD Read Buffer is added to the plate and the plate is inserted into an MSD plate reader, where voltage applied to the plate electrodes causes the MSD SULFO-TAG™ to emit light proportional to the amount of raxibacumab present in the serum. The concentration of raxibacumab in serum samples is interpolated from an 8-point reference standard curve. The lower limit of quantitation (LLOQ) is 800 ng/mL of raxibacumab in 100% serum.

The LOD of the standard curve is considered to be less than 0.0046 ng/mL. Repeatability precision is expressed as the ratio of the square root of the sum of error variance and between-plate variance to the mean. Repeatability precision ranged from 4-9%. Intermediate precision is expressed as the ratio of the square root of the sum of operator, day and operator*day variance components to the mean. Intermediate precision ranged from 3-17%. The overall CV of spiked samples ranged from 5.9% to 27.6%. The calculated accuracies of the spiked samples ranged from 83.9 to 96.0%.

Plasma samples were analyzed for ciprofloxacin using a high performance liquid chromatography (HPLC)/mass spectrometry method. The analyte ciprofloxacin and its internal standard enrofloxacin were extracted from a 100 µL aliquot of human EDTA plasma by solid phase extraction procedure. The extracted samples were injected onto a Zorbax SB-C8, 4.6 x 75 mm, 3.5 µm HPLC column equipped with a triple quadrupole mass spectrometer detector. The mobile phase used was an isocratic mixture of 50% acetonitrile, 50% 10 mM ammonium acetate +

formic acid (pH 2.7). The assay was validated over a calibration range of 10 ng/mL to 5000 ng/mL in human plasma.

PHARMACOKINETIC/PHARMACODYNAMIC/STATISTICAL ANALYSIS:
PK parameters were to be calculated from the plasma ciprofloxacin concentration-time results and the serum raxibacumab concentration-time results, using noncompartmental techniques. Plasma or serum drug concentration data and drug PK are summarized via descriptive statistics. Equivalence of ciprofloxacin PK parameters for ciprofloxacin alone and ciprofloxacin plus raxibacumab (Group 1) was based on the 90% CI. Raxibacumab PK parameters for Groups 1 and 3 (with ciprofloxacin) were compared to those for Group 2 (raxibacumab alone) using a 2-sample t-test; the exception was tmax, which was compared using the Mann-Whitney test. Although not prospectively planned, based on review of the study results it was desired to assess potential differences in raxibacumab exposure among subgroups of subjects based on occurrence of rash and diphenhydramine use. For the purpose of this assessment, one-way analysis of variance (ANOVA) was used. The raxibacumab PK results were inspected for possible effects of age, gender, race, physical characteristics (such as body surface area), and study site. Their impact on raxibacumab PK was assessed by visual inspection of graphical displays.

RESULTS:
Study Population
A total of 375 subjects were screened to provide 90 randomized subjects for the study. The number of screening failures was primarily due to individuals not meeting entry (inclusion/ exclusion) criteria. A total of 88 subjects were randomized and treated (32 in Group 1, 28 in Group 2, and 28 in Group 3); 70 (79.5%) subjects completed the study and 18 (20.5%) subjects withdrew from the study. None of these 18 subjects withdrew due to an adverse event (AE). Two subjects in Group 1 (US003-010, US003-036) received ciprofloxacin and withdrew due to subject request prior to receiving raxibacumab. Three subjects (2 in Group 1 and 1 in Group 2) who received partial doses of raxibacumab due to AEs completed the study. All 13 subjects who were lost to follow-up were followed at least 2 weeks post raxibacumab dosing for safety monitoring. None of these subjects had AEs resulting in discontinuation of study agent and there was one ongoing AE of tooth abscess.

Demographics
A summary of demographic and baseline characteristics for the study population is presented in Table 1. There were no substantial differences in demographic characteristics between the study groups; the only statistically significant difference among groups in any of the demographic or baseline variables evaluated was premedication with diphenhydramine. The reason for this difference is that a requirement to premedicate with diphenhydramine was added to the protocol after several subjects experienced infusion-related rashes.

Table 1. Demographics and Baseline Characteristics – Study HGS1021-C1064

		Group:	1	2	3
		N:	32	28	28
Sex		Male	16 (50.0%)	13 (46.4%)	14 (50.0%)
		Female	16 (50.0%)	15 (53.6%)	14 (50.0%)
Race		Black or African American	13 (40.6%)	8 (28.6%)	10 (35.7%)
		Black or African American & Native Hawaiian/Other Pacific Islander	-	1 (3.6%)	-
		Not Listed	-	2 (7.1%)	1 (3.6%)
		White	19 (59.4%)	17 (60.7%)	17 (60.7%)
Age (years)		Mean ± SD	35 ± 11	36 ± 11	35 ± 13
		Median	34	35	32
		Range	(18, 58)	(21, 57)	(18, 60)
Height (cm)		Mean ± SD	169.4 ± 10.8	169.9 ± 10.4	168.8 ± 10.6
		Median	172.1	167.5	169.2
		Range	(145.9, 187.0)	(152.2, 198.0)	(153.6, 193.0)
Weight (kg)		Mean ± SD	74.0 ± 14.3	74.0 ± 11.2	68.8 ± 13.2
		Median	71.8	75.6	66.0
		Range	(52.6, 97.6)	(53.8, 98.0)	(49.0, 98.6)

Source: HGS1021-C1064 Pharmacokinetic Report, Section 3.2

Table 2 provides a summary of the subjects treated and considered evaluable for PK in each treatment group.

Table 2. Summary of Pharmacokinetically Evaluable Subjects – Study HGS1021-C1064

	Number of Subjects:					
	Treated		Evaluable[1]		Non-Evaluable[1]	
Treatment Group	Ciprofloxacin	Raxi	Ciprofloxacin	Raxi	Ciprofloxacin	Raxi
Group 1	32	30	30	30	2	0
Group 2	NA	28	NA	28	NA	0
Group 3	28	28	28	28	0	0
Total[2]	60	86	58	86	2	0

Abbreviation: NA, not applicable.

1 Classification as evaluable or non-evaluable for the purpose of PK analysis. In Group 1, subjects were considered evaluable for ciprofloxacin PK if they received the first 11 of the planned ciprofloxacin doses as well as the raxibacumab dose, and had predose through at least 8 hours postdose plasma ciprofloxacin concentrations measured for the 9th and 11th doses (with and without raxibacumab, respectively). Two subjects in Group 1 did not receive the raxibacumab dose and were considered non-evaluable. For Group 3, subjects were considered evaluable for ciprofloxacin PK if they received the 1st IV ciprofloxacin dose as well as the raxibacumab dose and had predose through at least 8 hours postdose plasma ciprofloxacin concentrations measured for the 1st ciprofloxacin dose. Subjects were considered evaluable for raxibacumab PK if they received a raxibacumab dose and had at least one measurable postdose serum raxibacumab concentration. A non-evaluable subject was one who did not meet these criteria.

2 Total for all dose groups combined.

Source: HGS1021-C1064 Pharmacokinetic Report, Section 3.1

Raxibacumab Serum Pharmacokinetics

Data Excluded From Analysis

For Subject US003-000029 (Group 1), the serum raxibacumab concentration-time profile was inconsistent with IV administration, in that the peak serum raxibacumab concentration did not occur until 3 days postdose (the subject's profile appeared to be more consistent with extravascular administration). However, the dosing information reported for that subject indicates that a dose volume of 250 mL was administered over 2.38 hours, a volume and duration that is incompatible with subcutaneous or intramuscular administration. PK analysis of that subject's profile was not done and that subject's serum raxibacumab concentration data were excluded from calculation of descriptive statistics for the concentration-time data for Group 1.

Three of the 86 subjects administered raxibacumab in this study (Subjects US003-000002, US003-000006, and US003-000028) did not receive the complete raxibacumab dose, since their infusions were stopped due to mild adverse events. For these subjects, PK parameters were calculated based on the actual dose administered. Therefore, Cmax and AUC0-∞ were dose normalized for any comparisons among dose groups or subgroups of subjects.

Raxibacumab Serum Concentrations and PK Parameters

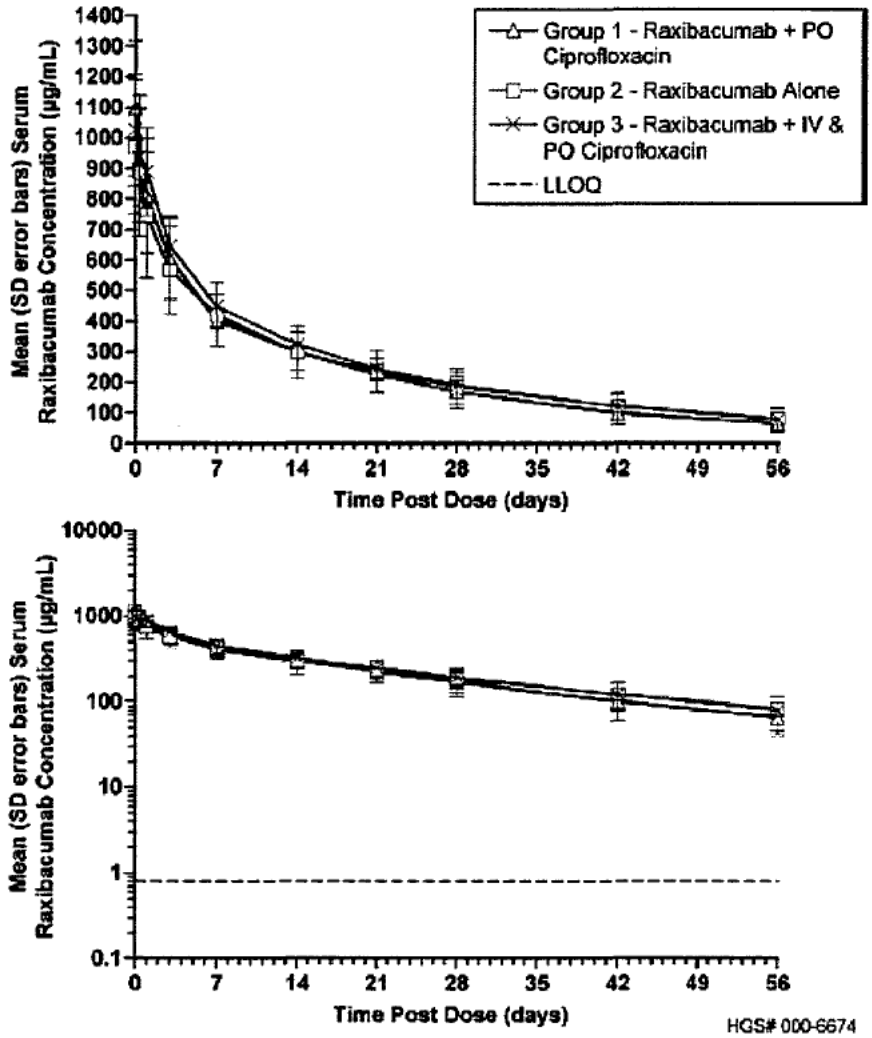
Serum concentration-time profiles for raxibacumab following single intravenous administration of 40 mg/kg with and without coadministration of ciprofloxacin PO (Group 1) or IV/PO (Group 3) in healthy, male and female subjects are presented in Figure 1. Pharmacokinetic parameters and results of statistical comparisons for raxibacumab following single intravenous administration of 40 mg/kg with and without coadministration of ciprofloxacin PO (Group 1) or IV/PO (Group 3) in healthy, male and female subjects are summarized in Table 3.

The only significant differences in raxibacumab PK between the groups studied were that the median tmax for Group 1 was significantly lower (-5%, p = 0.0108) than that for Group 2, and that the mean Cmax/Dose for Group 1 was significantly higher (14%, p = 0.0015) than that for Group 2. These differences are small, and were not replicated for Group 3, which also received ciprofloxacin. Hence, it is unlikely that these differences are meaningful. There were no other statistically significant differences in PK parameters between Group 3 and Group 2 or between Group 1 and Group 2. Overall, raxibacumab PK were not affected by co-administration of IV or PO ciprofloxacin.

The mean Vss ranged from 65 to 72 mL/kg among the dose groups. The smallest mean Vss is about 52% greater than the plasma volume (~42.8 mL/kg), while the largest mean Vss is about 68% greater than the plasma volume, suggesting that raxibacumab does distribute to tissues. The disappearance of raxibacumab from serum appears to be biphasic. The mean terminal phase elimination half-lives (t1/2,z) range from 20 to 22 days. Mean clearance (CL) ranged from 2.6 to 3.0 mL/day/kg among the dose groups. CL values were much smaller than the glomerular filtration rate indicating that, as expected, there is virtually no renal clearance of this monoclonal antibody. Based on graphical assessment of covariates, none of the potential covariates assessed had any meaningful impact on raxibacumab PK.

Several subjects in Group 1 (Subjects No. US001-000017, US001-000020, US002-000001, and US002-000005), in Group 2 (Subjects No. US003-000008, US003-000011, US003-000014, and US003-0000022), and in Group 3 (Subjects No. US003-000048, US003-000050, and US003-0000058) had specimens collected for serum raxibacumab measurement for periods of 21 days or less after their raxibacumab dose. For those subjects, estimates of t1/2,z ranged from 4.27 to 21.07 days, whereas t1/2,z for all other subjects ranged from 12.61 to 45.80 days. Given the short duration of specimen collection for the indicated subjects, it is possible that t1/2,z may be underestimated for those subjects. Hence, the results for those subjects should be interpreted with caution.

Figure 1. Mean (± SD) Serum Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 40 mg/kg with and without Ciprofloxacin PO or IV in Healthy, Male and Female Subjects (Linear and Log Scale)



Source: HGS1021-C1064 Pharmacokinetic Report, Section 3.5.2.4

Table 3. Summary of Raxibacumab Pharmacokinetic Parameters Following Single Intravenous Administration of 40 mg/kg with and without Ciprofloxacin PO or IV in Healthy, Male and Female Subjects

Parameter	Group 1 With PO Ciprofloxacin (n = 29)	Group 2 Raxibacumab Alone (n = 28)	Group 3 With IV & PO Ciprofloxacin (n = 28)
C_{max} (µg/mL)	1103 ± 225	988 ± 220	1048 ± 180
C_{max}/Dose (kg/mL)	0.0287 ± 0.0042 (p = 0.0015[1])	0.0251 ± 0.0040	0.0262 ± 0.0045 (p = 0.3310[1])
t_{max} (day)[2]	0.098 (0.049 to 0.430) (p = 0.0108[1])	0.103 (0.038 to 0.439)	0.107 (0.099 to 0.914) (p = 0.1120[1])
$AUC_{0-\infty}$ (µg·day/mL)	14362 ± 4208	15328 ± 5059	16349 ± 4256
$AUC_{0-\infty}$/Dose (kg·day/mL)	0.3733 ± 0.0931 (p = 0.6157[1])	0.3873 ± 0.1145	0.4085 ± 0.1064 (p = 0.4743[1])
$t_{1/2,z}$ (day)	19.66 ± 7.89 (p = 0.6886[1])	20.44 ± 6.46	21.50 ± 8.92 (p = 0.6125[1])
MRT (day)	24.68 ± 7.87 (p = 0.2246[1])	27.30 ± 8.24	27.79 ± 9.69 (p = 0.8406[1])
CL (mL/day/kg)	2.98 ± 1.33 (p = 0.6865[1])	2.85 ± 1.03	2.63 ± 0.82 (p = 0.3904[1])
V_{ss} (mL/kg)	64.85 ± 11.60 (p = 0.0825[1])	71.74 ± 17.36	67.17 ± 12.61 (p = 0.2646[1])
V_z (mL/kg)	74.33 ± 23.43 (p = 0.6150[1])	77.29 ± 20.50	74.27 ± 18.94 (p = 0.5696[1])

Abbreviations: C_{max}, maximum serum raxibacumab concentration for a single dose; t_{max}, time of occurrence for C_{max}; $AUC_{0-\infty}$, area under the serum raxibacumab concentration-time curve from time 0 to infinite time for a single dose; $t_{1/2,z}$, elimination half-life for the terminal phase; MRT, mean residence time; CL, clearance; V_{ss}, volume of distribution at steady-state; V_z, volume of distribution in the terminal phase.

[1] P-value from a 2-sample t-test with Group 2 (raxibacumab alone) as the reference treatment; the exception is t_{max}, for which the p-value is from a Mann-Whitney test..

[2] Median and range are presented.

Source: HGS1021-C1064 Pharmacokinetic Report, Section 3.5.2.4

Ciprofloxacin Plasma Pharmacokinetics

Data Excluded From Analysis
No data was excluded from the analysis.

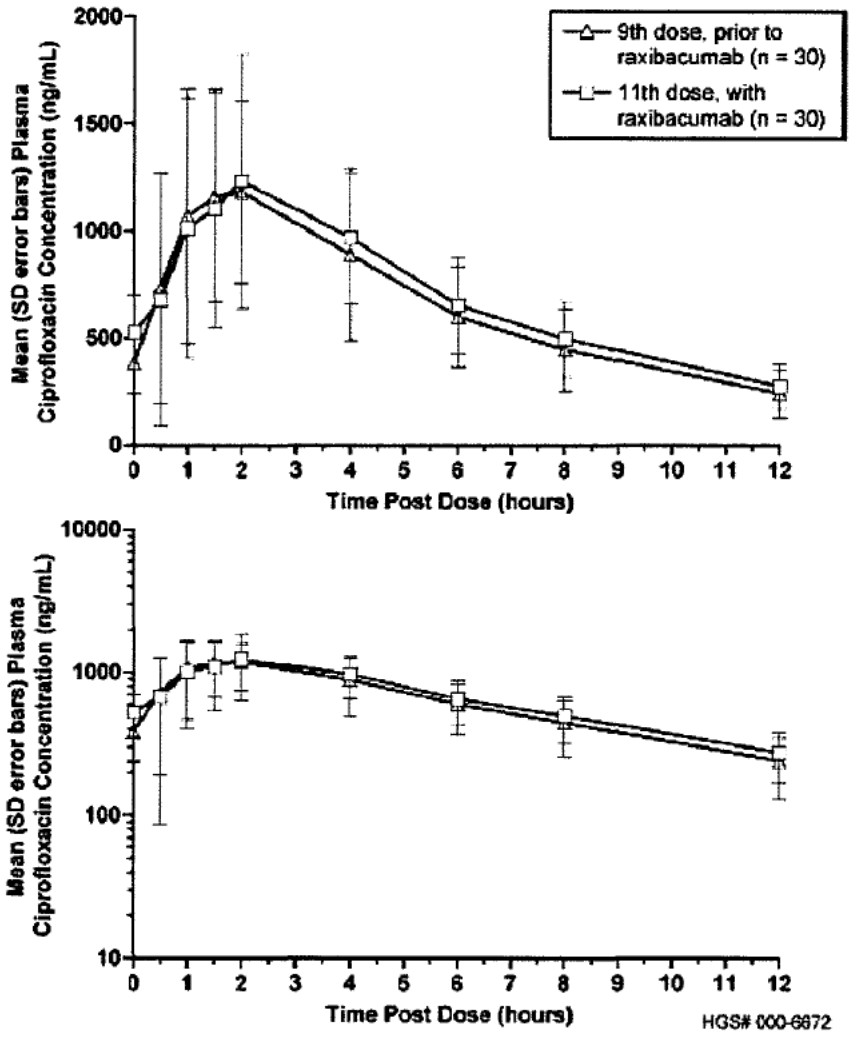
Ciprofloxacin Plasma Concentrations and PK Parameters
Plasma concentration-time profiles for ciprofloxacin following multiple oral administration with and without coadministration of a raxibacumab 40 mg/kg single dose in healthy, male and female subjects in Group 1 are presented in Figure 2. Plasma concentration-time profiles for ciprofloxacin following IV and oral administration with and without coadministration of a raxibacumab 40 mg/kg single dose in healthy, male and female subjects in Group 3 are presented in Figure 3. Peak and trough concentrations of ciprofloxacin in Groups 1 and 3 are presented in Table 4. Pharmacokinetic parameters ciprofloxacin following oral administration and IV followed by oral administration with and without coadministration of a raxibacumab 40 mg/kg single dose in healthy, male and female subjects are summarized in Table 5.

The plasma ciprofloxacin concentration-time profiles for Dose 9 (prior to raxibacumab administration) and Dose 11 (with raxibacumab) were quite similar, with overlapping SD bars, suggesting that raxibacumab did not affect ciprofloxacin PK. There was some accumulation of plasma ciprofloxacin concentrations in either dose group, with Cmin,n ranging from 193 to 527 ng/mL. While comparisons of 95% CI for Cmax,n and Cmin,n across days within a dose group shows some differences, there is no consistent pattern of increasing or decreasing values across days, suggesting that steady-state was attained prior to the 4th and 10th ciprofloxacin doses in Group 1 and Group 3, respectively. In addition, examination of the 95% CI for Cmax,n and Cmin,n across days within Group 1 shows no differences that could be attributed to altered ciprofloxacin PK after raxibacumab administration. Within a day, Cmin,n were higher for the PM dose (even numbered doses, ie, Doses 6, 8 ,10, etc) than for the AM doses (odd numbered doses, ie, Doses 5, 7, 9, etc). In contrast, there were no differences in Cmax,n for the AM and PM doses.

For Subject No. US003-000023 (Group 1), no specimen was collected at 12 hours post the 11th dose. Hence, AUCτ could not be determined for that subject. For the purpose of comparing AUC between the 9th and 11th doses for that subject, the area under the plasma ciprofloxacin concentration-time curve from 0 to 8 hours post dose was used. For Subject No. US003-000025 (Group 1) and Subject No. US003-000034 (Group 3), the plasma ciprofloxacin concentration-time profile did not allow estimation of λz; hence t1/2,z and Vz/F could not be determined for those subjects.

Based on comparison of AUC0-∞ and AUCτ for IV and PO, respectively, dosed ciprofloxacin in Group 3, the fraction of dose absorbed (F) for PO dosing is estimated to be about 59%. The PK for PO ciprofloxacin co-administered with raxibacumab were similar between Group 1 and Group 3, with median tmax between 1 and 2 hours and mean t1/2,z ranging from 4.6 to 5.3 hours. For Group 1, mean PK parameters were in good agreement for Dose 9 (prior to raxibacumab administration) and Dose 11 (with raxibacumab). The mean ciprofloxacin Css,max following raxibacumab administration was 1% lower than that for ciprofloxacin administered alone, while mean AUCτ for Dose 11 (with raxibacumab) was 6% higher than for Dose 9 (prior to raxibacumab). These results suggest that ciprofloxacin PK were not affected by co-administration of raxibacumab.

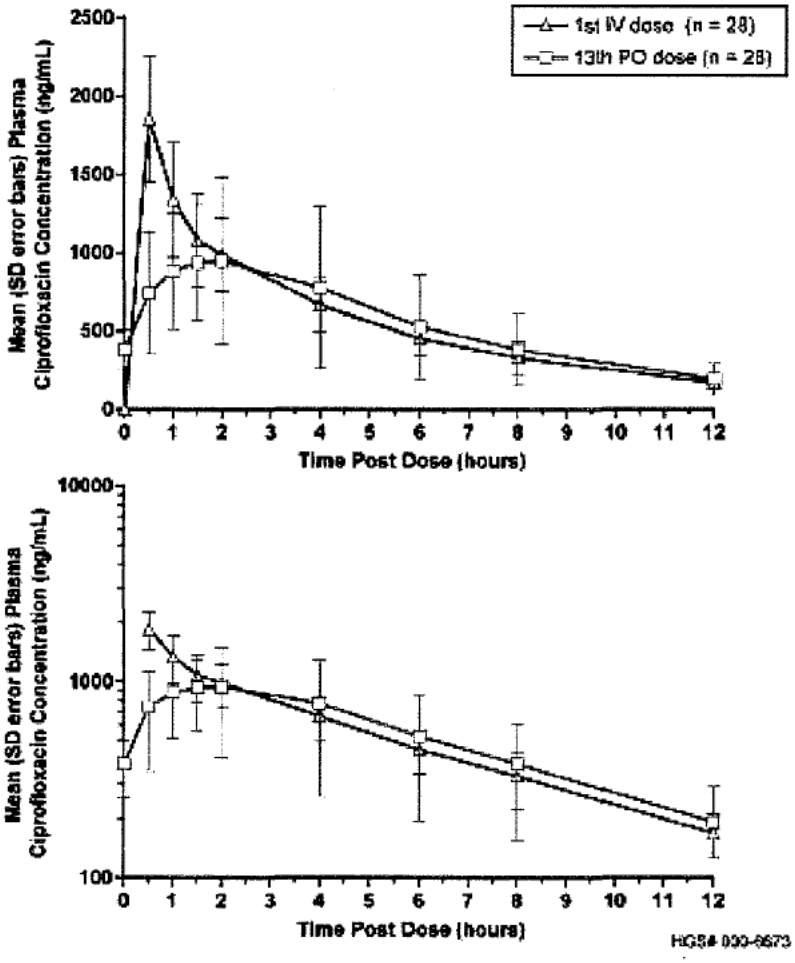
Figure 2. Mean (± SD) Plasma Concentration-Time Profiles for Ciprofloxacin Following Oral Administration with and without a Single Intravenous Dose of Raxibacumab 40 mg/kg in Healthy, Male and Female Subjects (Group 1; Linear and Log Scale)



Graph depicts plasma ciprofloxacin concentrations following the 9th and 11th of 15 consecutive 500 mg PO ciprofloxacin doses given q12h; a single 40 mg/kg raxibacumab IV infusion dose was administered just prior to the 11th ciprofloxacin dose.

Source: HGS1021-C1064 Pharmacokinetic Report, Section 3.5.1.4

Figure 3. Mean (± SD) Plasma Concentration-Time Profiles for Ciprofloxacin Following Intravenous and Oral Administration with and without a Single Intravenous Dose of Raxibacumab 40 mg/kg in Healthy, Male and Female Subjects (Group 3; Linear and Log Scale)



Graph depicts plasma ciprofloxacin concentrations following the 1st of two 400 mg IV ciprofloxacin doses given 12 hours apart, and following the 13th consecutive 500 mg PO ciprofloxacin doses given q12h starting 12 hours after the 2nd IV dose; a single 40 mg/kg raxibacumab IV infusion dose was administered immediately after the 1st IV ciprofloxacin dose.

Source: HGS1021-C1064 Pharmacokinetic Report, Section 3.5.1.4

Table 4. Summary of Ciprofloxacin Peak and Trough Plasma Concentrations Following Oral Administration (Group 1) or Intravenous Followed by Oral Administration (Group 3) with and without a Single Intravenous Dose of Raxibacumab 40 mg/kg in Healthy, Male and Female Subjects

		Group 1			Group 3[1]		
		N	Mean ± SD	95% CI	N	Mean ± SD	95% CI
Dose 4	$C_{min,3}$ (ng/mL)	30	367 ± 139	(316,419)		NA	
Dose 5	$C_{max,n}$ (ng/mL)	30	1048 ± 485	(867,1229)		NA	
	$C_{min,3}$ (ng/mL)	30	240 ± 93	(205,275)		NA	
Dose 6	$C_{max,n}$ (ng/mL)	30	1050 ± 741	(774,1327)		NA	
	$C_{min,3}$ (ng/mL)	30	423 ± 146	(368,477)		NA	
Dose 7	$C_{max,n}$ (ng/mL)	30	923 ± 421	(766,1081)		NA	
	$C_{min,3}$ (ng/mL)	30	228 ± 85	(196,260)		NA	
Dose 8	$C_{max,n}$ (ng/mL)	30	1161 ± 791	(866,1457)		NA	
	$C_{min,3}$ (ng/mL)	30	384 ± 145	(329,438)		NA	
Dose 9	$C_{max,n}$ (ng/mL)	30	1068 ± 597	(845,1291)		NA	
	$C_{min,3}$ (ng/mL)	31	240 ± 111	(199,280)		NA	
Dose 10	$C_{max,n}$ (ng/mL)	30	725 ± 626	(491,959)		NA	
	$C_{min,3}$ (ng/mL)	30	527 ± 172	(463,591)	28	371 ± 128	(321,420)
Dose 11	$C_{max,n}$ (ng/mL)	30	1013 ± 606	(786,1240)	28	922 ± 563	(703,1140)
	$C_{min,3}$ (ng/mL)	29	275 ± 106	(235,315)	28	315 ± 113	(271,358)
Dose 12	$C_{max,n}$ (ng/mL)	30	973 ± 647	(731,1214)	28	1222 ± 666	(963,1480)
	$C_{min,3}$ (ng/mL)	30	437 ± 194	(365,509)	28	412 ± 156	(352,472)
Dose 13	$C_{max,n}$ (ng/mL)	30	1078 ± 604	(852,1304)	28	776 ± 556	(560,991)
	$C_{min,3}$ (ng/mL)	30	262 ± 90	(229,296)	28	230 ± 89	(195,264)
Dose 14	$C_{max,n}$ (ng/mL)	30	1107 ± 822	(801,1414)	28	1161 ± 715	(864,1439)
	$C_{min,3}$ (ng/mL)	30	378 ± 159	(318,437)	28	381 ± 124	(333,429)
Dose 15	$C_{max,n}$ (ng/mL)	30	1118 ± 661	(871,1364)	28	882 ± 370	(739,1026)
	$C_{min,3}$ (ng/mL)	30	273 ± 124	(227,319)	28	193 ± 99	(154,231)

Abbreviations: $C_{max,n}$, maximum plasma ciprofloxacin concentration after the n^{th} dose, defined as the concentration measured 1 hour after the dose; $C_{min,n}$, minimum plasma ciprofloxacin concentration after the n^{th} dose, defined as the concentration measured just prior to the subsequent dose; NA, not applicable.

[1] The first 2 doses were administered IV.

Source: HGS1021-C1064 Pharmacokinetic Report, Section 3.5.1.4

Table 5. Summary of Ciprofloxacin Pharmacokinetic Parameters Following Oral Administration (Group 1) or Intravenous Followed by Oral Administration (Group 3) with and without a Single Intravenous Dose of Raxibacumab 40 mg/kg in Healthy, Male and Female Subjects

	Group 1		Group 3	
Parameter	PO, Without Raxibacumab (n = 30)	PO, With Raxibacumab (n = 30)	IV, With Raxibacumab (n = 28)	PO, With Raxibacumab (n = 28)
C_{max} (ng/mL)	NA	NA	1854 ± 402	NA
$C_{ss,max}$ (ng/mL)	1436 ± 519	1419 ± 599	NA	1195 ± 566
t_{max} (h)	NA	NA	1.32 (1.23 to 1.60)[1]	NA
$t_{ss,max}$ (h)	1.75 (0.50 to 4.00)[1]	2.00 (0.58 to 4.12)[1]	NA	1.00 (0.5 to 6.00)[1]
$AUC_{0-\infty}$ (ng-h/mL)	NA	NA	6770 ± 1577	NA
AUC_{0-8h} (ng-h/mL)	6413 ± 2241	6742 ± 2279	NA	NA
AUC_{τ} (ng-h/mL)	7694 ± 2680	8151 ± 2673[2]	NA	6615 ± 3224
F (%)	NA	NA	NA	56.99 ± 21.25
$t_{1/2,z}$ (h)	4.74 ± 2.09	5.25 ± 2.47[2]	4.53 ± 0.69	4.62 ± 1.07[3]
MRT (h)	NA	NA	6.01 ± 0.66	NA
CL or CL/F (mL/h)	72422 ± 23995	66781 ± 21874[2]	47555 ± 9547	91061 ± 36852
V_{ss} (mL)	NA	NA	285928 ± 72569	NA
V_z or V_z/F(mL)	510699 ± 340305	436047 ± 212832[2]	312015 ± 93369	630702 ± 309589[3]

Abbreviations: C_{max}, maximum plasma drug concentration for a single dose; $C_{ss,max}$, maximum plasma drug concentration during a steady-state dosing interval; t_{max}, time of occurrence for C_{max}; $t_{ss,max}$, time of occurrence for $C_{ss,max}$; AUC_{0-8h}, area under the plasma drug concentration-time curve from 0 to 8 h post dose; $AUC_{0-\infty}$, area under the plasma drug concentration-time curve from time zero to infinite time for a single dose; AUC_{τ}, area under the plasma drug concentration-time curve during a steady-state dosing interval; F, bioavailable fraction for oral dosing; $t_{1/2,z}$, elimination half-life for the terminal phase; MRT, mean residence time; CL, clearance; CL/F, apparent clearance for oral dosing; V_{ss}, volume of distribution at steady-state; V_z, volume of distribution in the terminal phase; V_z/F, apparent volume of distribution in the terminal phase for oral dosing; NA, not applicable.

[1] Median and range are reported.

[2] n = 29.

[3] n = 27.

Source: HGS1021-C1064 Pharmacokinetic Report, Section 3.5.1.4

Results of the statistical analysis of natural log transformed PK parameters for PO ciprofloxacin administered with and without raxibacumab to Group 1 subjects are summarized in Table 6. Although the prospectively defined assessments of equivalence were to be based on Css,max and AUCτ, as previously noted for 1 subject (US003-000023), no 12 hour specimen was collected after Dose 11. Therefore, the assessment based on AUCτ was based on only 29 subjects and equivalence was also assessed using AUC0-8h. For Css,max, AUCτ, or AUC0-8h, the 90% CI fell within the 80% to 125% range, demonstrating that for those primary parameters Dose 11 (with raxibacumab) was equivalent to Dose 9 (prior to raxibacumab administration). The secondary parameter CL/F also had a 90% CI, indicating equivalence before and after raxibacumab dosing. Based on the results obtained for Css,max and AUCτ, this analysis indicates that ciprofloxacin exposure is equivalent for ciprofloxacin administered alone and when administered with raxibacumab.

Table 6. Summary of Statistical Analysis of Pharmacokinetic Parameters Following Oral Administration (Group 1) or Intravenous Followed by Oral Administration (Group 3) with and without a Single Intravenous Dose of Raxibacumab 40 mg/kg in Healthy, Male and Female Subjects

Parameter	Geometric Means ± Standard Error (n = 30) Reference[1]	Test[2]	Test/ Reference (%)[3]	90% Confidence Interval[4]
Primary Assessments:				
$C_{ss,max}$ (ng/mL)	1349 ± 95	1310 ± 109	100.42	92.17, 108.66
AUC_τ (ng·h/mL)	7273 ± 489	7751 ± 498[5]	107.99	102.53, 113.45
Secondary Assessments:				
AUC_{0-8h} (ng·h/mL)	6060 ± 409	6391 ± 416	107.15	101.22, 113.09
$t_{1/2,z}$ (h)	4.43 ± 0.38	4.86 ± 0.46[5]	122.99	104.65, 141.33
CL/F (mL/h)	68587 ± 4379	63487 ± 4062[5]	92.67	88.30, 97.04
V_z/F(mL)	438639 ± 62131	445031 ± 39522[5]	114.42	96.15, 132.70

Abbreviations: $C_{ss,max}$, maximum plasma drug concentration during a steady-state dosing interval; AUC_τ, area under the plasma drug concentration-time curve during a steady-state dosing interval; AUC_{0-8h}, area under the plasma drug concentration-time curve from 0 to 8 h post dose; $t_{1/2,z}$, elimination half-life for the terminal phase; CL/F, apparent clearance for oral dosing; V_z/F, apparent volume of distribution in the terminal phase for oral dosing; NA, not applicable.

1 9th PO ciprofloxacin dose (Day 4, prior to raxibacumab) from Group 1.

2 11th PO ciprofloxacin dose (Day 5, with raxibacumab) from Group 1.

3 Mean of the individual test:reference ratios, expressed as a percentage.

4 90% CI for the individual test:reference ratios, expressed as percentages.

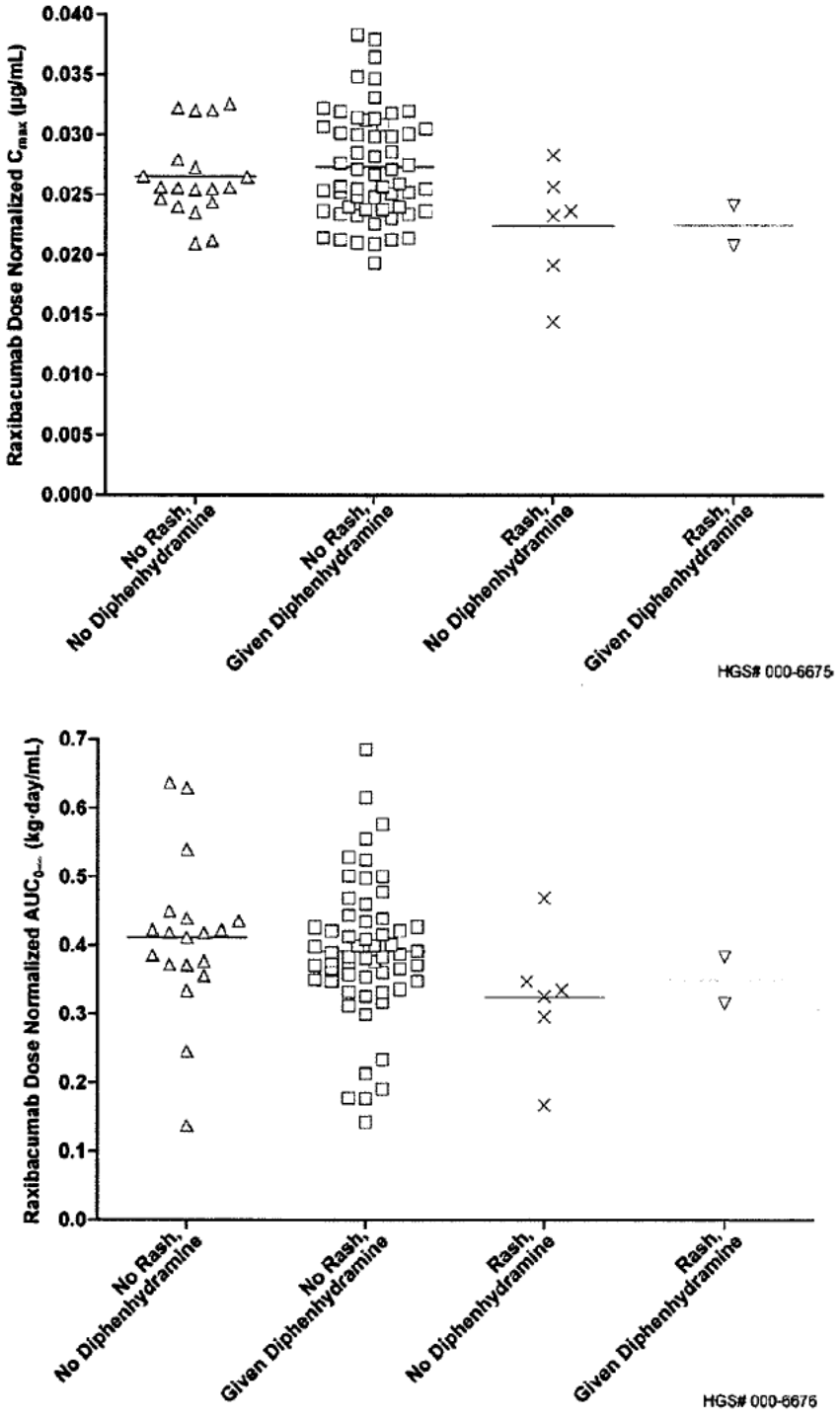
5 n = 29.

Pharmacodynamics

Several subjects treated with raxibacumab experienced mild to moderate rashes in this study (n = 8/86 subjects). Plots of Cmax/Dose or AUC0-∞/Dose in subjects experiencing rash versus those who did not experience rash are displayed in Figure 4. Diphenhydramine was administered within 60 minutes prior to raxibacumab administration in 61/86 subjects. Mean Cmax/Dose for subjects who experienced rash was lower (by approximately 17%) than those for the subjects who did not experience rash, while diphenhydramine use had little, if any, impact on Cmax/Dose; this difference was statistically significant (p = 0.0334), but none of the Bonferroni's multiple comparison post tests achieved significance (p > 0.05). Mean AUC0-∞/Dose for subjects who experienced rash tended to be lower (by approximately 11% to 21%) than those for the subjects who did not experience rash, while diphenhydramine use had no consistent impact on AUC0-∞/Dose; this difference was not statistically significant (p = 0.3312). Overall, raxibacumab exposure appears to be somewhat lower in the subjects who experienced rash, relative to those

who did not. Raxibacumab exposure does not appear to differ between subjects who were administered diphenhydramine and those who were not.

Figure 4. Individual and Mean Raxibacumab Dose Normalized Cmax and AUCinf Following a Single 40 mg/kg Raxibacumab IV Infusion Dose, Administered with or without PO or IV Ciprofloxacin Coses, in Subjects Who Did or Did Not Experience Rash, and Who Were or Were Not Administered Diphenhydramine



HGS1021-C1064 Pharmacokinetic Report, Section 3.6

Immunogenicity
None of the subjects in this study had a positive anti-raxibacumab antibody response.

APPLICANT'S DISCUSSION:
The objectives of this study were to determine the effect of co-administration of raxibacumab on PO ciprofloxacin PK, and to characterize the effect of co-administration of PO and IV ciprofloxacin on raxibacumab PK. Subjects in this study were administered a single 40 mg/kg IV infusion raxibacumab dose, with or without co-administered ciprofloxacin (IV or PO). Plasma ciprofloxacin concentration-time profiles were very similar for Group 1 at PO Dose 9 (prior to raxibacumab) and PO Dose 11 (with raxibacumab), with overlapping SD error bars. The 90% CI for the primary PK parameters Css,max and AUCτ were within the 80% to 125% equivalence limits. Thus, it can be concluded that ciprofloxacin PK were equivalent when ciprofloxacin was administered alone or when it was administered with raxibacumab. For Group 3, which was administered IV ciprofloxacin in combination with raxibacumab, followed by q12h PO ciprofloxacin doses, the ciprofloxacin exposure for the PO doses was similar to or slightly lower than that for the subjects administered PO ciprofloxacin doses in Group 1.

Serum raxibacumab concentration-time profiles were very similar among treatment groups, with overlapping SD error bars. Statistically significant differences in PK parameters were not encountered for Group 3 (raxibacumab + IV ciprofloxacin) vs the raxibacumab alone group. Overall, exposure to ciprofloxacin appears to have no consistent or meaningful impact on raxibacumab PK.

Eight of the 86 subjects administered raxibacumab in this study experienced adverse events of mild to moderate severity rash following raxibacumab administration. In subjects who experienced rash, raxibacumab exposure tended to be decreased, relative to that for subjects who did not experience rash. This suggests that the probability that a subject would experience rash is more closely linked to some other subject characteristic(s) than the magnitude of raxibacumab exposure. This is supported by the observation that rashes were observed in the 3 subjects who did not receive the entire planned raxibacumab dose, and hence had the lowest raxibacumab exposures. Raxibacumab exposure was not altered in subjects administered diphenhydramine, relative to those who did not take diphenhydramine, independent of whether the subjects experienced rash or not.

Raxibacumab PK following a single 40 mg/kg IV dose to healthy subjects were previously assessed in a Phase 1 safety study (Clinical Study Report No. PAM-NH-01.CSR, 2006). Raxibacumab PK for raxibacumab administered alone in the current study (n = 28 subjects) are similar to those obtained in the previous study (n = 7 subjects). Mean Cmax were similar, at 988 μg/mL in the current study and 1042 μg/mL in the previous study. Mean AUC0-∞ were also similar (15328 and 15554 μg·day/mL in the current and previous studies, respectively).
The mean t1/2,z in the current study was 20 days, compared with a mean t1/2,z of 16 days in the previous study. The mean CL in the previous study was 2.6 mL/kg/day, while that in the current study was 2.9 mL/kg/day. Overall, there is reasonable agreement in the raxibacumab PK obtained in these two studies.

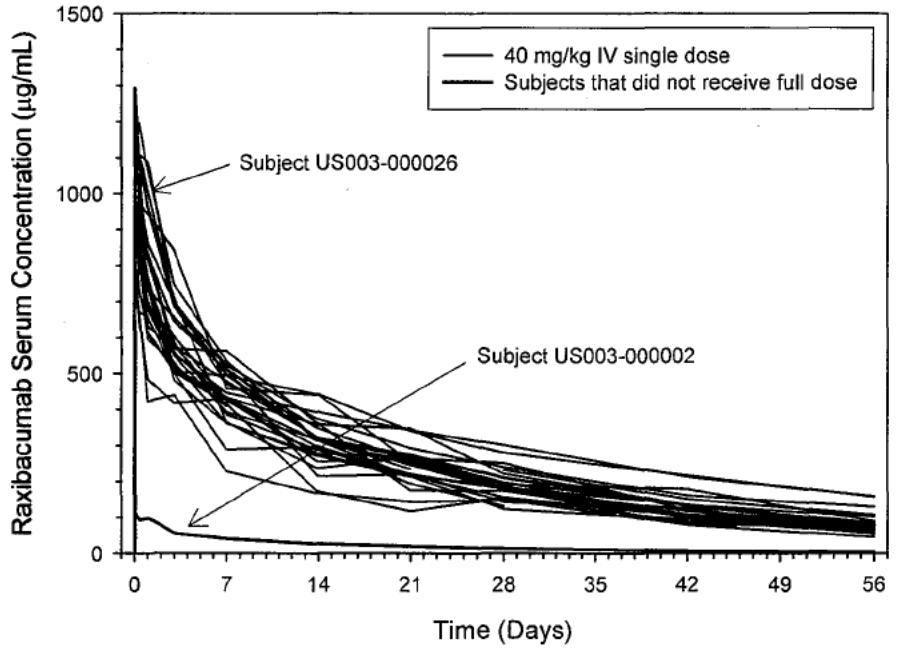
APPLICANT'S CONCLUSIONS:
Co-administration of IV raxibacumab had no effect on ciprofloxacin exposure for PO ciprofloxacin doses. Exposure to ciprofloxacin appears to have no consistent or meaningful impact on raxibacumab PK. In subjects who experienced rash, raxibacumab exposure tended to be decreased, relative to that for subjects who did not experience rash, suggesting that rash is more closely linked to other subject characteristic(s) than magnitude of raxibacumab exposure.

Raxibacumab exposure was not altered in subjects administered diphenhydramine, relative to those who did not take diphenhydramine, independent of whether the subjects experienced rash or not.

REVIEWER ASSESSMENT:
Results from Study HGS1021-C1064 adequately determined the effect of co-administration of raxibacumab on oral ciprofloxacin pharmacokinetics. The applicant's pharmacokinetic conclusions based on these findings are valid.

In Group 2 (raxibacumab administered alone), two subjects (Subject US003-000002 and US003-000026) were reported in summary tables to have not received the complete raxibacumab dose. In the remainder of the applicant's report, Subject US003-000026 was not reported to have been one of the subjects that received an incomplete infusion; only the following three subjects were reported to have received an incomplete infusion: Subjects US003-000002, US003-000006, and US003-000028. Figure 5 shows the raxibacumab serum concentration-time profiles for both subjects compared to the group profiles. The serum concentration-time profile for Subject US003-000026 was not consistent with an incomplete dose. Conversely, the profile for Subject US003-000002 was consistent with an incomplete infusion. This subject was included in summary descriptive statistics for PK parameters in the applicant's report. For this subjects PK parameters were calculated based on the actual dose administered. Therefore, Cmax and AUC0-∞ were dose normalized for any comparisons among dose groups or subgroups of subjects. For completeness and a more accurate comparison of PK parameters across groups, descriptive statistics for raxibacumab PK parameters excluding Subject US003-000002 are presented in Table 7. Exclusion of this subject does not impact the final conclusions from this study.

Figure 5. Individual Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 40 mg/kg in Healthy, Male and Female Subjects (Linear and Log Scale)



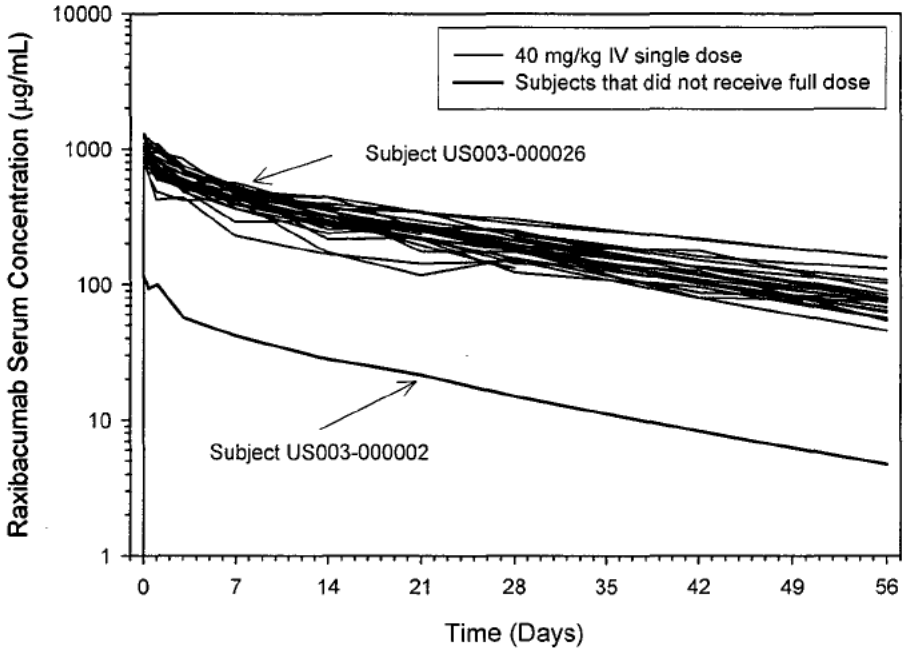


Table 7. Summary of Raxibacumab Pharmacokinetic Parameters Following a Single 40 mg/kg Raxibacumab IV Infusion Administered with or without PO or IV Ciprofloxacin

Parameter	Group 1[a] (n = 27)	Group 2[b] (n = 27)	Group 3 (n = 28)
Cmax (µg/ml)	1143.3 ± 169.5 (833.6 – 1517.8)	1020.3 ± 140.6 (766.2 – 1293.7)	1047.8 ± 180.3 (772.5 – 1458.4)
AUCinf (µg·day/ml)	14871.9 ± 3821.1 (5494.5 – 21116.6)	15845.8 ± 4333.5 (7614.9 – 25464.6)	16349.3 ± 4255.7 (7117.5 – 27465.2)
Half life (days)	19.8 ± 8.18 (4.27 – 45.8)	20.6 ± 6.54 (5.93 – 32.3)	21.5 ± 8.92 (6.53 – 42.9)
CL (ml/day/kg)	2.99 ± 1.38 (1.85 – 7.30)	2.73 ± 0.84 (1.57 – 5.24)	2.63 ± 0.82 (1.46 – 5.62)
Vss (ml/kg)	64.9 ± 11.9 (44.1 – 99.8)	69.7 ± 13.7 (45.6 – 106.7)	67.2 ± 12.6 (47.6 – 93.2)

Group 1, raxibacumab 40 mg/kg + cipro PO
Group 2, raxibacumab 40 mg.kg only
Group 3, raxibacumab 40 mg/kg + cipro IV/PO
Data presented represents mean ± one standard deviation and (minimum – maximum)
[a] Subjects US003-000006 and US003-000028 excluded for receiving only partial doses of raxibacumab. Subject US003-00029 excluded for PK profile uncharacteristic of IV administration.
[b] Subject US003-000002 excluded for receiving only a partial dose of raxibacumab.

For further evalauation of the safety results for Study HGS1021-C1064, refer to the Medical Officers' review for BLA 125349.

HGS1021-C1069:
An Open-Label Study to Evaluate the Immunogenicity and Safety of Raxibacumab (Human Monoclonal Antibody to *B. anthracis* Protective Antigen) Administered in Healthy Subjects

Date(s): 08JAN2008 to 08MAY2008

Investigator and Clinical Site: 2 US sites

OBJECTIVES:

Primary: To evaluate immunogenicity in subjects receiving two doses of raxibacumab with a delay prior to the second dose.

Secondary: (1) To evaluate safety in subjects receiving two doses of raxibacumab with a delay prior to the second dose, and (2) to determine serum raxibacumab PK in subjects receiving two doses of raxibacumab with a delay prior to the second dose.

STUDY DESIGN:
HGS1021-C1069 was a single-group, open-label study to evaluate the immunogenicity and safety of raxibacumab in healthy adult male and female subjects. Subjects who had received raxibacumab ≥ 4 months prior to this study were to be enrolled (in Study HGS1021-C1064). A maximum of 25 subjects (to include at least 3 evaluable female subjects) were to receive a 2nd dose of raxibacumab equal to that of their previous dose ≥ 4 months following their first dose. Subjects were to remain in the clinical unit from Day 0 until Day 1 and were followed for 70 days after receiving their second dose of raxibacumab. Blood specimens were to be collected during the study and analyzed for anti-raxibacumab antibodies. Subjects with a positive anti-raxibacumab antibody response anytime following the second dose were to have a serum sample collected approximately six months after the last dose for a follow-up immunogenicity assessment. All subjects were to be treated with 50 mg oral (PO) diphenhydramine up to 60 minutes prior to infusion of raxibacumab.

FORMULATIONS:
Raxibacumab was supplied in 50 mL sterile, single-use vials containing 34 mL of liquid formulation per vial. Each vial contained 50 mg/mL raxibacumab in 0.12 mg/mL citric acid, 2.8 mg/mL sodium citrate, 10 mg/mL sucrose, 18 mg/mL glycine, 0.2 mg/mL polysorbate 80, pH 6.5. Lot 71044 was administered to the subjects as a 40 mg/kg IV infusion; the same lot was administered to these subjects in Study HGS1021-C1064.

PHARMACOKINETIC ASSESSMENTS:
Blood samples for serum raxibacumab concentration measurement were collected from all subjects just prior to administration of the second raxibacumab dose, at 5 minutes and 8 hours after completion of the second raxibacumab infusion, and at 1, 3, 7, 14, 21, 28, 42, and 56 days after the 2nd raxibacumab dose.

IMMUNOGENICITY ASSESSMENTS:
Blood samples were to be collected for immunogenicity assessment at screening, prior to raxibacumab dosing on Day 0, and on Days 28, 56, and 70 (End of Study). Subjects with a positive anti-raxibacumab antibody titer following raxibacumab administration on Day 0 were to have a serum sample collected approximately six months after the dose for follow-up immunogenicity assessment.

BIOANALYTICAL ANALYSIS:
Serum samples were analyzed for raxibacumab using an electrochemiluminescence (ECL)-based assay. Biotinylated PA is bound to a streptavidin coated Meso Scale Discovery (MSD) 96-well assay plate for raxibacumab capture. The raxibacumab in diluted serum samples binds to the biotinylated PA and is detected by the addition of rabbit anti-raxibacumab followed by goat anti-human antibody labeled with MSD SULFOTAG™, an ECL label. MSD Read Buffer is added to the plate and the plate is inserted into an MSD plate reader, where voltage applied to the plate electrodes causes the MSD SULFO-TAG™ to emit light proportional to the amount of raxibacumab present in the serum. The concentration of raxibacumab in serum samples is interpolated from an 8-point reference standard curve. The lower limit of quantitation (LLOQ) is 800 ng/mL of raxibacumab in 100% serum.

Immunogenicity testing was performed to determine if raxibacumab induced an anti-raxibacumab immune response. Testing comprised 2 assays (screening and confirmatory). The screening assay (direct binding) is an electrochemiluminescence (ECL)-based bridging assay. A rabbit polyclonal antibody was used as a positive control. Samples above the assay cut point were considered positive. Samples identified as positive in the screening assay were to be confirmed positive in a confirmatory assay. Samples must have demonstrated a significant percent drop in the confirmatory inhibition of binding assay to be considered positive. The inhibition of binding confirmatory assay was performed identically to the direct binding screening assay with the exception that the samples were tested in parallel with excess unlabeled raxibacumab. The assay limit of detection is 62.5 ng/mL.

PHARMACOKINETIC/PHARMACODYNAMIC/STATISTICAL ANALYSIS:
PK parameters were to be calculated from the serum raxibacumab concentration-time results. Serum raxibacumab concentration data and PK were summarized using mean, median, and standard error of the mean (SE), standard deviation (SD), coefficient of variation (CV%), geometric mean, 95% confidence interval (CI), and number of subjects.

For the purpose of comparison, the raxibacumab PK results for the first dose (administered during study HGS1021-C1064) were reproduced and summarized in this report, with descriptive statistics. To assess potential differences in raxibacumab PK between the two dosing occasions, paired t-tests were used; the exception was the time of occurrence for the maximum serum drug concentration (tmax), for which the Wilcoxon matched pairs test was used. A significance level of $\alpha = 0.05$ was used for all statistical comparisons, unless otherwise noted.

To correct for the minimal carryover that the measurable predose concentrations might represent, the area under the serum raxibacumab concentration time curve from predose to infinite time ($AUC_{predose-\infty}$) was calculated by dividing the predose concentration by λz.

RESULTS:
Study Population
A total of 23 subjects were screened to provide 20 subjects for the study. All 20 subjects were treated with 40 mg/kg IV raxibacumab per protocol, and all subjects completed the study. The mean time between raxibacumab doses was 7.6 months with a range of 6 to 9 months. All 20 subjects enrolled in this study were considered evaluable for raxibacumab PK.

Demographics
A summary of demographic and baseline characteristics for the study population is presented in Table 1.

Table 1. Demographics and Baseline Characteristics – Study HGS1021-C1069

Demographic	Raxibacumab 40 mg/kg (n = 20)
Sex	12 (60.0%) Male 8 (40.0%) Female
Race	13 (65.0%) White 7 (35.0%) Black or African American 4 (20.0%) Hispanic
Age (years)	40.6 (23 – 61)
Weight (kg)	80.1 (56.1 – 100.5)

Data presented represents mean (range).
Source: Study Report HGS1021-C1069, Section 6.5.1.1

Raxibacumab Serum Pharmacokinetics

Eleven of the 20 subjects administered raxibacumab in this study (Subjects US001-001, US002-001, US002-002, US002-004, US002-007, US002-009, US002-011, US002-012, US002-013, US002-014, and US002-017) had measurable serum raxibacumab concentrations prior to administration of the second raxibacumab dose (the dose administered in this study), ranging from 0.74 to 7.13 μg/mL. For these subjects, the predose concentration was less than 0.7% of the peak concentration, and was less than 5% of the concentration measured at 56 days postdose. PK calculations were corrected for the minimal carryover that these measurable predose concentrations might represent (as described in PK/PD Analysis method section).

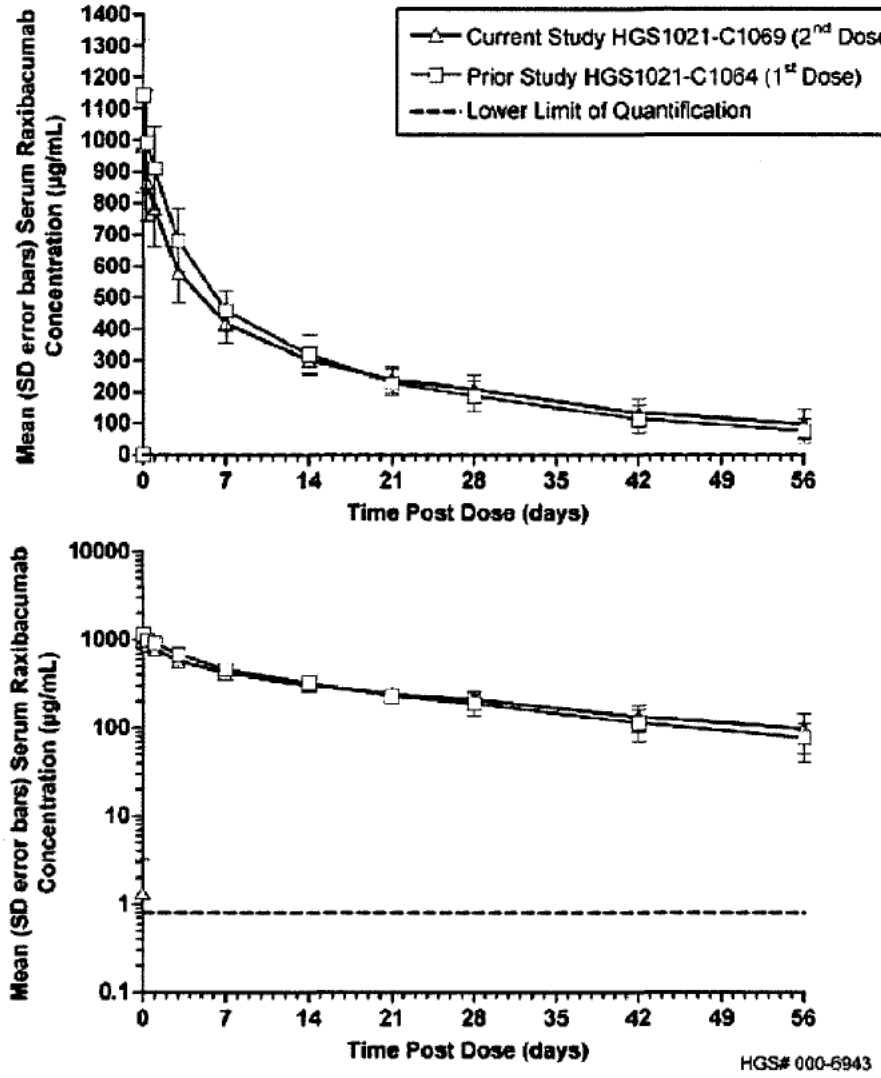
Missing Data

Only one specimen was not received for serum raxibacumab analysis for this study. For Subject US002-014, the 14 days postdose specimen was not collected.

Raxibacumab Serum Concentrations and PK Parameters

Serum concentration-time profiles for raxibacumab following the first (Study HGS1021-C1064) and second (current study) doses of raxibacumab 40 mg/kg IV administered at least four months apart in healthy, male and female subjects are presented in Figure 1. Serum raxibacumab concentration-time profiles were very similar for the first and second doses, with overlapping SD error bars.

Figure 1. Mean (± SD) serum concentration-time profiles for raxibacumab following the first (Study HGS1021-C1064) and second (current study) doses of raxibacumab 40 mg/kg IV administered at least four months apart in healthy, male and female subjects (linear and log scale)



Source: Study Report HGS1021-C1069, Section 9.2.4

Pharmacokinetic parameters for raxibacumab following the first (Study HGS1021-C1064) and second (current study) doses of raxibacumab 40 mg/kg IV administered at least four months apart in healthy, male and female subjects are summarized in Table 2.

The disappearance of raxibacumab from serum appeared to be biphasic, and the mean t1/2,z ranged from 21 to 26 days. The mean Vss ranged from 65 to 76 mL/kg and were at least 51% greater than plasma volume (approximately 42.8 mL/kg). Results suggest that raxibacumab distributes to tissues. Mean CL ranged from 2.37 to 2.59 mL/day/kg; these CL values are much smaller than the glomerular filtration rate indicating that, as expected, there is virtually no renal clearance of this mAb.

Results of the statistical analysis of PK parameters for the first and second doses are also presented in Table 2. Subject US002-014 (US003-022 in the prior study) only had serum raxibacumab concentrations determined through seven days postdose in the prior study, it is possible that subject's results for $AUC_{0-\infty}$, t1/2,z, MRT, CL, Vss, and Vz could have been biased. Therefore, the statistical analyses for those parameters were were performed with and without this subject's data. Since statistical significance was unaffected by inclusion of that subject's results, the statistical analysis results including that subject are presented in Table 2.

There were no statistically significant differences ($p \geq 0.0594$) in mean AUC0-∞, t1/2,z, MRT, CL, or Vz between the second dose administered in the current study and the first dose administered in study HGS1021-C1064. The mean Cmax for the second dose (979 µg/mL) was significantly lower (-15%, $p = 0.0008$) than that for the first dose (1152 µg/mL). Relative to this, the mean Vss for the second dose in this study (76 mL/kg) was significantly larger (17%, $p = 0.0122$) than that for the first dose in study HGS1021-C1064 (65 mL/kg), suggesting that the difference in mean Cmax between the studies is related to a shift in volume of distribution between the first and second dose. Although the difference in mean Cmax between this study and the prior study is statistically significant, the difference is less than 20%. Furthermore, out of the 20 subjects, nine had decreases in Cmax of more than 20% for the second dose relative to the first, while another five subjects had increases in Cmax of more than 20%, and the remaining six subjects had a Cmax for their second dose that was within 20% of the Cmax for their first dose. Given the lack of consistent decreased Cmax for the second dose relative to the first dose among the subjects, it is questionable if the statistical significance of the difference in mean Cmax is meaningful.

The only other parameter for which there was a statistically significant difference between the first and second dose was tmax. For the second dose in the current study, the median tmax was 0.099 days, while the median tmax for the first dose in study HGS1021-C1064 was 0.102 days. Review of the individual results show that the slightly larger tmax for the first dose may be driven by two subjects who appear to be outliers for that dose. Although the small difference in median tmax (-3%) attained statistical significance, it is unlikely to be clinically meaningful.

Table 2. Summary of raxibacumab pharmacokinetic parameters following first (Study HGS1021-C1064) and second (current study) doses of raxibacumab 40 mg/kg IV administered at least four months apart in healthy, male and female subjects

Parameter	Current Study (HGS1021-C1069) (2^{nd} dose) (n = 20)	Prior Study (HGS1021-C1064) (1^{st} dose) (n = 20)
C_{max} (µg/mL)	979 ± 148	1152 ± 176
	(p = 0.0008[1])	
C_{max}/Dose (kg/mL)	0.0245 ± 0.0037	0.0290 ± 0.0047
	(p = 0.0011[1])	
t_{max} (day)[2]	0.099 (0.097 to 0.104)	0.102 (0.097 to 0.431)
	(p = 0.0019[1])	
$AUC_{0-\infty}$ (µg·day/mL)	18239 ± 6179	16440 ± 4140
	(p = 0.1798[1])	
$AUC_{0-\infty}$/Dose (kg·day/mL)	0.4566 ± 0.1538	0.4122 ± 0.1017
	(p = 0.1853[1])	
$t_{1/2,z}$ (day)	25.68 ± 11.19	21.20 ± 8.62
	(p = 0.1535[1])	
MRT (day)	35.09 ± 15.58	27.21 ± 8.62
	(p = 0.0594[1])	
CL (mL/day/kg)	2.37 ± 0.63	2.59 ± 0.77
	(p = 0.3017[1])	
V_{ss} (mL/kg)	75.72 ± 11.42	64.73 ± 14.02
	(p = 0.0122[1])	
V_z (mL/kg)	80.07 ± 12.33	72.60 ± 18.96
	(p = 0.1425[1])	

Abbreviations: C_{max}, maximum serum raxibacumab concentration for a single dose; t_{max}, time of occurrence for C_{max}; $AUC_{0-\infty}$, area under the serum raxibacumab concentration-time curve from time 0 to infinite time for a single dose; $t_{1/2,z}$, elimination half-life for the terminal phase; MRT, mean residence time; CL, clearance; V_{ss}, volume of distribution at steady-state; V_z, volume of distribution in the terminal phase.

1 P-value from a paired t-test; the exception is t_{max}, for which the p-value is from a Wilcoxon matched pairs test.

2 Median and range are presented.

Source: HGS1021-C1069, Section 3.5.1.4

Immunogenicity

None of the subjects in study HGS1021-C1064 developed an anti-raxibacumab antibody response, and none of the subjects in this study had a positive anti-raxibacumab antibody response following raxibacumab readministration.

APPLICANT'S DISCUSSION:

The objective of this study was to determine raxibacumab pharmacokinetics (PK) for a second intravenous (IV) dose administered at least four months after the first dose. No immunogenicity was detected in these subjects following either the first or second raxibacumab dose.

Serum raxibacumab concentration-time profiles were very similar for the first and second doses, with overlapping SD error bars. Although there were statistically significant differences in volume of distribution (Vss) that were reflected by differences in Cmax, those differences were small (≤ 17%). **In addition, not all subjects had a decreased Cmax** following the 2nd dose in this study. It is questionable if the statistical significance of the difference in Cmax means is clinically meaningful. The small (3%) but statistically significant difference in tmax between the

first and second doses is unlikely to be clinically meaningful. There were no other statistically significant differences between the 1st and 2nd doses for other PK parameters ($AUC_{0-\infty}$, CL, t1/2,z, MRT and Vz). These results indicate that raxibacumab PK were not meaningfully different for two doses that were administered at least four months apart.

Although not prespecified in the protocol, the subjects enrolled in this study represented a subset of the subjects enrolled in the previous raxibacumab–ciprofloxacin interaction study (Clinical Study HGS1021-C1064). The mean PK parameters for this subgroup of subjects (Table 3-2) are similar to those obtained for the three treatment groups in the prior study.

APPLICANT'S CONCLUSIONS:

- None of the subjects in this study had a positive anti-raxibacumab antibody response.
- Raxibacumab PK were not meaningfully different for 2 single IV doses administered at least 4 months apart.
- The subgroup of subjects enrolled in this study had PK similar to that for all subjects from the prior study.

REVIEWER ASSESSMENT:

Results from Study HGS1021-C1069 adequately achieved the objectives of 1) evaluating immunogenicity in subjects receiving two doses of raxibacumab with a delay prior to the second dose, and 2) determining serum raxibacumab PK in subjects receiving two doses of raxibacumab with a delay prior to the second dose. The applicant's pharmacokinetic conclusions based on these findings are valid.

Interestingly, eleven of the 20 subjects administered raxibacumab in this study had measurable serum raxibacumab concentrations prior to administration of the second raxibacumab dose. These measurable concentrations occurred as far out as 6 to 9 months in these subjects. This finding was unexpected since the reported half-life of raxibacumab is approximately 20 days. PK calculations were appropriately corrected for the minimal carryover that these measurable predose concentrations might represent.

For further evalauation of the safety results for subjects receiving two doses of raxibacumab with a delay prior to the second dose in Study HGS1021-C1069, refer to the Medical Officers' review for BLA 125349.

4.2. Pharmacometric Review

OFFICE OF CLINICAL PHARMACOLOGY PHARMACOMETRIC REVIEW

1 SUMMARY OF FINDINGS

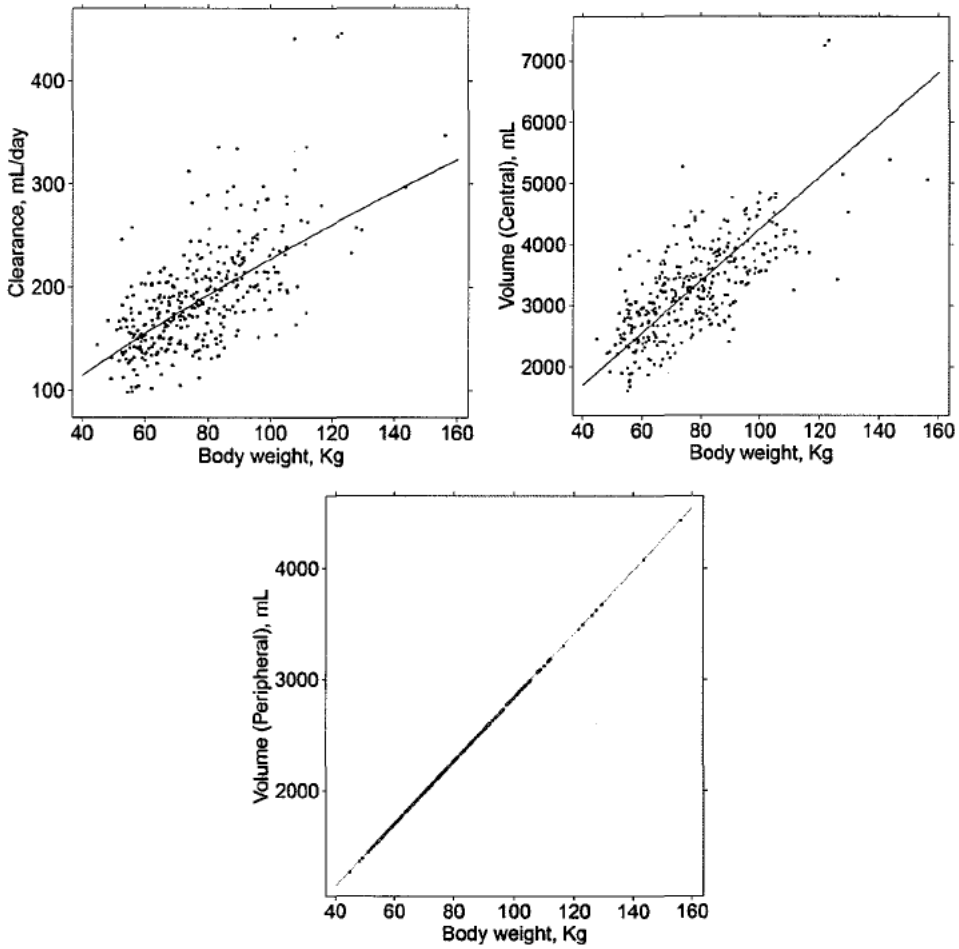
1.1 Key Review Questions

The purpose of this pharmacometrics review is to address the following key questions:

1.1.1 Does body weight affect raxibacumab PK to justify mg/kg dosing?

Body weight is a significant covariate for raxibacumab PK, specifically on clearance and volume of distribution parameters.

Figure 1.1.1-1 Effect of Body Weight on Raxibacumab PK



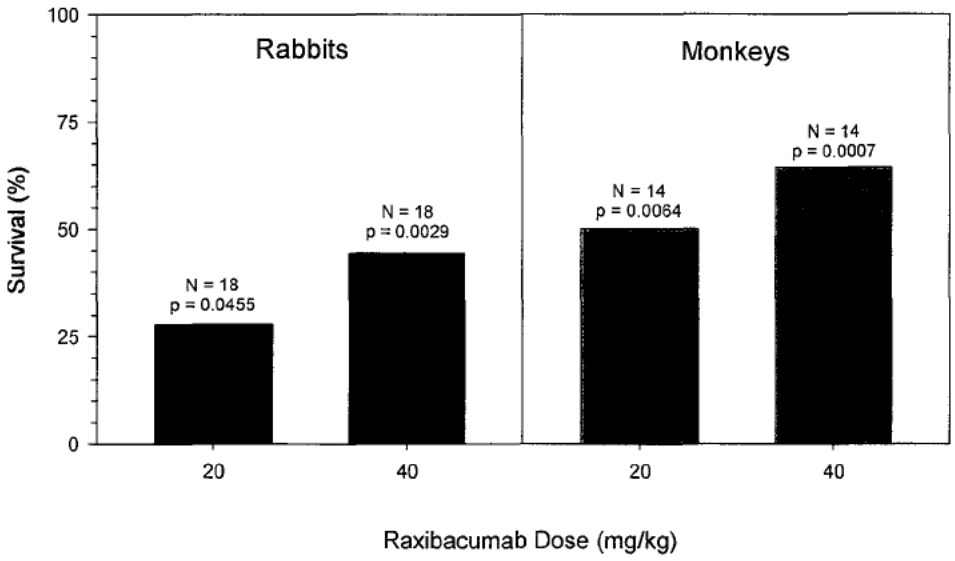
Based on simulated subjects receiving the same 40 mg/kg dose, Cmax increases by 61% over a weight range of 45 to 156 kg, while AUC0-∞ increases by 22%. Therefore, the applicant's proposed weight-based dosing accounts for increase in clearance with body weight. The simulated exposures for body weights of 45 and 156 kg are within the range of exposures observed in human subjects receiving raxibacumab 40 mg/kg in clinical trials. Other factors, such as age and race, did not affect raxibacumab PK to warrant dose adjustments. In the reviewer's analysis, body weight did not affect inter-compartmental clearance.

1.1.2 What is the relationship (if any) between protective antigen (PA) concentrations, raxibacumab concentrations, and outcome (survival or death) in monkeys and rabbits?

Survival rates by raxibacumab dose in the pivotal animal efficacy studies are summarized in Figure 1.1.2-1. Survival rates in both studies exhibited a dose-response for 20 mg/kg and 40 mg/kg doses of raxibacumab IV.

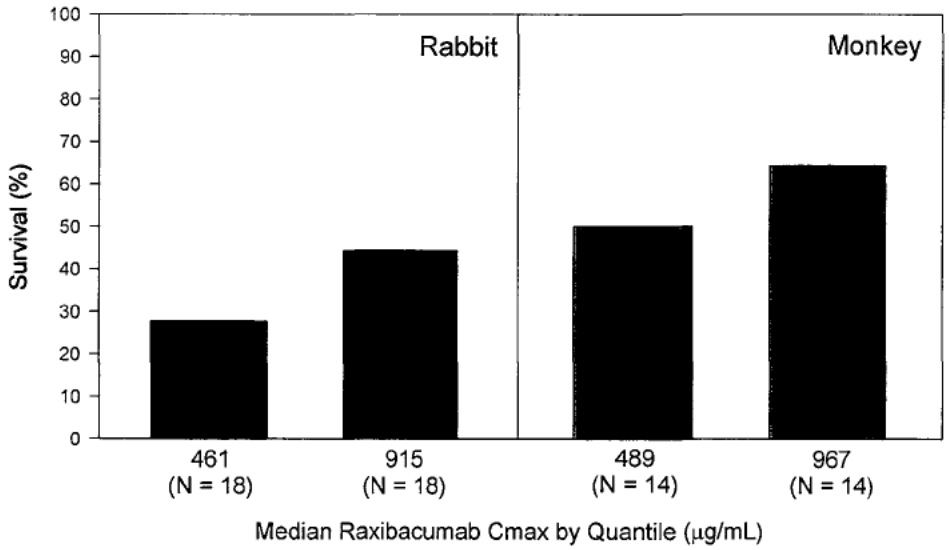
However, a PK/PD relationship between plasma/serum PA concentrations and serum raxibacumab concentrations could not be described based on the limitations of the PA data collected in the pivotal animal efficacy studies (e.g. poor parameter estimation, analytical complications, significant inherent intra- and inter-individual variability, etc.).

Figure 1.1.2-1 Percent Survival in Rabbits and Monkeys Treated with Raxibacumab for Anthrax



The applicant concluded that a human raxibacumab dose should attain a sufficiently high Cmax to optimize the likelihood of achieving efficacy. Mirroring the dose-response seen with 20 and 40 mg/kg doses of raxibacumab, a concentration-response is seen between the probability of survival and quantiles of raxibacumab Cmax, as presented in Figure 1.1.2-2. The data suggest a relationship between raxibacumab dose, concentration, and survival. Although the applicant proposes a sufficiently high Cmax optimizes the likelihood of efficacy, a threshold of Cmax was not identified.

Figure 1.1.2-2 Comparison of Probabilities of Survival by Raxibacumab Cmax Quantile

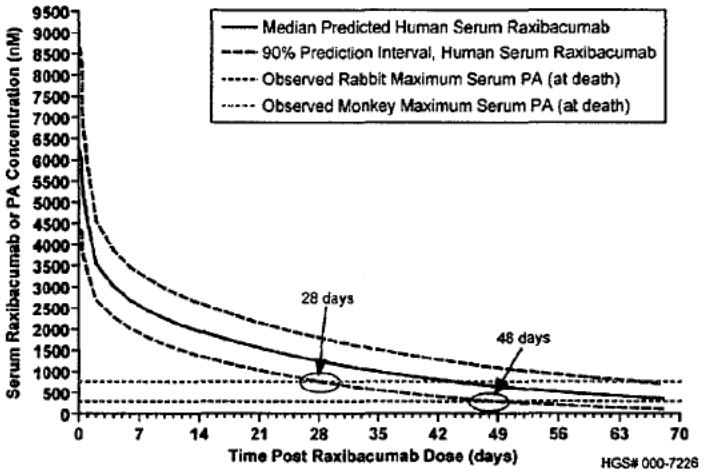


1.1.3 Does the applicant's assessment of PA kinetics and the estimated proportion of humans achieving protective raxibacumab concentrations support the proposed dose of 40 mg/kg?

The applicant's rationale on the proportion of a human population that would attain protective serum raxibacumab concentrations when administered a single 20 mg/kg or 40 mg/kg intravenous (IV) raxibacumab dose support the proposed clinical dose of raxibacumab 40 mg/kg. The applicant defined an effective dose in humans as one that achieved 1) a sufficiently high Cmax equal to or in excess of concentration needed to bind systemic PA, 2) at least 28 days duration of protective serum raxibacumab levels to allow the innate immune response to PA to develop, and 3) equimolar or higher serum raxibacumab levels than the highest expected PA levels for 28 or 48 days, using PA levels from monkeys and rabbits, respectively. Median serum raxibacumab profiles (with 90% prediction intervals) for a 40 mg/kg single IV dose in comparison to the expected highest PA concentrations to be encountered are presented in Figure 1.1.3-1. Following a 40 mg/kg dose, serum raxibacumab levels are equimolar to or greater than the highest expected PA levels for 28 or 48 days, using PA levels from monkeys and rabbits, respectively, following up to 200 × LD_{50} inhalational exposure to *B. anthracis*.

The rationale presented by the applicant (with reasonable assumptions) suggests appropriateness of 40 mg/kg. However, dose and concentration response for the endpoint of survival indicates potential to achieve higher response rate at higher doses. If future trials are conducted, higher doses (60 or 80 mg/kg) should be explored along with 40 mg/kg (see the following question in section 1.1.4 for the safety aspects of this recommendation).

Figure 1.1.3-1 Median and 90% Prediction Interval Serum Raxibacumab Profiles for a 40 mg/kg Single IV Dose Versus the Highest Expected PA Concentrations to be Encountered

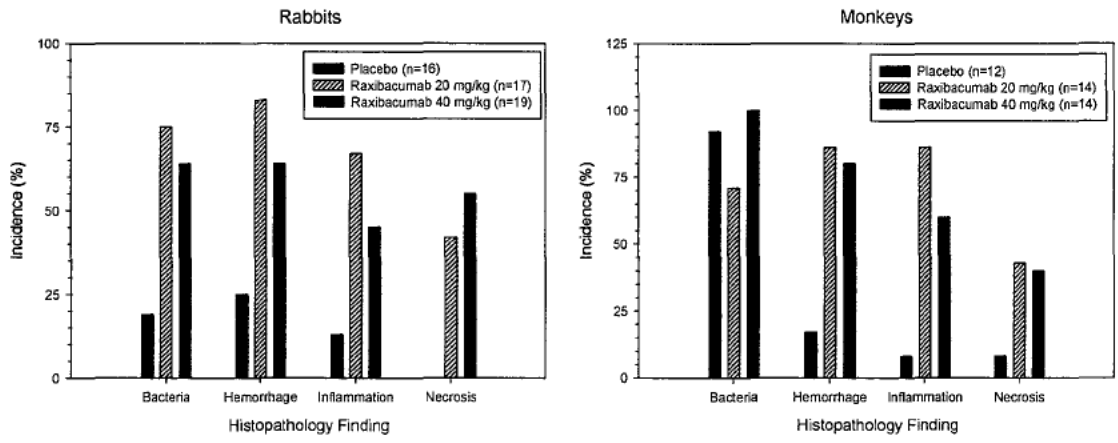


1.1.4 What is the relationship (if any) between raxibacumab PK and CNS findings (ie meningitis) in animals in the pivotal efficacy studies (682-G005758 and 724-G005829)?

No clear relationship between exposure and CNS pathology can be discerned based on the limited amount of data in animals that died.

In the pivotal animal studies, animals that were treated with raxibacumab and died exhibited a higher incidence of histopathologic findings in the brain, as presented in Figure 1.1.4-1. Histopathologic findings were also of higher severity in raxibacumab treated animals versus animals receiving placebo treatment. The raxibacumab 20 mg/kg dose group had a higher incidence and severity of CNS findings versus the 40 mg/kg group, suggesting an absence of clear dose-response relationship for brain histopathology.

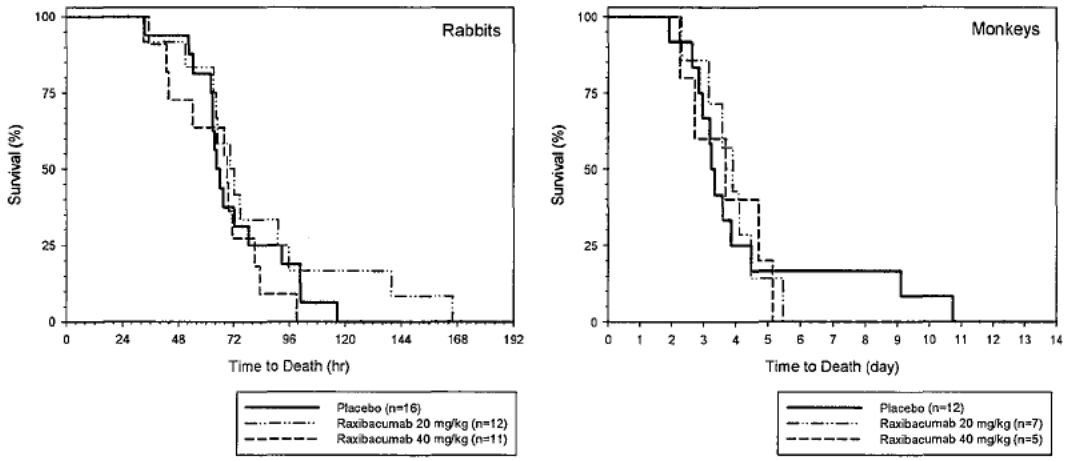
Figure 1.1.4-1 Histopathology Findings by Treatment in Rabbits and Monkeys that Died



The major limitation of these data is the absence of data from animals that survived as those animals were not sacrificed and evaluated for brain histopathology findings.

A potential hypothesis proposed by the applicant to explain the finding that adverse CNS findings were more prevalent in animals that received raxibacumab and died versus animals that received placebo and died is that animals that received raxibacumab survived longer and brain lesions increase over time. Figure 1.1.4-2 shows percent survival and survival time by raxibacumab dose for rabbits and monkeys that died in the pivotal studies. The survival plots show that in rabbits that died, placebo animals have comparable survival times versus animals that received 40 mg/kg of raxibacumab. In monkeys that died, two or three animals that received placebo lived longer than those receiving raxibacumab. Thus, the potential for raxibacumab treatment to result in adverse CNS findings is not likely to be related to prolonged survival time. In addition, an examination of time to death by exposure quantiles indicated the magnitude of raxibacumab exposure did not affect survival time in animals that died.

Figure 1.1.4-2 Time to Death in Rabbits and Monkeys That Died

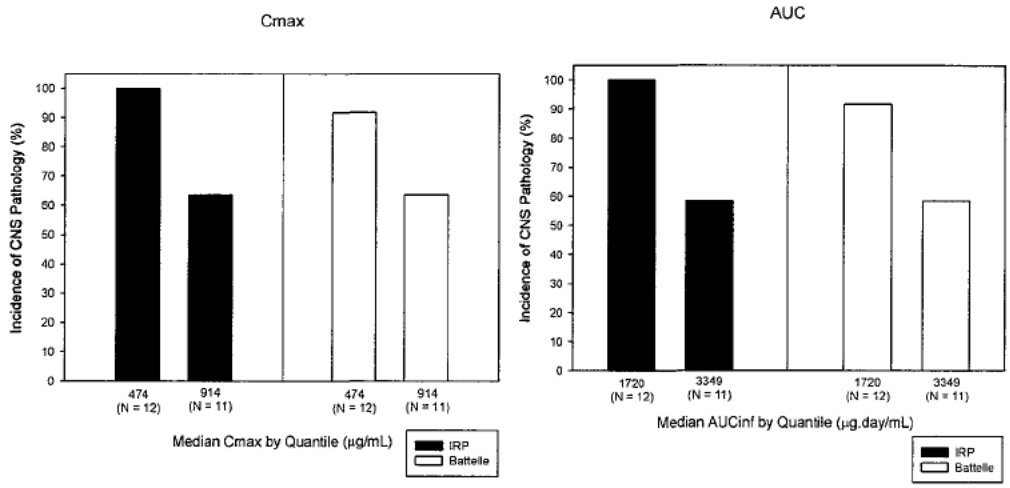


In rabbits, a lower incidence of CNS pathology was observed in higher quantiles of exposure for both Cmax and AUC, as presented in Figure 1.1.4-3. In addition, a lower incidence of bacteria, hemorrhage, and inflammation, but not necrosis, was observed with higher exposures (both Cmax and AUC).

In monkeys, all but one monkey (11/12, 91.7%) that received raxibacumab treatment exhibited CNS findings on necroscopy (based on Battelle assessment; all raxibacumab monkeys that received raxibacumab and died had CNS pathology in the independent pathologist's assessment). In monkeys, a lower incidence of hemorrhage and inflammation with higher exposures (primarily for Cmax) is suggested. Conversely, higher exposure had a higher incidence of bacteria in the CNS for both Cmax and AUC. The incidence of necrosis did not appear to be related to magnitude of exposure in monkeys.

Due to the small numbers of animals that exhibited specific pathology findings, no definitive conclusions about exposure-response for bacteria, hemorrhage, inflammation, or necrosis could be made.

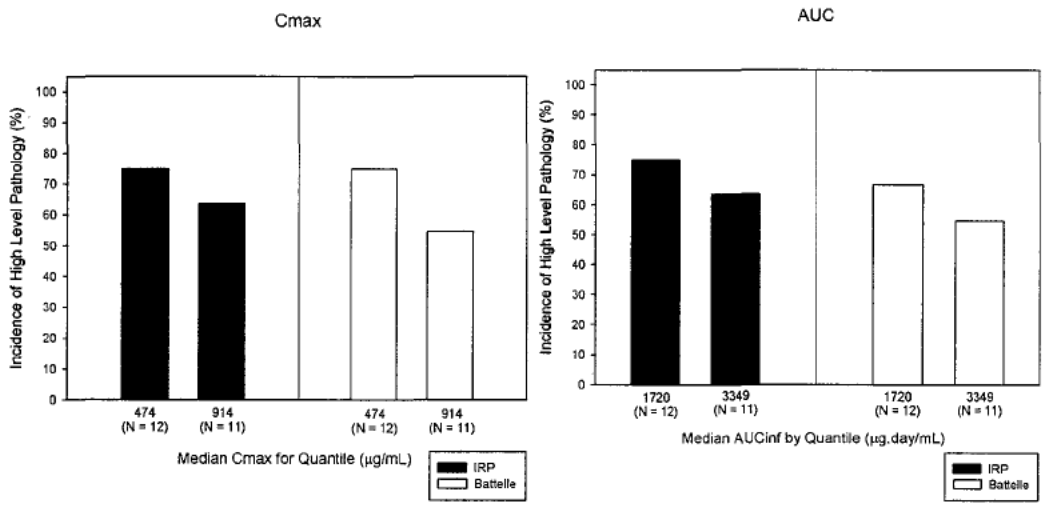
Figure 1.1.4-3 **Comparison of Incidence of CNS Pathology in Rabbits that Died by Quantile of Raxibacumab Exposure**



Cmax quantile ranges: Quantile 1 (342 – 543), Quantile 2 (624 – 1166)
AUCinf quantile ranges: Quantile 1 (1380 – 2144), Quantile 2 (2971 – 3978)
IRP, readings performed by independent pathologist review
Battelle, readings performed by board-certified pathologist at study site

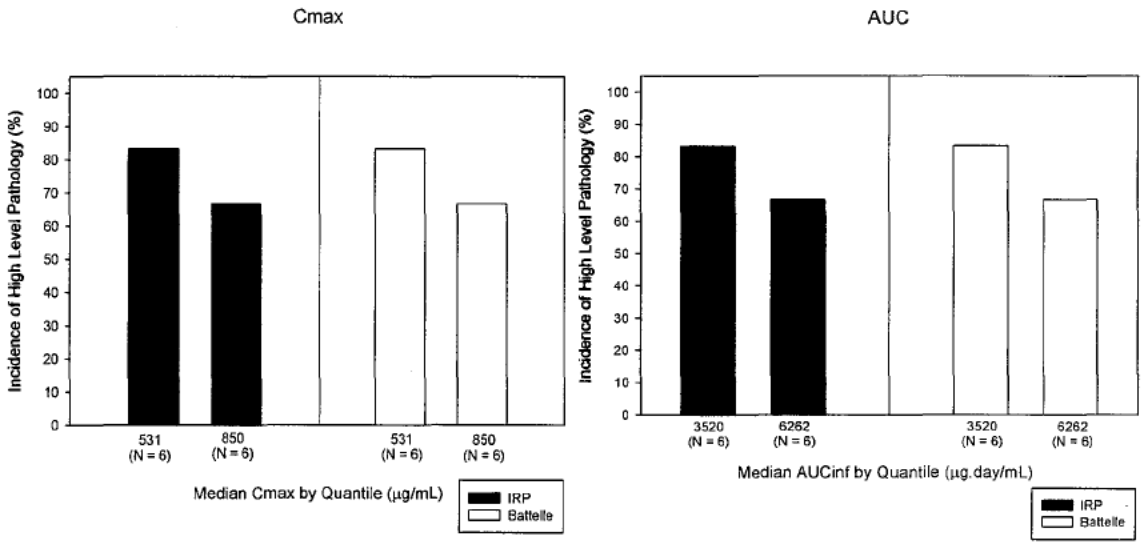
As presented in Figures 1.1.4-3 and 1.1.4-4, a lower incidence of high level CNS pathology was observed with higher quantiles exposures in both rabbits and monkeys. No clear relationship between exposure and response for CNS pathology grade severity can be discerned since these differences between grades were attributed to small numbers of animals.

Figure 1.1.4-3 **Comparison of Incidence of High Level (Grades ≥ 3) CNS Pathology in Rabbits that Died by Quantile of Raxibacumab Exposure**



Cmax quantile ranges: Quantile 1 (342 – 543), Quantile 2 (624 – 1166)
AUCinf quantile ranges: Quantile 1 (1380 – 2144), Quantile 2 (2971 – 3978)
IRP, readings performed by independent pathologist review
Battelle, readings performed by board-certified pathologist at study site

Figure 1.1.4-4 **Comparison of Incidence of High Level (Grades ≥ 3) CNS Pathology in Monkeys that Died by Quantile of Raxibacumab Exposure**



Note: A lower incidence of high level CNS pathology was observed in the higher quantiles of exposure, but this finding can be attributed to one animal (N = 4 in the lower quantile versus N = 5 in the higher quantile).
IRP, readings performed by independent pathologist review
Battelle, readings performed by board-certified pathologist at study site

1.2 Recommendations

1. The proposed weight-based dosing of raxibacumab is acceptable.
2. Dose and concentration response for the endpoint of survival indicates potential to achieve higher response rates at higher doses. If future trials are conducted, higher doses (60 or 80 mg/kg) should be explored along with 40 mg/kg.
3. In any future trials studying higher doses in animals, safety assessments should include an examination of CNS pathology.

1.3 Label Statements

See section 3 (LABELING RECOMMENDATIONS) of the question-based review for clinical pharmacology reviewer recommendations to the proposed labeling.

2 PERTINENT REGULATORY BACKGROUND

Raxibacumab (HGS1021, PA mAb, or ABthrax™) is a fully human monoclonal antibody that specifically binds the Protective Antigen (PA) of *Bacillus anthracis*, thereby preventing its association with the anthrax toxin receptor on host cells. The indication currently being sought for raxibacumab is therapeutic treatment of inhalation anthrax, (b) (4) The current BLA for raxibacumab seeks approval under the "Animal Efficacy Rule" (21 CFR 601, Subpart H, "Approval of Biological Products when Human Efficacy Studies are not Ethical or Feasible"). In accordance with the Animal Rule and because evaluation of new treatment options for inhalational anthrax is not possible in controlled clinical trials in humans for ethical concerns, the efficacy of raxibacumab was evaluated in two animal species, specifically New Zealand white rabbits and cynomolgus monkeys, with symptomatic anthrax disease (Studies Study 682-

G005758 and 724-G005829, respectively). Raxibacumab doses evaluated in both pivotal animal model efficacy studies were 20 and 40 mg/kg single doses administered intravenously. To allow selection of an effective dose in humans for therapeutic treatment of anthrax, the applicant has submitted clinical pharmacology data for raxibacumab in humans and in the two pivotal animal species, rabbits and non-human primates. The pharmacokinetics (PK) and safety of raxibacumab administered as the product proposed for licensure has been evaluated in a Phase 1 dose-ranging study (PAM-NH-01), an antibiotic interaction study with ciprofloxacin (HGS1021-C1064), a repeat dose immunogenicity study (HGS1021-C1069) and a Phase 2/3 safety study (HGS1021-C1063).

3 RESULTS OF APPLICANT'S ANALYSIS

The applicant conducted a population pharmacokinetic (PK) analysis as described in the report entitled "Human Raxibacumab Pharmacokinetics and Rabbit/Monkey PA Kinetic Results for Protocol HGS1021-POP01".

3.1 Objectives

The primary objectives of this analysis were 1) to develop a population PK model of serum raxibacumab concentrations in humans, and 2) to identify and quantify the impact of covariates on raxibacumab PK in humans. The secondary objectives of this analysis were 1) to model the kinetics of PA in untreated, inhalation *Bacillus anthracis* spore-challenged New Zealand white (NZW) rabbits and cynomolgus monkeys, and 2) to estimate the proportion of a human population that would attain protective serum raxibacumab concentrations when administered a single 40 mg/kg intravenous (IV) raxibacumab dose, based on serum/plasma PA concentrations in untreated inhalation *B. anthracis* spore-challenged rabbits and monkeys.

3.2 Methods

Four different analyses were performed to meet the objectives of this study:

1. Human raxibacumab PK were determined from the pooled serum raxibacumab concentration-time data collected in the clinical studies.
2. Serum/plasma PA kinetic parameters were determined for untreated rabbits that died in the therapeutic intervention studies.
3. Serum/plasma PA kinetic parameters were determined for untreated monkeys that died in the therapeutic intervention studies.
4. Human raxibacumab PK results were compared with data for the nonclinical therapeutic intervention studies to confirm selection of the 40 mg/kg IV raxibacumab dose for human use.

3.2.1 Data Sets

Since only the 40 mg/kg IV raxibacumab dose was evaluated in the Phase 3 clinical trial and has been proposed for use as a therapeutic treatment of inhalation anthrax, the population PK analysis characterized the PK of the 40 mg/kg raxibacumab dose. Studies that contributed results to the population analysis of raxibacumab in humans are summarized in Table 3.2.1-1.

For the serum/plasma PA kinetic analyses, results for NZW rabbits and cynomolgus monkeys were analyzed separately. Since animals administered either antibiotics or raxibacumab (as a therapeutic treatment) have been shown to have serum/plasma PA concentration-time profiles that differed from those for control group or untreated animals that died, only control group

animals that died were included in the dataset for analysis. Studies that contributed results to the population analyses for serum/plasma PA kinetics in rabbits and monkeys are summarized in Tables 3.2.1-2 and 3.2.1-3. Analysis data sets used in the applicant's population analysis are summarized in Table 3.2.1-4.

Table 3.2.1-1. Studies used as data sources for the raxibacumab population PK analysis

Study Number and Title	Phase (number of centers)	Study Population	IV dose (mg/kg)	Regimen	Number of evaluable subjects
HGS1021-C1063 A Randomized, Single-Blind, Placebo-Controlled Study to Evaluate the Safety and Tolerability of Raxibacumab (Human Monoclonal Antibody to *Bacillus anthracis* Protective Antigen) in Healthy Subjects	Phase 3 (6 US centers)	Healthy volunteers (54% females; 12% Hispanic, 21% non-white; **≥ 18 years of age, 9% ≥ 65 years of age)**	SD Grp:40	Single dose	217
			DD Grp: 40	Two doses given 14 days apart	23
HGS1021-C1064 An Open-Label Study to Evaluate the Pharmacokinetics and Safety of Raxibacumab (Human Monoclonal Antibody to *Bacillus anthracis* Protective Antigen) Administered in Combination with Ciprofloxacin in Healthy Subjects	Phase 2/3 (3 US centers)	Healthy volunteers (43 males, 45 females; 53 White, 31 Black or African American, 4 other; 18 to 60 years of age)	Grp 1[a]: 40	Single dose	30
			Grp 2: 40	Single dose	28
			Grp 3[b]: 40	Single dose	28
HGS1021-C1069 An Open-Label Study to Evaluate the Immunogenicity and Safety of Raxibacumab (Human Monoclonal Antibody to *Bacillus anthracis* Protective Antigen) Administered in Healthy Subjects	Phase 2/3 (2 US centers)	Healthy volunteers recruited from HGS1021-C1064 subjects (12 males, 8 females; 13 White, 7 Black or African American; 23 to 61 years of age)	40	Two doses given ≥ four months after prior dose (in HGS1021-C1064)	20[c]
				Total number of subjects in population PK analysis	326

SD, single dose; Grp, group; DD, double dose

[a] Raxibacumab was administered concurrent with oral (PO) 500 mg q12 hour x 15 ciprofloxacin doses; the raxibacumab dose was administered immediately following the 11th ciprofloxacin dose.

[b] Raxibacumab was administered concurrent with IV 400 mg q12 hour x 2 ciprofloxacin doses, which were followed by PO 500 mg q12 hour x 13 ciprofloxacin doses; the raxibacumab dose was administered immediately following the 1st IV ciprofloxacin dose.

[c] Number of subjects not included in the total (these subjects were enrolled in the previous study HGS1021-C1064).

Source: Population PK Study Report HGS1021-POP01

Table 3.2.1-2. Studies used as data sources for the rabbit population serum/plasma PA kinetic analyses

Study Number and Title	Target Spore Challenge ($\times LD_{50}$)	Animal Characteristics	Treatment Criteria[1]	Treatment	Number of Animals[2]
615-N104504 Exploratory Study to Evaluate Markers of Disease Course of *Bacillus anthracis* in New Zealand White Rabbits	200	4 males 4 females	No treatment	None	8
682-G005758 Evaluation of Raxibacumab Efficacy as Therapeutic Treatment Against Inhalation Anthrax in the Rabbit Model	200	8 males 8 females	PA toxemia or increased temperature	IV raxibacumab vehicle	16
723-G005835 Evaluation of Levofloxacin for Post-exposure Treatment in the New Zealand White Rabbit Inhalation Anthrax Model	200	3 males	Increased temperature	None	3
781-G923701 Evaluating the Efficacy of Raxibacumab in Combination with Levofloxacin for Post-exposure Treatment in the New Zealand White Rabbit Inhalational Anthrax Model.	200	6 males 6 females	PA toxemia or increased temperature	Intragastric sterile water for injection plus IV raxibacumab vehicle	12
				Total number	39

[1] Treatment was initiated following either first detection of detectable serum/plasma PA concentrations using an electrochemiluminescence-based screening assay, or after the first two consecutive hourly body temperature measurements two or more degrees Fahrenheit higher than the individual's baseline average temperature.
[2] Only control group animals (those not treated with raxibacumab or antibiotic) were included.

Source: Population PK Study Report HGS1021-POP01

Table 3.2.1-3. Studies used as data sources for the monkey population serum/plasma PA kinetic analyses

Study Number and Title	Target Spore Challenge ($\times LD_{50}$)	Animal Characteristics	Treatment Criteria	Treatment	Number of Animals
685-G005762 Natural History Study to Evaluate Criteria for Evidence of Illness due to Inhalation Anthrax in Cynomolgus Macaques	200	5 males 1 female	No treatment	None	6
724-G005829 Evaluation of Raxibacumab Efficacy as Therapeutic Treatment Against Inhalation Anthrax in the Cynomolgus Macaque	200	6 males 6 females	PA toxemia	IV raxibacumab vehicle	12
789-G923702 Evaluation of the Efficacy of Raxibacumab in Combination with Ciprofloxacin for Therapeutic Treatment in the Cynomolgus Monkey Inhalation Anthrax Model. Report No. 789-G923702	200	6 males 6 females	PA toxemia	Intragastric sterile water for injection plus IV raxibacumab vehicle	12
				Total number	30

[1] Treatment was initiated following either first detection of detectable serum PA concentrations using an electrochemiluminescence-based screening assay.
[2] Only control group animals (those not treated with raxibacumab or antibiotic) were included.

Source: Population PK Study Report HGS1021-POP01

Table 3.2.1-4. Analysis Data Sets – Study HGS1021-POP01

Study Number	Name	Link to EDR
Human population PK analysis (Studies HGS1021-C1063, HGS1021-C1064, and HGS1021-C1069)	dat_0001.txt (complete dataset)	\\cbsap58\M\eCTD_Submissions\STN125349\0010\m5\datasets\hgs1021-pop01-pk\human-analysis
	dat_0002.txt (model building)	\\cbsap58\M\eCTD_Submissions\STN125349\0010\m5\datasets\hgs1021-pop01-pk\human-analysis
	dat_0003.txt (model evaluation)	\\cbsap58\M\eCTD_Submissions\STN125349\0010\m5\datasets\hgs1021-pop01-pk\human-analysis
Rabbit PA Analysis (Studies 615-N104504, 682-G005758, 723-G005835, and 781-G923701)	dat-005.csv (complete dataset)	\\cbsap58\M\eCTD_Submissions\STN125349\0010\m5\datasets\hgs1021-pop01-pk\rabbit-analysis
Monkey PA Analysis (Studies 685-G005762, 724-G005829, and 789-G923702)	dat_0001.txt (complete dataset)	\\cbsap58\M\eCTD_Submissions\STN125349\0010\m5\datasets\hgs1021-pop01-pk\human-analysis

3.2.2 Software

For pre- and post-processing of data for NONMEM runs, SAS Release 9.1.3 service pack 4 was used. NONMEM VI was used for the analysis. Selected figures were prepared using GraphPad Prism (Version 4.02). Selected tables and simulations were done using WinNonlin Enterprise (Version 5.0.1; Pharsight Corp.).

3.2.3 Models

3.2.3.1 Raxibacumab Pharmacokinetic Model

PK model building was performed in a stepwise fashion. First, a base model (no covariates) was built by identifying the appropriate structural model and the appropriate error models of intersubject variability. Results from the previously conducted raxibacumab PK studies indicated that human raxibacumab concentration-time profiles exhibit multiphasic disposition, therefore, PK models based on two or more compartments were evaluated. First-order elimination from the central (blood) compartment was assumed. Compartment models were specified using the PREDPP ADVAN and TRAN subroutines in NONMEM. To judge goodness of fit, a decrease in -2LL of ≥ 3.84 was considered as significant, at $\alpha \leq 0.05$. Once the best structural model was identified for PK, the error model (if any) for each parameter was optimized. Additive, proportional, and exponential error models for inter-individual variability (ETA) and residual variability (EPS), as well as the additive with proportional error model for EPS were prospectively identified for potential inclusion in the model.

Plots of individual PK parameter values vs individual values of potential covariates and plots of individual ETA parameter values vs individual values of potential covariates were examined to obtain an overview of potential covariate effects. Covariates included age, gender, race, weight, and clinical laboratory parameters. The data set also included a variable for immunogenicity outcome (overall positive or negative for anti-raxibacumab antibodies [Ab]), as well as a variable for positive/negative status for anti-raxibacumab Ab at different times during the study. Concomitant use of diphenhydramine was also evaluated as a potential covariate.

Model validation was performed on the final PK model. The following aspects of the model were assessed: assumption checking; stability and precision of parameter estimates; model performance (predictive check); and sensitivity analysis. Prospectively, it was intended to evaluate model performance by predicting observations for a test data set using the final model parameter estimates. Diagnostic plots were used to assess the predictive performance of the final model for the test data set. Bias and precision were evaluated using the mean error (ME) and root mean squared error (RMSE), respectively.

3.2.3.2 Serum/Plasma PA Kinetic Models

In monkey and rabbit studies (615-N104504, 682-G005758, 723-G005835, 685-G005762, 724-G005829, and 789-G923702), serum/plasma total PA concentration-time profiles comprised three phases; an initial rapid rise, followed by a 'plateau' period of more slowly increasing or even decreasing levels, and a terminal phase during which levels increased more rapidly again. The first two phases of the profile are consistent with the Gompertz equation that is used to describe biological growth, which displays a lag phase, followed by an exponential growth phase, which then approaches an asymptote, as follows:

$$y = a \cdot e^{-e^{(b-ct)}}$$

where y is the number of bacteria present at time t, and a, b, and c are constants.

A modified Gompertz equation expressed in terms of parameters with biological meaning is as follows:

$$y = A \cdot e^{-e^{\left(\frac{\mu_m \cdot e}{A}(\lambda - t)+1\right)}}$$

where $y=\ln(N/N_0)$ and $A=\ln(N_\infty/N_0)$. N is the number of bacteria present at time t, N_0 is the number of bacteria initially present at time 0, N_∞ is the number of bacteria present at infinite time (the asymptotic phase), μ_m is the maximum specific growth rate, and λ is the lag time.

Many bacteria display a diauxic growth pattern that consists of an initial phase as described by the Gompertz model; following establishment of the asymptotic plateau, a new exponential growth phase begins, followed by a terminal asymptotic phase, as follows:

$$y = w_\alpha \alpha + w_\beta \beta$$

where α and β are the functions describing the two phases of the diauxic growth curve, and w_α and w_β are weights applied to the two functions; the sum of the two weights is constrained to equal 1.

This function was to be applied to the diauxic serum or plasma PA concentration-time profiles, using the modified Gompertz equation of to describe the two phases of the growth curve in the following system of equations:

$$\begin{aligned} y &= w_\alpha \alpha + w_\beta \beta \\ 1 &= w_\alpha + w_\beta \\ \alpha &= A \cdot e^{-e^{\left(\frac{\mu_m \cdot e}{A}(\lambda - t)+1\right)}} \\ \beta &= \left(A \cdot T_A\right) \cdot e^{-e^{\left(\frac{\left(\mu_m \cdot T_{\mu m}\right)e}{\left(A \cdot T_A\right)}(\lambda + T_\lambda - t)+1\right)}} \end{aligned}$$

with variables as defined above, and T_A, $T_{\mu m}$, and T_λ as the factors by which A, μ_m, and λ, respectively, in the β phase differ from those in the α phase.

The structural model prospectively selected for application is described above. The need for the more complex model was to be justified by comparing goodness of fit for the competing models. The form of the model that provides a better fit was to be accepted as the base structural model. To judge goodness of fit, a decrease in -2LL of ≥ 3.84 was to be considered as significant, at $\alpha \leq 0.05$. The selection of error models and covariate model building was performed in a manner analogous to that described for the raxibacumab PK model.

3.2.3.3 Assessment of Protective Raxibacumab Exposure in Humans

The proportion of human subjects for whom raxibacumab exposure can be considered as protective for survival was estimated in the following ways:

- Human raxibacumab PK results were compared with those for *B. anthracis* spore-challenged rabbits and monkeys that survived, following treatment with IV 40 mg/kg raxibacumab alone. The proportion of humans expected to attain a Cmax or AUCinf greater than the lowest Cmax or AUCinf values for a surviving rabbit or monkey were to be determined.
- Human raxibacumab PK results were used to calculate the predicted percent of PA bound by raxibacumab at selected times post-dose, based on the dissociation equilibrium constant (Kd) determined in vitro. For this analysis, the highest observed serum/plasma PA concentrations prior to death in a rabbit or monkey were used as the expected human serum/plasma PA concentration. The percent of PA bound was based on the lower 90% CI bound for the predicted human serum raxibacumab concentration-time profile (thus ensuring 95% of human subjects would have concentrations equal to or greater than that value).
- The percent of PA bound was also assessed in a manner similar to that described above, with the exception that the predicted serum/plasma PA concentration to time of death for individual rabbits and monkeys was estimated using the respective serum/plasma PA kinetic models, which were then used in lieu of the highest observed serum/plasma PA concentrations prior to death for calculation of percent PA bound.

3.3 Results

3.3.1 Raxibacumab Pharmacokinetics in Humans

Data were obtained for 2229 specimens from 322 subjects from the three human clinical trials in which 40 mg/kg IV infusion raxibacumab doses were administered. Most subjects received a single 40 mg/kg raxibacumab dose, but 43 subjects were administered two 40 mg/kg raxibacumab doses given either 14 days apart (in study HGS1021-C1063) or at least 4 months apart (in study HGS1021-C1069).

Demographics and Baseline Characteristics

The demographics and baseline characteristics subjects are summarized in Table 3.3.1-1. Of the 322 subjects, 150 (47%) were male and 172 (53%) were female; 230 (71%) were White, 52 (16%) were Black, and 15 (5%) were Asian; 272 (84%) were non-Hispanic and 50 (16%) were Hispanic; and, 301 (93%) were < 65 years of age and 21 (7%) were ≥ 65 years of age with a range of 18 to 87 years.

Table 3.3.1-1 Summary of Demographics and Baseline Characteristics in Study HGS1021-POP01

	N	Mean	SD	CV%	Median	Minimum	Maximum
Age (y)[1]	322	39	15	39.2	37	18	87
Body Weight (kg)[1]	322	76.9	17.4	22.6	75.6	44.6	155.9
ALT (IU)[1]	322	21	12	55.6	18	7	108
AST (IU)[1]	322	21	8	38.0	19	10	87
Bilirubin (mg/dL)[1]	322	0.5	0.3	62.0	0.4	0.1	2.3
Albumin (g/dL)[1]	322	4.3	0.3	7.2	4.3	3.5	5.1
Total Protein (g/dL)[1]	322	7.1	0.5	7.0	7.0	5.5	8.4
Albumin:Globulin Ratio	322	1.59	0.25	15.5	1.60	0.90	2.65
Serum Creatinine (mg/dL)[1]	322	0.9	0.2	20.7	0.9	0.5	1.8

[1] Value at baseline for the 1st raxibacumab dose administered.

Raxibacumab Dosing

Of the 322 subjects, 279 (87%) were administered a single raxibacumab dose. Twenty-three subjects (7%) were administered 2 raxibacumab doses given 14 days apart. Another 20 subjects (6%) received 2 raxibacumab doses given 4 or more moths apart. For that group, the mean time between doses was 226 days, and ranged from 191 to 279 days. On average, the actual doses administered were close to the nominal 40 mg/kg dose. The exceptions were ID 61, ID 270 and ID 273 who received doses of 13.09, 8.05, and 14.68 mg/kg, respectively; all other doses were close to 40 mg/kg.

Reviewer Comment: Subjects ID 61, ID 270 and ID 273 who received doses of 13.09, 8.05, and 14.68 mg/kg, respectively, were excluded from comparisons of exposure across species for determination of suitability of the proposed 40 mg/kg dose.

Pharmacokinetic Analysis

The raxibacumab data was best fit to a two-compartment model, with exponential error for CL, volume of distribution for the V1, and CLD2, and with proportional error for V2. The final model included the following covariate effects: effects of body weight on CL, V1, CLD2, and V2; effect of sex on V1; and, effect of Black race on CLD2. Diagnostic plots for the final model are displayed in Figures 3.3.1-1 through 3.3.1-3. There was good agreement between observations in the test dataset and predicted values from the final model, as illustrated in Figure 3.3.1-4.

Reviewer Comment: The choice of proportional error model for V2 is questionable. See reviewer's analysis (Appendix 1) using allometric structural model and exponential error model for all parameters. The interindividual variability on intercompartmental clearance and peripheral volume of distribution could not be estimated.

Figure 3.3.1-1 Goodness-of-Fit Plots for the Final Population Model Describing Raxibacumab Pharmacokinetics in Healthy Subjects

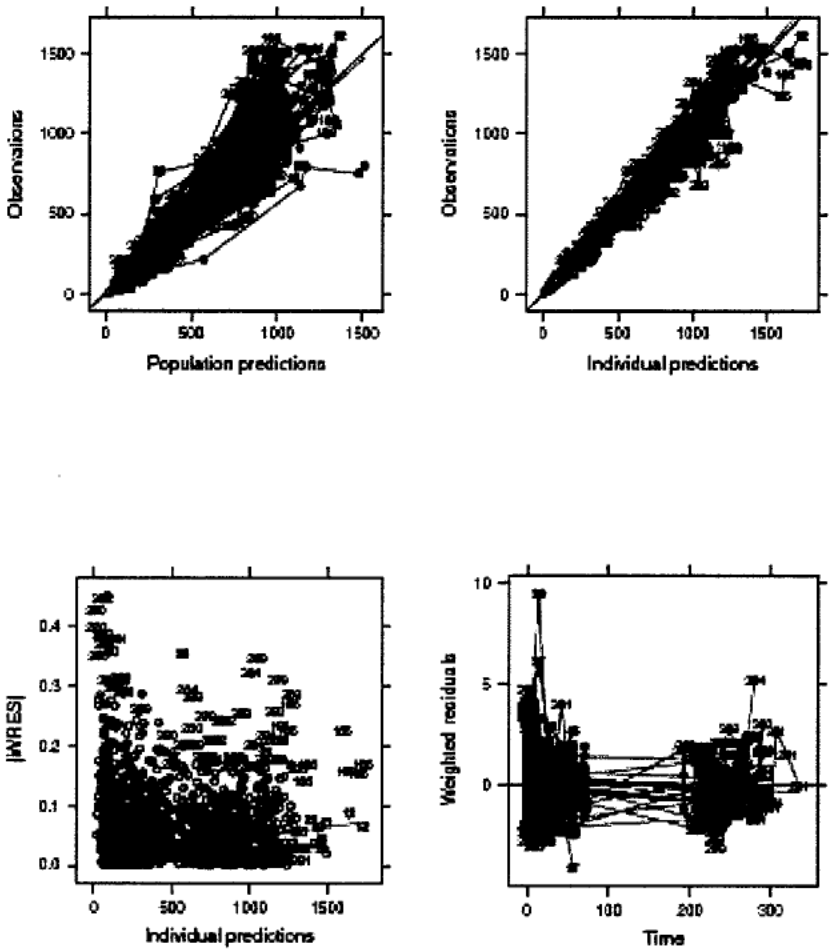
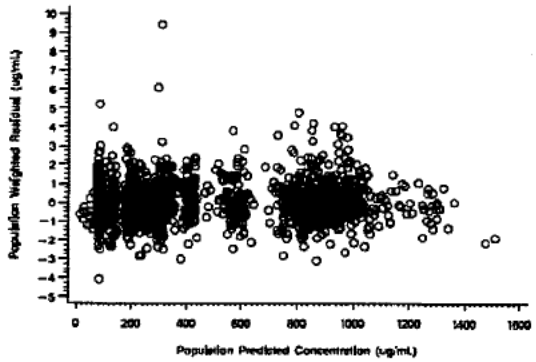


Figure 3.3.1-2 Population and Individual Weighted Residual Plots for the Final Population Model Describing Raxibacumab Pharmacokinetics in Healthy Subjects

Population WRES Plot



Individual WRES Plot

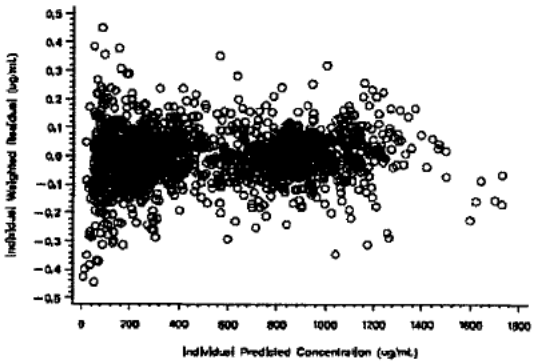
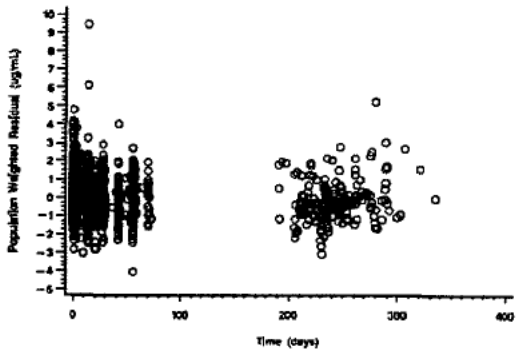


Figure 3.3.1-3 Weighted Residuals versus Time Post Dose for the Final Population Model Describing Raxibacumab Pharmacokinetics in Healthy Subjects

Population WRES Plot



Individual WRES Plot

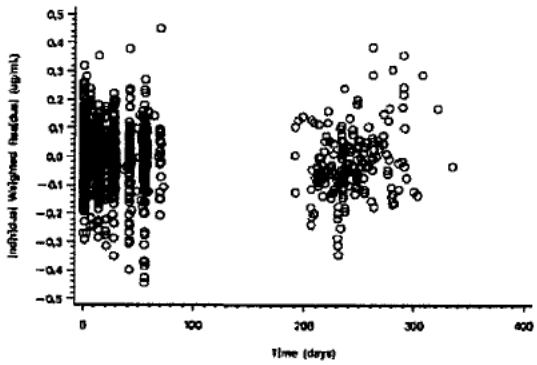
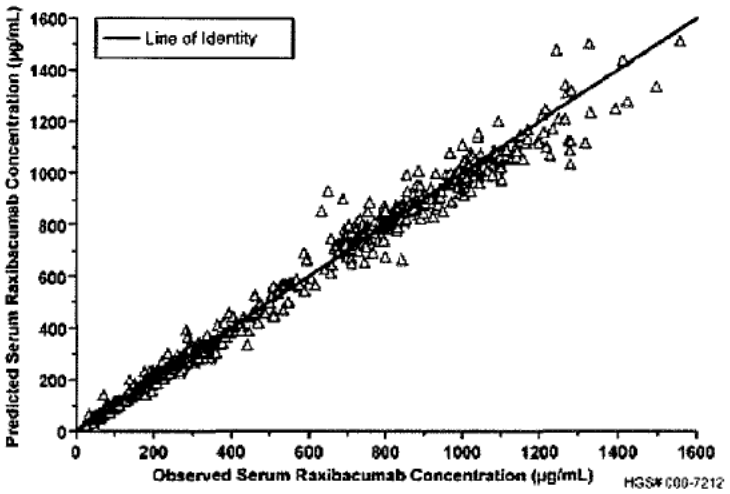


Figure 3.3.1-4 Predicted Versus Observed Individual Serum Concentrations for the Test Dataset for the Final Population Model Describing Raxibacumab Pharmacokinetics in Healthy Subjects



The population model estimates for the primary PK parameters are summarized in Table 3.3.1-2. Body weight was a significant covariate for all PK parameters, with parameter values increasing two- to four-fold across the weight range for the population, as displayed in Figure 3.3.1-5. Although sex and Black race were covariates for V1 and CLD2, respectively, the impact of these covariates was much smaller than that for weight, at 11% and 3%, respectively.

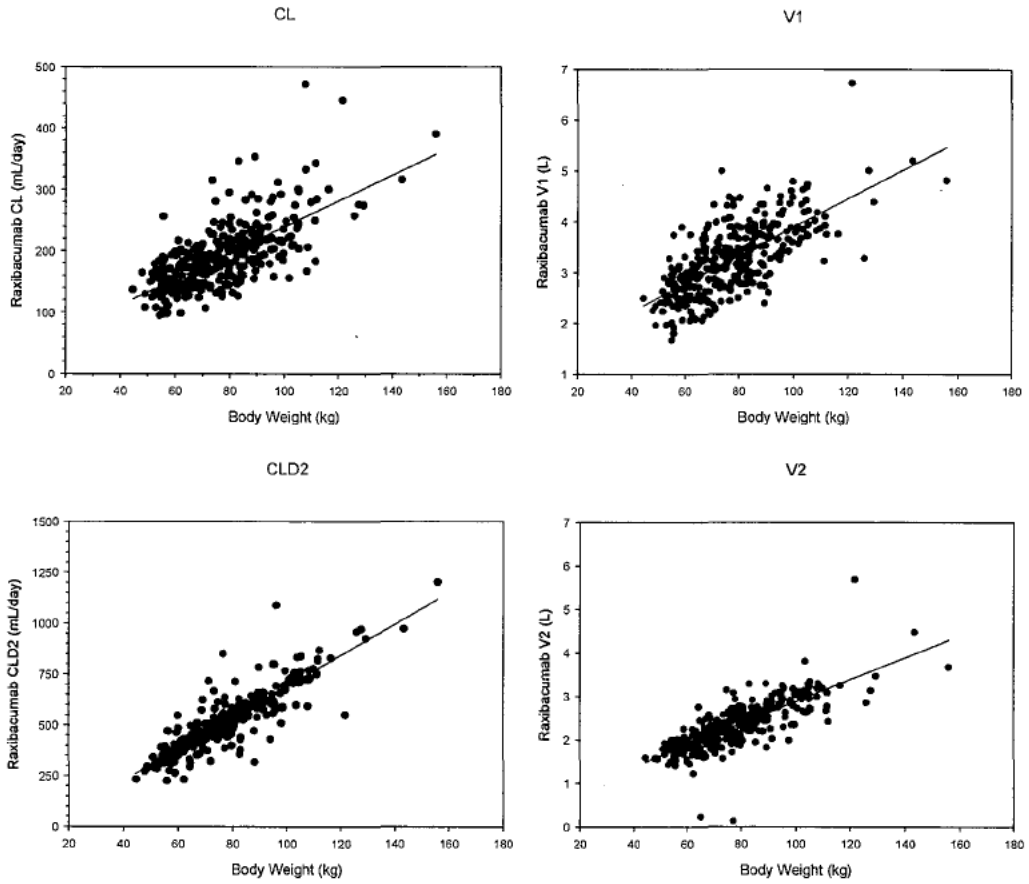
Reviewer Comment: The lack of effect of sex and race on raximacumab is consistent in the reviewer's analysis. The effect of body weight was confirmed on CL, V1 and V2 but not on CLD2.

Table 3.3.1-2 Raxibacumab Pharmacokinetic Parameters

Primary Parameter	Value (RSE [%])	CV% (RSE [%])
V1 (mL)	3312 (1.4)	16.0 (9.8)
Effect of sex on V1	V1 = 3312 × (1+(-0.0992 × sex)) (20.1)	
Males	3312	
Females	2984	
Effect of weight on V1	V1 = 3312 + (26.3 × (weight – 74)) (8.3)	
At 45 kg	2549	
At 62 kg	2996	
At 86 kg	3628	
At 156 kg	5469	
CL (mL/day)	180 (1.2)	20.0 (10.6)
Effect of weight on CL	CL = 180 + (2.02 × (weight – 74)) (6.7)	
At 45 kg	122	
At 62 kg	156	
At 86 kg	205	
At 156 kg	346	
V2 (mL)	2243 (1.9)	21.2 (33.3)
Effect of weight on V2	V2 = 2243 + (26 × (weight – 74)) (7.8)	
At 45 kg	1489	
At 62 kg	1931	
At 86 kg	2555	
At 156 kg	4374	
CLD2 (mL/day)	487 (5.4)	33.4 (35.4)
Effect of race on CLD2	CLD2 = 487 × (1+-0.0328) if race = Black (426.4)	
Black	471	
Non-black	487	
Effect of weight on CLD2	CLD2 = 487 + (7.98 × (weight – 74)) (8.2)	
At 45 kg	256	
At 62 kg	391	
At 86 kg	583	
At 156 kg	1142	

Reviewer Comment: In the reviewer's analysis (see Appendix 1), the parameter estimates are approximately similar to that of reported by the applicant. However, the effect of Sex on V1 and weight on CLD2 cannot be substantiated.

Figure 3.3.1-5 Effect of Body Weight on Raxibacumab PK



Simulations were performed to assess the impact of sex (covariate for V1) and Black race (covariate for CLD2) on secondary (derived) PK parameters. The simulations were performed for hypothetical subjects that weighed the same, so that the substantial covariate effect of weight did not confound the assessment of the impact of sex and Black race on PK. Secondary PK parameters resulting from the simulations are summarized in Table 3.3.1-3. Cmax values (886 and 983 μg/mL for males and females, respectively) were 11% higher in females than in males. Since CL was unaffected by sex or race, AUCinf did not differ among the subgroups. There were small differences in t1/2,α due to sex (< 6%) or due to race (< 3%). Similarly, there were minimal differences in the t1/2,β due to sex (< 6%) or due to race (< 1%). There were minimal differences in MRT or Vss due to sex (< 7%), with no differences due to race. Overall, this assessment indicates that neither sex nor race have a clinically meaningful impact on raxibacumab PK.

Table 3.3.1-3 Effect of Sex and Race on Secondary PK Parameters for Raxibacumab

Secondary Parameters[1]	Non-Black		Black	
	Males	Females	Males	Females
C_{max} (µg/mL)	886	983	886	983
$AUC_{0-\infty}$ (µg·day/mL)	16393	16393	16393	16393
$t_{1/2,\alpha}$ (days)	1.79	1.70	1.84	1.75
$t_{1/2,\beta}$ (days)	22.73	21.56	22.78	21.61
MRT (days)	30.77	28.95	30.77	28.95
V_{ss} (mL/kg)	75.07	70.63	75.07	70.63

Abbreviations: C_{max}, maximum serum drug concentration; $AUC_{0-\infty}$, area under the serum drug concentration-time curve from time 0 to infinite time; $t_{1/2,\alpha}$, elimination half-life for the 1st phase; $t_{1/2,\beta}$, elimination half-life for the 2nd (terminal) phase; MRT, mean residence time; V_{ss}, volume of distribution at steady-state.

[1] Assuming the weight of 74 kg (median weight for model building data set).

Simulations were also performed to assess the impact of weight on secondary PK parameters. The simulations were performed for hypothetical subjects with weights ranging from the minimum to the maximum weight for the subjects, but without accounting for effects of sex or race (ie, in essence, for White males), so that only the effect of weight on PK was assessed. Secondary PK parameters resulting from the simulations are summarized in Table 3.3.1-4. Cmax increased by 61% from 45 to 156 kg, while AUCinf increased by 22% over the weight range. The values of t1/2,α decreased by 42%, whereas t1/2,β and MRT decreased by 15% and 14%, respectively, as weight increased from 45 to 156 kg. Vss also decreased by 30% as weight increased. Overall, these results show that weight has substantial impact on raxibacumab PK.

Table 3.3.1-4 Effect of Weight on Secondary PK Parameters for Raxibacumab

Secondary Parameters	45 kg	62 kg	74 kg	86 kg	156 kg
C_{max} (µg/mL)	702	821	886	940	1128
$AUC_{0-\infty}$ (µg·day/mL)	14760	15873	16393	16807	18018
$t_{1/2,\alpha}$ (days)	2.38	1.95	1.79	1.67	1.39
$t_{1/2,\beta}$ (days)	24.61	23.33	22.73	22.30	20.97
MRT (days)	33.11	31.54	30.77	30.21	28.42
V_{ss} (mL/kg)	89.74	79.47	75.07	71.90	63.10

Abbreviations: C_{max}, maximum serum drug concentration; $AUC_{0-\infty}$, area under the serum drug concentration-time curve from time 0 to infinite time; $t_{1/2,\alpha}$, elimination half-life for the 1st phase; $t_{1/2,\beta}$, elimination half-life for the 2nd (terminal) phase; MRT, mean residence time; V_{ss}, volume of distribution at steady-state.

Reviewer Comment: The finding that weight is a significant covariate on raxibacumb PK is consistent with other monoclonal antibodies demonstrating a strong relationship between weight and PK characteristics. The applicant's simulation results demonstrate that the proposed weight-based dose of raxibacumab 40 mg/kg is appropriate. Dosing raxibacumab based on body weight accounts for increases in clearance seen with higher body weights. The simulated exposures for a body weight of 156 kg are within the range of exposures observed in human subjects receiving raxibacumab 40 mg/kg in clinical trials.

The population model was further evaluated by a cross-study comparison and visual predictive checks. Post hoc PK parameter estimates were compared to non-compartmental PK analysis results from Studies HGS1021-C1064 and HGS1021-C1069, as presented in Table 3.3.1-5. There was good agreement in PK among the studies, indicating that the population PK analysis results are consistent with the results for the prior studies.

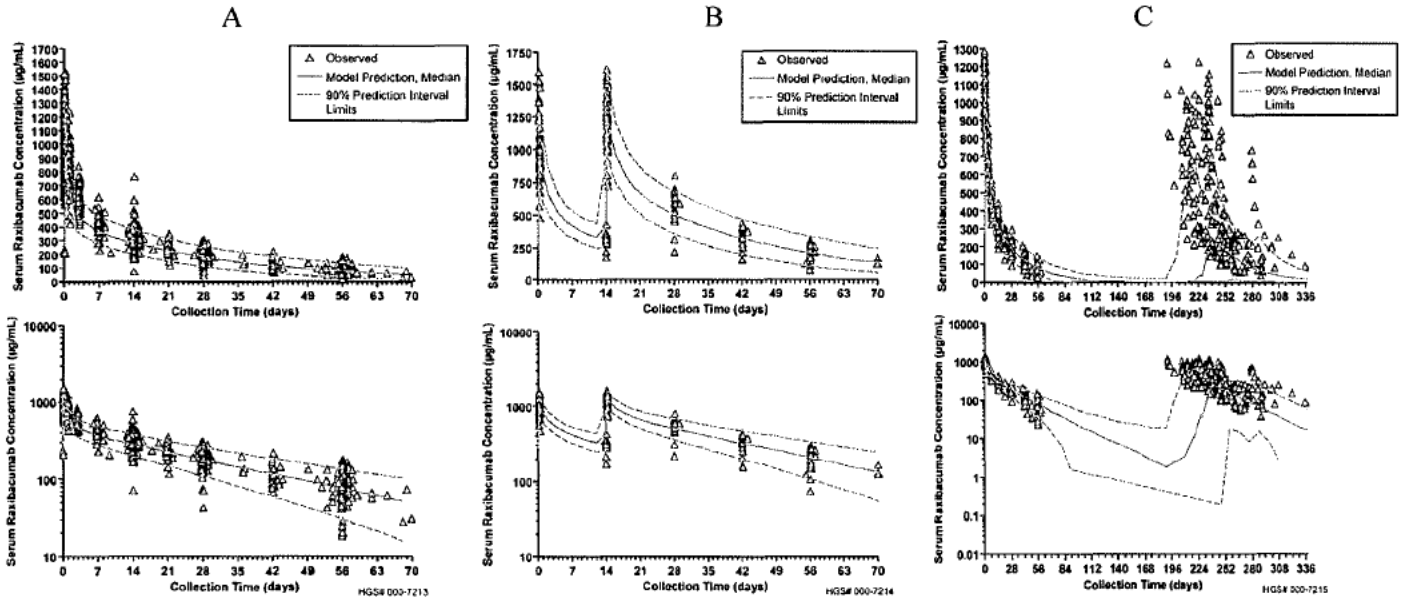
Table 3.3.1-5 Summary of Raxibacumab Pharmacokinetic Parameters Across Studies

Parameter	Population Analysis (N = 322)	HGS1021-C1064 (N = 28)	HGS1021-C1069 (N = 20)
Cmax (μg/mL)	960 (164)	988 (220)	979 (148)
AUC0-∞ (μg·day/mL)	16667 (3198)	15328 (5059)	18239 (6179)
T1/2, β (day)	22.35 (4.04)	20.44 (6.46)	25.68 (11.19)
CL (mL/day/kg)	2.49 (0.49)	2.85 (1.03)	2.37 (0.63)
Vss (mL/kg)	72.92 (10.07)	71.74 (17.36)	75.72 (11.42)

Separate predictive checks were performed by running the final model in simulation mode and generating 1,000 replicate simulations for each subject for subjects that were administered as single raxibacumab dose, subjects that were administered two raxibacumab doses 14 days apart, and subjects that were administered two raxibacumab doses at least four months apart, as displayed in Figure 3.3.1-6. The majority of the observed serum raxibacumab concentrations are within the 90% prediction interval. In addition, the precision of the population PK model parameter estimates was assessed by log likelihood profiling. The 95% CI were relatively narrow, showing that the parameters were satisfactorily precise.

Reviewer Comment: The visual predictive check attempted by the applicant evaluates if the range of concentrations in the original data are obtained in the simulated data. It is less informative towards qualifying model for any use. If attempted, the predictive check should be done by dividing data based on covariates of interest. The reviewer did not attempt to perform visual predictive check as other diagnostics were used for model qualification.

Figure 3.3.1-6 Visual Predictive Checks for Subjects Administered Raxibacumab 40 mg/kg as a Single Dose (A), as Two Doses Given 14 Days Apart (B), and as Two Doses Given at Least Four Months Apart (C)

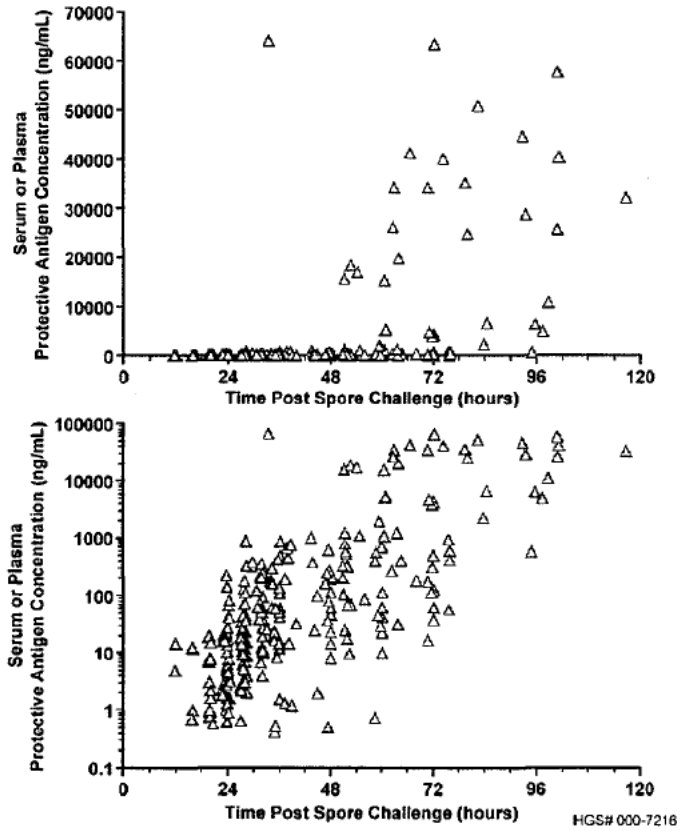


3.3.2 PA Kinetics in Rabbits

The population PA kinetics analysis was performed using data for 344 specimens from 38 rabbits that were obtained in four studies (see Table 3.2.1-2). All animals used for the PA kinetics analysis were not treated with either raxibacumab or antibiotics. Individual serum or plasma PA concentrations in rabbits are presented in Figure 3.3.2-1. PA concentrations were variable among animals but displayed a pattern of increasing concentration as time post challenge increases, with a marked increase prior to death.

Reviewer Comment: The applicant only modeled PA kinetics in animals that were not treated with raxibacumab or antibiotics. An attempt to model PA kinetics in animals that were treated with modalities other than placebo was not made.

Figure 3.3.2-1 Individual Serum or Plasma PA Concentrations in Rabbits Following *Bacillus anthracis* Spore Challenge



A reduced diauxic Gompertz model was used to model the data since no rabbit had evidence of the 2nd plateau phase in their profile. This model consisted of the standard Gompertz model, with a 2nd exponential rising phase that occurs after a 2nd lag phase. This model results in a profile that has an initial lag phase, 1st rising phase, plateau phase, and 2nd rising phase, and is described by the following set of equations:

$$y = e^{\alpha} + \beta$$

$$\alpha = \ln(N_0) + A \cdot e^{-e^{\left(\frac{\mu_{m,2} \cdot e}{A}(\lambda - t)+1\right)}}$$

$$\beta = e^{\mu_{m,2}(t-\lambda_2)-1}, \text{ for } \beta \geq 0, \text{ else } \beta = 0$$

where $\mu_{m,2}$ is the maximum specific growth rate for the 2nd rising phase, and λ_2 is the lag time for the 2nd rising phase. The final rabbit PA model included the following covariate effects: 1) the effect of time to first positive bacteremia by culture on lag time; 2) the effect of body weight on maximum specific growth rate; and 3) the effects of survival time on the natural log of the ratio of the number of bacteria, or the plasma or serum PA concentration, present at infinite time to the number of bacteria, or the plasma or serum PA concentration, present at time 0; on lag time for the 2nd growth phase; and on the maximum specific growth rate in the 2nd phase. Diagnostic plots for the final model are displayed in Figure 3.3.2-2. The population model estimates for the PA kinetic parameters are summarized in Table 3.3.2-1. Post hoc PA kinetic parameter estimates are presented in Table 3.3.2-2. The model was further evaluated by visual predictive checks, as displayed in Figure 3.3.2-3. Separate predictive checks were performed for rabbits with survival times less than 55 h, for rabbits with survival times greater than or equal to 55 h but less than 79 h, and for rabbits with survival times greater than or equal to 79 h. The majority of the observed serum or plasma PA concentrations were within the 90% prediction interval, suggesting that the model describes the data well.

Figure 3.3.2-2 Goodness-of-Fit Plots for the Final Population Model Describing Protective Antigen Kinetics in Rabbits

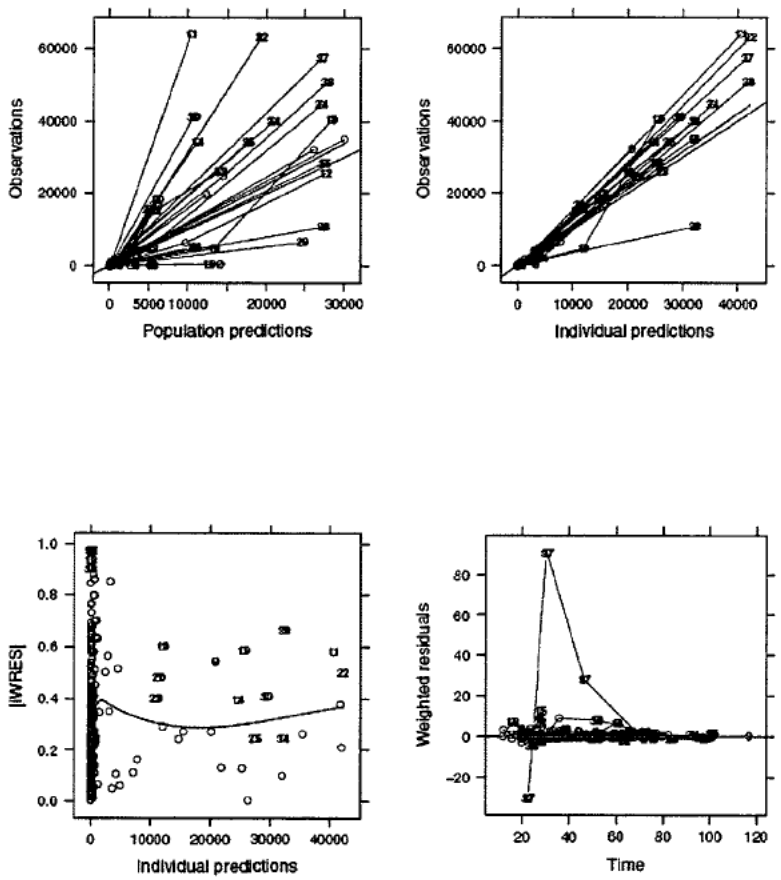


Table 3.3.2-1 Protective Antigen Kinetic Parameters in Rabbits

Primary Parameters	Value (RSE [%])	CV% (RSE [%])
N_0 (ng/mL)	1.43 (68.72)	114.4 (39.0)
λ (h)	22.3 (4.15)	6.3 (59.6)
Effect of time to 1st positive bacteremia by culture (TBAC) on λ (h)	$\lambda(TBAC/24)^{1.09}$	(6.93)
At 19.39 h	17.7	
At 24 h	22.3	
At 94.85 h	99.6	
μ_m (h^{-1})	0.775 (3.41)	-
Effect of weight (WT) on μ_m (h^{-1})	$\mu_m - 0.518$ (WT – 3.2)	(20.67)
At 2.70 kg	1.034	
At 3.24 kg	0.754	
At 3.85 kg	0.438	
A (unitless)	4.34 (18.86)	30.5 (6.9)
Effect of survival time (TSUR) on A	$A(TSUR/72.83)^{-1.47}$	(20.39)
At 33.27 h	13.71	
At 71.25 h	4.49	
At 116.50 h	2.18	
λ_2 (h)	43.4 (3.89)	6.0 (59.58)
Effect of survival time (TSUR) on λ_2 (h)	$\lambda_2(TSUR/72.83)^{0.425}$	(30.08)
At 33.27 h	31.13	
At 71.25 h	43.04	
At 116.50 h	53.06	
$\mu_{m,2}$ (h^{-1})	0.335 (7.74)	-
Effect of survival time (TSUR) on $\mu_{m,2}$ (h^{-1})	$\mu_{m,2}(TSUR / 72.83)^{-1.58}$	(15.10)
At 33.27 h	1.150	
At 71.25 h	0.346	
At 116.50 h	0.160	
Residual variability (CV%)	44.7 (18.6)	

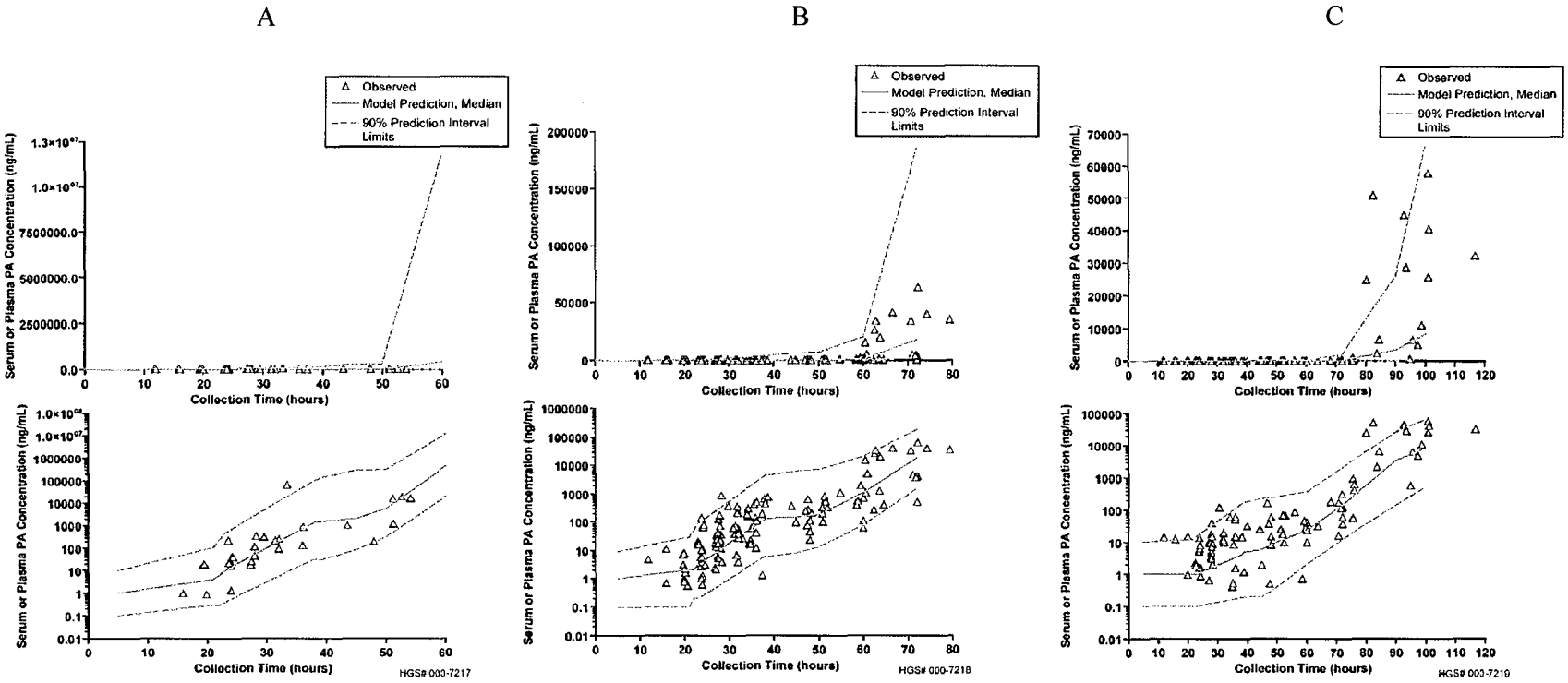
Abbreviations: CV%, coefficient of variation; N_0, PA concentration at time 0; A, natural log of the ratio of the PA concentration in the asymptotic phase to N_0; μ_m, maximum specific growth rate for the 1st phase; λ, lag time for the 1st phase; λ_2, lag time for the 2nd growth phase; $\mu_{m,2}$, maximum specific growth rate for the 2nd phase; TBAC, time to 1st positive bacteremia by culture; WT, body weight; TSUR, survival time; RSE, relative standard error.

Table 3.3.2-2 Post-Hoc Protective Antigen Kinetic Parameters in Rabbits

Parameter	Value Obtained via Population Analysis (n = 39)
N_0 (ng/mL)	3.596 ± 10.493
λ (h)	30.6 ± 19.2
μ_m (h^{-1})	0.753 ± 0.153
A (unitless)	4.77 ± 2.64
λ_2 (h)	44.0 ± 5.4
$\mu_{m,2}$ (h^{-1})	0.359 ± 0.179

Values presented as mean ± SD.

Figure 3.3.2-3 Visual Predictive Checks for Rabbits who Survived Less Than 55 h (A), for at Least 55 h but Less Than 79 h (B), and for at Least 79 h Following Spore Challenge (C)

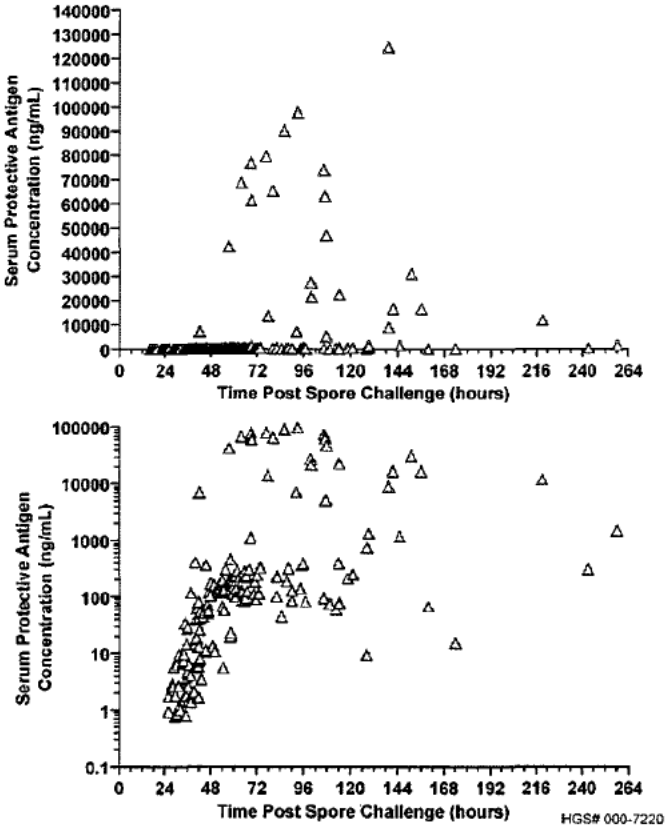


3.3.3 PA Kinetics in Monkeys

The population PA kinetics analysis was performed using data for 265 specimens from 30 monkeys that were obtained in three studies (see Table 3.2.1-3). All animals used for the PA kinetics analysis were not treated with either raxibacumab or antibiotics. Individual serum PA concentrations in rabbits are presented in Figure 3.3.3-1. PA concentrations were variable among animals but displayed a pattern of increasing concentration as time post challenge increases, with a marked increase prior to death.

Reviewer Comment: The applicant only modeled PA kinetics in animals that were not treated with raxibacumab or antibiotics. An attempt to model PA kinetics in animals that were treated with modalities other than placebo was not made.

Figure 3.3.3-1 Individual Serum PA Concentrations in Monkeys Following *Bacillus anthracis* Spore Challenge



The reduced diauxic Gompertz model utilized in the rabbit analysis was used to model the monkey PA data since no monkey had evidence of the 2nd plateau phase in their profile (see Section 3.3.2). The final monkey PA model included the following covariate effects: 1) the effect of time to first positive bacteremia by culture on lag time; and 2) the effects of survival time on lag time for the 2nd growth phase and on the maximum specific growth rate in the 2nd phase. Diagnostic plots for the final model are displayed in Figure 3.3.3-2. The population model estimates for the PA kinetic parameters are summarized in Table 3.3.3-1. Post hoc PA kinetic parameter estimates are presented in Table 3.3.3-2. The model was further evaluated by visual

predictive checks, as displayed in Figure 3.3.3-3. Separate predictive checks were performed for monkeys with survival times less than 90 h, for monkeys with survival times greater than or equal to 90 h but less than 150 h, and for monkeys with survival times greater than or equal to 150 h. The majority of the observed serum PA concentrations were within the 90% prediction interval, suggesting that the model describes the data well.

Figure 3.3.3-2 Goodness-of-Fit Plots for the Final Population Model Describing Protective Antigen Kinetics in Monkeys

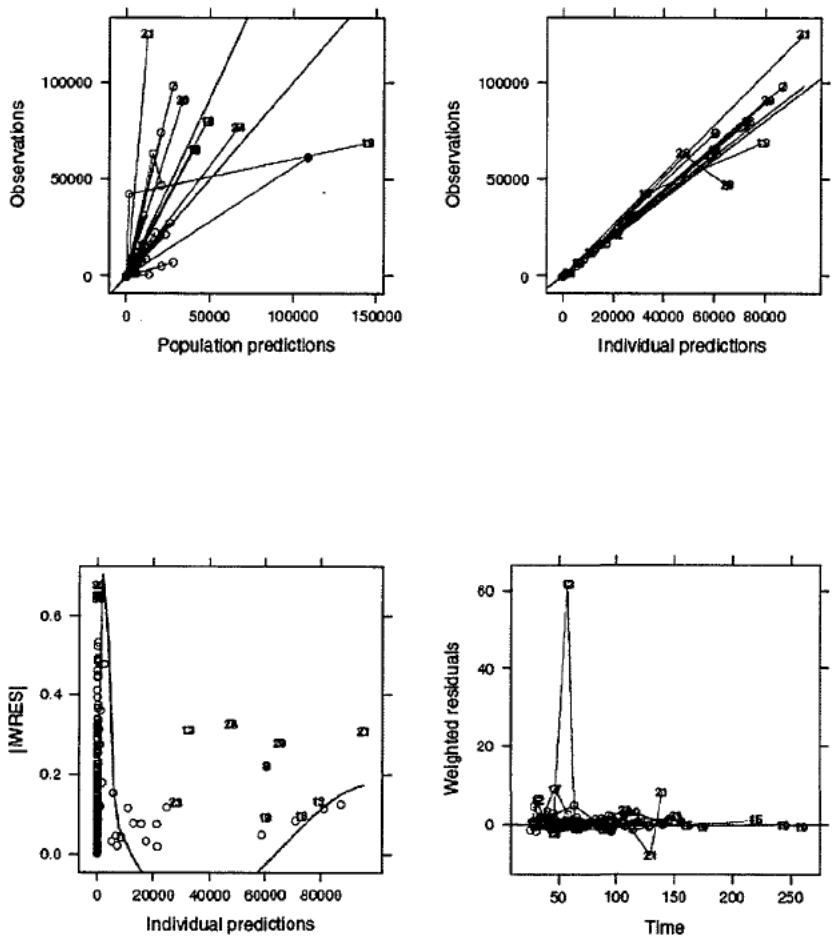


Table 3.3.3-1 Protective Antigen Kinetic Parameters in Monkeys

Parameters	Value (RSE [%])	CV% (RSE [%])
N_0 (ng/mL)	1.21 (16.5)	23.5 (126.8)
λ (h)	35.0 (1.8)	7.2 (36.9)
Effect of time to 1st positive bacteremia by culture (TBAC) on λ (h)	$\lambda + 0.8779(TBAC - 35.72)$	(6.4)
At 26.00 h	26.5	
At 35.72 h	35.0	
At 128.14 h	116.2	
μ_m (h^{-1})	0.515 (9.0)	41.4 (59.0)
A (unitless)	4.94 (3.7)	20.3 (74.3)
λ_2 (h)	71.3 (2.1)	0.001 (4142.6)
Effect of survival time (TSUR) on λ_2 (h)	$\lambda_2 + 0.7979(TSUR - 93)$	(3.6)
At 46.20 h	34.0	
At 92.79 h	71.2	
At 257.89 h	202.9	
$\mu_{m,2}$ (h^{-1})	0.474 (8.1)	11.5 (55.2)
Effect of survival time (TSUR) on $\mu_{m,2}$ (h^{-1})	$\mu_{m,2}(TSUR / 93)^{-1.11478}$	(10.1)
At 46.20 h	1.034	
At 92.79 h	0.475	
At 257.89 h	0.152	
Residual variability (CV%)	28.6 (21.9)	

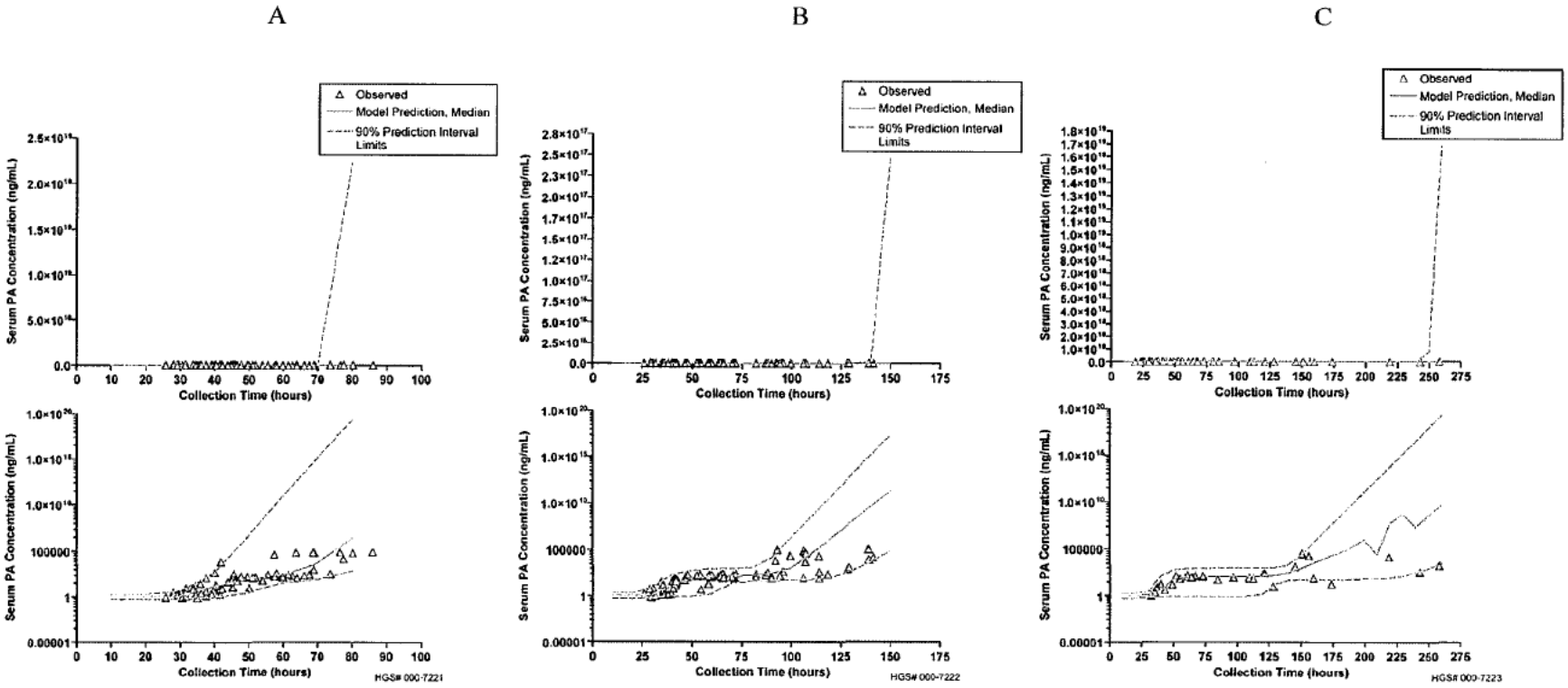
Abbreviations: CV%, coefficient of variation; N_0, PA concentration at time 0; A, natural log of the ratio of the PA concentration in the asymptotic phase to N_0; μ_m, maximum specific growth rate for the 1st phase; λ, lag time for the 1st phase; λ_2, lag time for the 2nd growth phase; $\mu_{m,2}$, maximum specific growth rate for the 2nd phase; TBAC, time to 1st positive bacteremia by culture; TSUR, survival time; RSE, relative standard error.

Table 3.3.3-2 Post-Hoc Protective Antigen Kinetic Parameters in Monkeys

Parameter	Value Obtained via Population Analysis (n = 30)
N_0 (ng/mL)	1.19 ± 0.14
λ (h)	39.3 ± 17.3
μ_m (h^{-1})	0.558 ± 0.174
A (unitless)	5.07 ± 1.14
λ_2 (h)	81.4 ± 36.9
$\mu_{m,2}$ (h^{-1})	0.448 ± 0.196

Values presented as mean ± SD.

Figure 3.3.3-3 Visual Predictive Checks for Monkeys who Survived Less Than 90 h (A), for at Least 90 h but Less Than 150 h (B), and for at Least 150 h Following Spore Challenge (C)

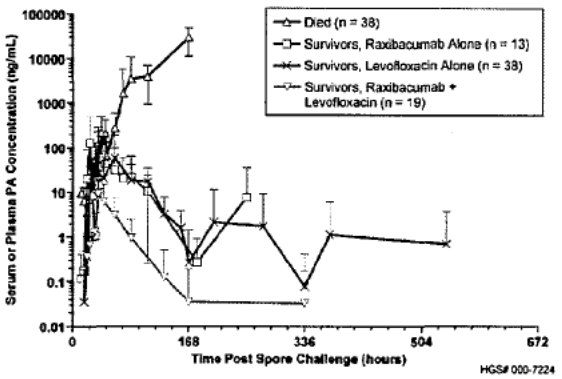


3.3.4 Assessment of Protective Raxibacumab Exposure in Humans

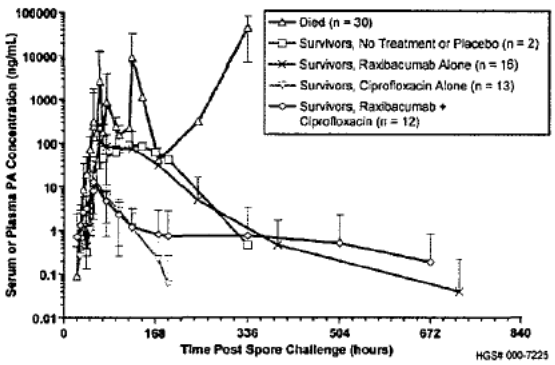
The applicant defined two characteristics of an effective human raxibacumab dose based on PK and PA findings from the animals studies, as follows: 1) a sufficiently high Cmax, and 2) at least 28 days duration of protective serum raxibacumab levels. This was based on a comparison of serum/plasma PA concentration-time profiles for *B. anthracis* inhalation spore-challenged animals that were untreated and died or were administered placebo and died verus animals that survived, as displayed in Figure 3.3.4-1. In surviving animals, PA concentrations reach a peak, but then decrease, whereas in animals that died, PA levels continue to increase until death occurs. In addition, the peak PA levels in animals that died are orders of magnitude higher than the peak levels in the animals that survived. According to the applicant, the data suggest that an efficacious human raxibacumab dose needs to be large enough to rapidly attain serum levels equal to or in excess of those needed to bind systemic PA concentrations, to prevent the mortality associated with PA toxemia. In other words, ***a human raxibacumab dose should attain a sufficiently high Cmax to optimize the likelihood of achieving efficacy***.

Figure 3.3.4-1 Serum/Plasma PA Concentration Profiles in Rabbits and Monkeys

Rabbits



Monkeys



In the rabbit and monkey efficacy studies, animals that survived generally developed measurable anti-PA Ab concentrations and toxin neutralizing Ab (TNA) titers by 14 to 28 days post spore challenge. Thus, the applicant proposes an efficacious human raxibacumab dose ***should maintain protective systemic raxibacumab exposure for at least one month after administration, to allow the innate immune response to PA to develop***.

The applicant assessed these two characteristics for the single 40 mg/kg IV infusion raxibacumab dose in humans by determining the proportion of human subjects for whom raxibacumab exposure can be considered as protective for survival against inhalation anthrax. Comparisons of raxibacumab PK between humans, rabbits and monkeys are presented in Tables 3.3.4-1 and 3.3.4-2. Cmax was similar for a 40 mg/kg dose across species. Given the raxibacumab mechanism of action (binding PA to block lethal effects of toxemia) it is reasonable to expect that attaining similar Cmax should result in similar efficacy in the therapeutic intervention setting. The lowest Cmax and AUC0-∞ in a human subject were 589 µg/mL and 8720 µg·day/mL, respectively, while the lowest observed Cmax and AUC0-∞ for surviving spore-challenged

rabbits were 405 µg/mL and 1411 µg·day/mL, respectively, and were 385 µg/mL and 2499 µg·day/mL, respectively, for surviving spore-challenged monkeys. Thus, a 40 mg/kg dose to humans can be expected to provide exposure associated with survival for virtually all subjects.

Reviewer Comment: The rationale presented by the applicant (with reasonable assumptions) suggests appropriateness of 40 mg/kg. However, dose and concentration response on survival endpoint indicates potential to achieve higher response rate at higher doses. If future trials are conducted, higher doses (60 or 80 mg/kg) should be explored along with 40 mg/kg.

Table 3.3.4-1 Raxibacumab PK in Rabbits and in Healthy Humans (Mean ± SD)

	Healthy Rabbits AB50409.INF.0.016		Spore-Challenged Rabbits That Survived Report 682-G005758		Report 781-G923701	Healthy Humans Population Analysis	
	1 mg/kg (n = 4)	10 mg/kg (n = 4)	20 mg/kg[1] (n = 5)	40 mg/kg[1] (n = 8)	40 mg/kg[2] (n = 18)	40 mg/kg[1] (n = 322)	20 mg/kg[3]
C_{max} (µg/mL)	26 ± 1	276 ± 19	460 ± 51	918 ± 105	929 ± 106	960 ± 164	480 ± 82
$AUC_{0-\infty}$ (µg·day/mL)	174 ± 60	1518 ± 408	1711 ± 323	3504 ± 647	4439 ± 856	16667 ± 3198	8334 ± 1599
$t_{1/2,\alpha}$ (h)	0.40 ± 0.22	0.24 ± 0.05	0.23 ± 0.04	0.26 ± 0.05	0.09 ± 0.01	1.76 ± 0.36	1.76 ± 0.36
$t_{1/2,\beta}$ (h)	8.7 ± 4.4	6.9 ± 2.7	3.86 ± 1.11	4.15 ± 1.22	4.58 ± 0.70	22.35 ± 4.04	22.35 ± 4.04
MRT (h)	12.0 ± 6.0	9.7 ± 3.7	5.41 ± 1.54	5.79 ± 1.69	6.55 ± 0.99	30.09 ± 5.76	30.09 ± 5.76
CL (mL/kg/day)	6.2 ± 1.8	6.9 ± 1.8	11.96 ± 2.36	11.60 ± 2.42	9.35 ± 1.91	2.49 ± 0.49	2.49 ± 0.49
CLD_2 (mL/kg/day)	NA	NA	37.18 ± 1.31	36.73 ± 2.12	86.38 ± 6.74	6.56 ± 0.91	6.56 ± 0.91
V_1 (mL/kg)	39.0 ± 2.1	36.4 ± 2.5	43.56 ± 4.21	43.22 ± 4.53	43.62 ± 5.49	42.86 ± 7.28	42.86 ± 7.28
V_2 (mL/kg)	NA	NA	18.34 ± 4.32	20.70 ± 4.55	16.11 ± 3.16	30.06 ± 4.34	30.06 ± 4.34
V_{ss} (mL/kg)	66.9 ± 9.9	63.2 ± 15.4	61.90 ± 6.45	63.92 ± 7.54	59.73 ± 6.34	72.92 ± 10.07	72.92 ± 10.07

Abbreviations: C_{max}, maximum serum drug concentration; $AUC_{0-\infty}$, area under the serum drug concentration-time curve from time 0 to infinite time; $t_{1/2,\alpha}$, elimination half-life for the 1st phase; $t_{1/2,\beta}$, elimination half-life for the 2nd (terminal) phase; MRT, mean residence time; CL, clearance; CLD_2, intercompartmental clearance; V_1, volume of distribution for the central compartment; V_2, volume of distribution for the peripheral compartment; V_{ss}, volume of distribution at steady-state; NA, not available.

[1] Based on individual post hoc estimates.

[2] Based on individual post hoc estimates; raxibacumab was administered with levofloxacin.

[3] Extrapolated values, assuming linear PK.

Table 3.3.4-2 Raxibacumab PK in Monkeys and in Healthy Humans (Mean ± SD)

	Healthy Monkeys AB50409.INF.0.017		Spore-Challenged Monkeys That Survived Report 724-G005829		Report 789-G923702	Healthy Humans Population Analysis	
	1 mg/kg (n = 4)	10 mg/kg (n = 4)	20 mg/kg[1] (n = 7)	40 mg/kg[1] (n = 9)	40 mg/kg[2] (n = 12)	40 mg/kg[1] (n = 322)	20 mg/kg[3]
C_{max} (µg/mL)	29 ± 6	262 ± 30	475 ± 50	1042 ± 177	1067 ± 158	960 ± 164	480 ± 82
$AUC_{0-\infty}$ (µg·day/mL)	267 ± 91	2030 ± 172	3379 ± 655	6544 ± 2400	9903 ± 2279	16667 ± 3198	8334 ± 1599
$t_{1/2,\alpha}$ (h)	1.10 ± 0.84	0.69 ± 0.53	0.69 ± 0.10	0.68 ± 0.14	0.64 ± 0.12	1.76 ± 0.36	1.76 ± 0.36
$t_{1/2,\beta}$ (h)	15.8 ± 4.1	11.8 ± 1.9	10.80 ± 1.79	9.95 ± 2.48	15.27 ± 4.53	22.35 ± 4.04	22.35 ± 4.04
MRT (h)	19.8 ± 4.3	15.8 ± 1.7	14.38 ± 2.64	13.06 ± 3.53	20.78 ± 6.33	30.09 ± 5.76	30.09 ± 5.76
CL (mL/kg/day)	4.1 ± 1.4	5.0 ± 0.4	6.09 ± 1.15	6.64 ± 2.00	4.25 ± 1.05	2.49 ± 0.49	2.49 ± 0.49
CLD_2 (mL/kg/day)	NA	NA	19.95 ± 3.11	19.03 ± 2.97	21.34 ± 2.98	6.56 ± 0.91	6.56 ± 0.91
V_1 (mL/kg)	36.0 ± 7.8	38.6 ± 4.3	42.41 ± 4.81	38.80 ± 6.05	38.18 ± 5.14	42.86 ± 7.28	42.86 ± 7.28
V_2 (mL/kg)	NA	NA	43.04 ± 2.60	42.07 ± 5.83	44.88 ± 7.58	30.06 ± 4.34	30.06 ± 4.34
V_{ss} (mL/kg)	78.8 ± 23.7	78.0 ± 8.3	85.45 ± 6.90	80.87 ± 10.40	83.06 ± 10.08	72.92 ± 10.07	72.92 ± 10.07

Abbreviations: C_{max}, maximum serum drug concentration; $AUC_{0-\infty}$, area under the serum drug concentration-time curve from time 0 to infinite time; $t_{1/2,\alpha}$, elimination half-life for the 1st phase; $t_{1/2,\beta}$, elimination half-life for the 2nd (terminal) phase; MRT, mean residence time; CL, clearance; CLD_2, intercompartmental clearance; V_1, volume of distribution for the central compartment; V_2, volume of distribution for the peripheral compartment; V_{ss}, volume of distribution at steady-state; NA, not available.

[1] Based on individual post hoc estimates.

[2] Based on individual post hoc estimates; raxibacumab was administered with ciprofloxacin.

[3] Extrapolated values, assuming linear PK.

Based on the median human serum raxibacumab concentration-time profile and lower 90% prediction interval bound profile for a single 40 mg/kg IV raxibacumab dose, the percentage of PA that could be bound were calculated and are summarized in Table 3.3.4-3. A single IV 40 mg/kg raxibacumab dose in humans can be expected to produce serum drug concentrations high enough to bind at least 99% of serum PA for at least 42 days post-dose in at least 95% of the

subjects. Greater than 99% of PA would be bound following a 20 mg/kg dose; however, that level of binding would only be maintained for 28 days.

Table 3.3.4-3 Serum Raxibacumab Concentrations in Humans and PA Binding

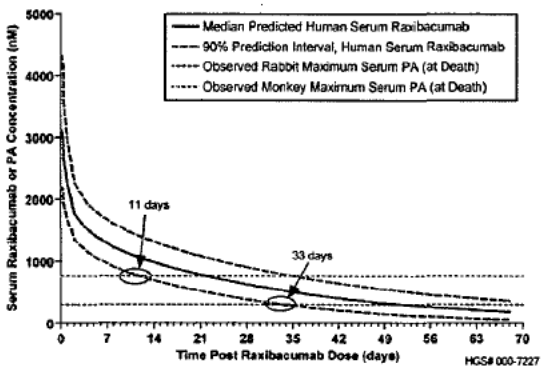
	Serum Raxibacumab Concentrations at:											
	20 mg/kg						40 mg/kg					
	Median			Lower 90% PI			Median			Lower 90% PI		
Time (Days)	µg/mL	nM	% Bound for PA	µg/mL	nM	% Bound for PA	µg/mL	nM	% Bound for PA	µg/mL	nM	% Bound for PA
End of Infusion	452	3115	99.9	317	2185	99.9	903	6230	100.0	634	4370	99.9
1	326	2251	99.9	243	1672	99.9	653	4502	99.9	485	3345	99.9
14	142	978	99.7	99	683	99.6	283	1955	99.9	198	1367	99.8
28	90	623	99.6	55	380	99.4	181	1246	99.8	110	759	99.7
42	58	403	99.4	30	203	98.8	117	806	99.7	59	407	99.4
56	38	260	99.1	15	104	97.7	75	520	99.5	30	209	98.8

Note: This table also includes the percentage of PA that would be bound for a single 20 mg/kg dose based on extrapolation from the 40 mg/kg dose results.

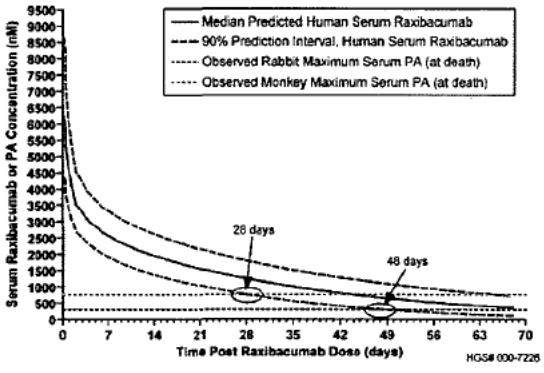
The highest observed serum/plasma PA concentrations observed prior to death in control rabbits and monkeys (ie, not administered raxibacumab or antibiotic) that died were 24752 and 63096 ng/mL (298 and 760 nM), respectively. The median and 90% prediction interval serum raxibacumab profiles for a 20 and 40 mg/kg single IV dose (expressed as nM concentrations) overlaid with the expected highest PA concentrations to be encountered (expressed as nM) is presented in Figure 3.3.4-2. Following a 40 mg/kg dose, serum raxibacumab levels are equimolar to or greater than the highest expected PA levels for 28 or 48 days, using PA levels from monkeys and rabbits, respectively. In contrast, following a 20 mg/kg dose, serum raxibacumab levels were equimolar to or greater than the highest expected PA levels for 11 to 33 days.

Figure 3.3.4-2 Median and 90% Prediction Interval Serum Raxibacumab Profiles for a 20 and 40 mg/kg Single IV Dose Versus the Highest Expected PA Concentrations to be Encountered

20 mg/kg



40 mg/kg



3.3.5 Applicant's Conclusions

- Human serum raxibacumab concentrations best fit a two-compartement open model with 1st-order elimination from the central compartment, with values for CL of 180 mL/day, 3312 mL for V1, 487 mL/day for CLD2, and 2243 mL for V2. Inter-individual variability was less than 33% for CL, V1, CLD2, and V2.
- Body weight was a significant covariate for CL, V1, CLD2, and V2, with parameter values increasing as weight increases. Although sex and Black race were covariates for V1 and CLD2, respectively, the impact of these covariates is small and not likely to be clinically meaningful.
- Serum/plasma PA concentration-time data in rabbits and monkeys were best described using a diauxic growth model consisting of a 1st phase that is consistent with a Gompertz model, followed by a second exponential growth phase that starts following a lag time.
- After a 40 mg/kg raxibacumab IV dose, all human subjects should attain a Cmax or AUC0-∞ greater than the lowest observed Cmax or AUC0-∞ **in any surviving rabbit** or monkey in any of the nonclinical studies of therapeutic intervention.
- More than 95% of humans administered a 40 mg/kg IV raxibacumab dose can be expected to have serum raxibacumab concentrations that are equimolar to or in excess of the highest observed serum/plasma PA concentration in any animal that died in the therapeutic intervention studies (for 48 and 28 days relative to rabbits and monkeys, respectively).
- A single 40 mg/kg IV raxibacumab dose in humans is adequate to bind at least 99.7% of serum PA for up to 28 days after administration, in at least 95% of subjects.
- Based on comparison of exposure in humans and rabbits and monkeys in the therapeutic intervention studies, a 40 mg/kg IV raxibacumab dose can be considered an efficacious dose for humans in the treatment of symptomatic inhalational anthrax.

3.3.6 Reviewer Assessment of Applicant's Analysis

The applicant's population analysis adequately addressed the primary objectives of developing a population PK model of serum raxibacumab concentrations in humans and identifying and quantifying the impact of covariates on raxibacumab PK in humans. The applicant's conclusions regarding the pharmacokinetics of raxibacumab and the significance of weight as a covariate are appropriate. The finding that weight is a significant covariate on raxibacumb PK is consistent with other monoclonal antibodies demonstrating a strong relationship between weight and PK characteristics. Dosing raxibacumab based on body weight accounts for increases in clearance seen with higher body weights.

The applicant also adequately modeled PA kinetics in untreated, *B. anthracis* spore-challenged rabbits and monkeys, and the applicant's conclusions based on this analysis are acceptable.

The applicant's estimates of the proportion of a human population that would attain protective serum raxibacumab concentrations when administered a single 20 mg/kg or 40 mg/kg IV raxibacumab dose support the proposed clinical dose of raxibacumab 40 mg/kg. The rationale presented by the applicant (with reasonable assumptions) suggests appropriateness of 40 mg/kg. However, dose and concentration response on survival endpoint indicates potential to achieve higher response rate at higher doses. If future trials are done, higher doses (60 or 80 mg/kg) should be explored along with 40 mg/kg.

4 REVIEWER'S ANALYSIS

4.1 Introduction

The applicant has concluded that an efficacious human raxibacumab dose needs to be large enough to rapidly attain serum levels equal to or in excess of those needed to bind systemic PA concentrations, to prevent the mortality associated with PA toxemia. In other words, a human raxibacumab dose should attain a sufficiently high Cmax to optimize the likelihood of achieving efficacy. To evaluate this conclusion, an exposure-response analysis was conducted to determine if a relationship exists between PA concentrations, raxibacumab concentrations (specifically Cmax), and outcome (survival or death) in monkeys and rabbits.

Gross necroscopy and microscopic histopathology findings in the CNS of rabbits that died of anthrax in the pivotal efficacy Study 682-G005758 suggested CNS findings were more prevalent in animals that received raxibacumab and died versus animals that received placebo and died. This prompted the FDA to request additional, independent, blinded microscopic histology review of CNS histology data in the pivotal efficacy trials in both rabbits and monkeys (Studies 682-G005758 and 724-G005829). To evaluate a potential relationship between CNS findings and exposure to raxibacumab, an exposure-response analysis was conducted.

4.2 Objectives

The FDA pharmacometric analysis addressed the following two objectives:

1. To evaluate the relationship between PA concentrations, raxibacumab concentrations, and outcome (survival or death) in monkeys and rabbits, and
2. To explore the relationship between plasma raxibacumab PK and CNS findings (ie meningitis) in animals in the pivotal efficacy studies (682-G005758 and 724-G005829).

4.3 Methods

4.3.1 Data Sets

Analysis data sets used in the reviewer's pharmacometric analysis are summarized in Table 4.3.1-1.

Table 4.3.1-1 FDA Pharmacometric Analysis Data Sets

Analysis	Name	Link
E/R PA, raxibacumab and outcome	ANIMAL PK_outcome_dataset.xls	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Shared Analysis Data\
	PKPAoutcome.xls	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Shared Analysis Data\
E/R for CNS findings	RABBIT_CNS_dataset.xls	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Shared Analysis Data\
	MONKEY_CNS_dataset.xls	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Shared Analysis Data\

4.3.2 Software

Additional pharmacometric analyses were performed using NONMEM VI, SPlus7.0 for Windows, Insightful Corp. (2005), Microsoft Office Excel 2003, Professional Edition, Microsoft, Corp (2003), and SigmaPlot Version 11.0, Systat Software, Inc. (2008).

4.3.3 Analytical Methods

4.3.3.1 Relationship Between Raxibacumab PK and PA

PK and PA data from the pivotal animal efficacy studies conducted in rabbits (Study 682-G005758) and monkeys (Study 724-G005829) were explored graphically and by descriptive statistics for relationships between raxibacumab serum concentrations, raxibacumab PK parameters, and PA concentrations in animals that died versus animals that survived.

4.3.3.2 Relationship Between Raxibacumab PK and CNS Findings

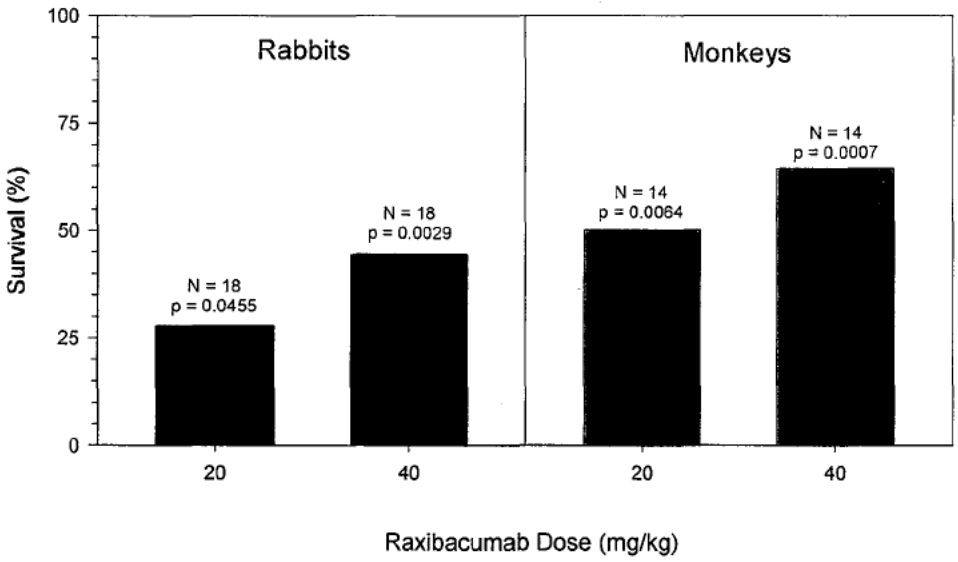
PK and CNS pathology data from the pivotal animal efficacy studies conducted in rabbits (Study 682-G005758) and monkeys (Study 724-G005829) were explored graphically and by descriptive statistics for relationships between raxibacumab exposure and CNS findings in animals that died.

4.3.4 Results

4.3.4.1 Relationship Between Raxibacumab PK, PA and Survival

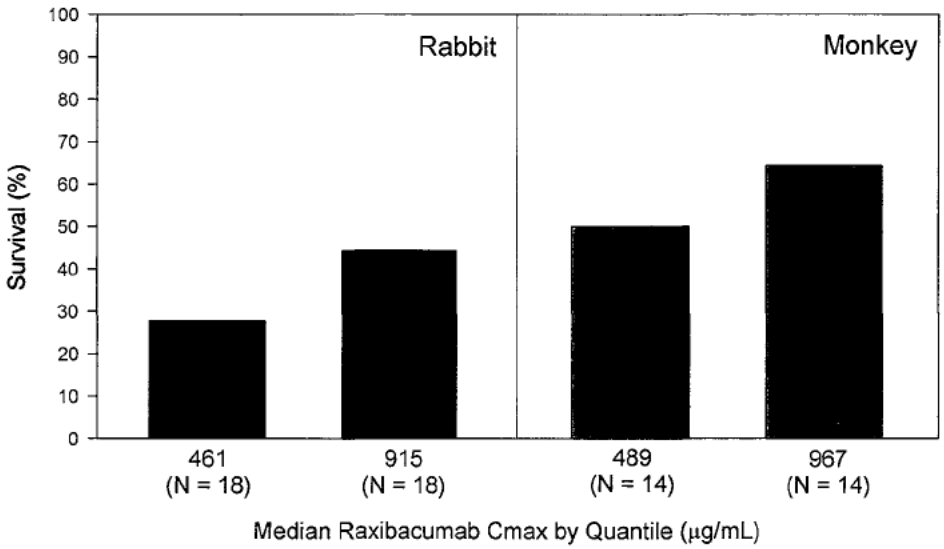
Survival rates by raxibacumab dose in the pivotal animal efficacy studies are summarized in Figure 4.3.4.1-1. Survival rates in both studies exhibited a dose-response for 20 mg/kg and 40 mg/kg doses of raxibacumab IV.

Figure 4.3.4.1-1 Percent Survival in Rabbits and Monkeys Treated with Raxibacumab for Anthrax



As stated previously, the applicant has concluded that a human raxibacumab dose should attain sufficiently high serum levels equal to or in excess of those needed to bind systemic PA concentrations (Cmax) to optimize the likelihood of achieving efficacy. In order to examine the role of Cmax in predicting efficacy, the probabilities of survival across quantiles of raxibacumab Cmax in rabbits and monkeys are presented in Figure 4.3.4.1.1-2. Percents survival in both rabbits and monkeys for Cmax quantiles mirrored findings for dose in that similar increases in survival were observed with increasing quantiles of exposure (Cmax). The data suggest a relationship between raxibacumab dose, concentration, and survival. Although the applicant proposes a sufficiently high Cmax optimizes the likelihood of efficacy, a threshold of Cmax was not identified.

Figure 4.3.4.1-2 **Comparison of Probabilities of Survival by Raxibacumab Cmax Quantile**



To determine a relationship (if any) of PA concentrations, raxibacumab concentration and survival, scatter plots of PA and raxibacumab concentrations over time by survival status in rabbits and monkeys were compared, as displayed in Figures 4.3.4.1-3 and 4.3.4.1-4, respectively. Individual plots of raxibacumab and PA concentrations for all animals over time are presented in Appendices 2 and 3. PA concentrations were highly variable across all animals. As observed previously, in surviving animals, PA concentrations reach a peak, but then decrease, whereas in animals that died, PA levels continue to increase until death occurs. In addition, the peak PA levels in animals that died are orders of magnitude higher than the peak levels in the animals that survived. As stated by the applicant, the overall magnitude of PA concentrations was higher in animals that died versus those that survived. Conversely, the magnitude of raxibacumab concentrations were very similar in animals that died compared to animals that survived. Although PA concentrations appear to decline in surviving animals as raxibacumab is being cleared from the body, a direct exposure/response relationship between serum raxibacumab concentrations and serum/plasma PA concentrations could not be determined due to the following confounding factors:

1. For both analytes, only total serum/plasma concentrations were measured. The time course of binding of PA by raxibacumab could therefore not be described and the relationship between free or bound drug and PA concentrations is unknown.
2. Measurement of PA concentrations is affected by the presence of raxibacumab in serum and plasma. The effect of raxibacumab was assessed by PA and raxibacumab-spiked samples and calculation of the percentage ratio of the measured concentration in these spikes to the measured concentrations of the PA-spiked samples. In rabbits, at concentrations of **raxibacumab at 100 µg/mL** or higher, recovery of PA was decreased by up to 65%. In monkeys, recovery of PA ranged from -17% to +26% of the spiked value **when the concentration of raxibacumab was 500 µg/mL or** higher. Thus, the reported PA values in animals receiving raxibacumab may not accurately reflect actual PA concentrations in those animals.

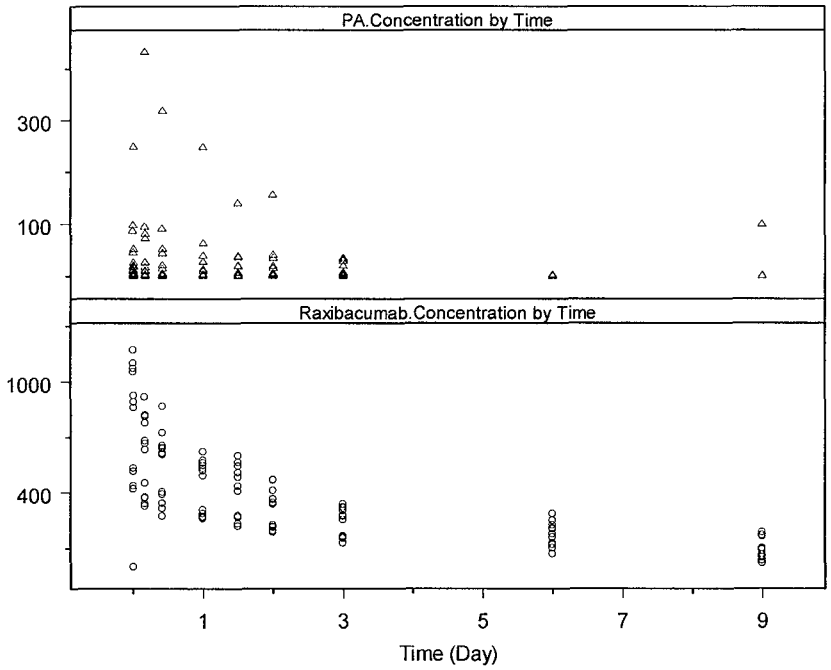
3. Significant inherent intra- and inter-individual variability of PA data exists. In the modeling of PA data from rabbits and monkeys, the variability components of the applicant's kinetic model were poorly defined suggesting the PA kinetic results should be interpreted with caution.

Due to these factors, definition of an exposure-response relationship between raxibacumab concentrations, PA concentrations, and survival is inappropriate given the available data.

In summary, the exposure-response data suggest a relationship between raxibacumab dose, concentration, and survival, but small samples sizes limit conclusions regarding the use of Cmax as a predictor of survival. Definition of an exposure-response relationship between raxibacumab concentrations, PA concentrations, and survival is inappropriate given the available data and limitations mentioned above. The rationale presented by the applicant (with reasonable assumptions) suggests appropriateness of 40 mg/kg. However, dose and concentration response on survival endpoint indicates potential to achieve higher response rate at higher doses. If future trials are conducted, higher doses (60 or 80 mg/kg) should be explored along with 40 mg/kg.

Figure 4.3.4.1-3 Raxibacumab and PA Concentrations in Rabbits (Study 682-G005758)

Rabbits that Survived



Rabbits that Died

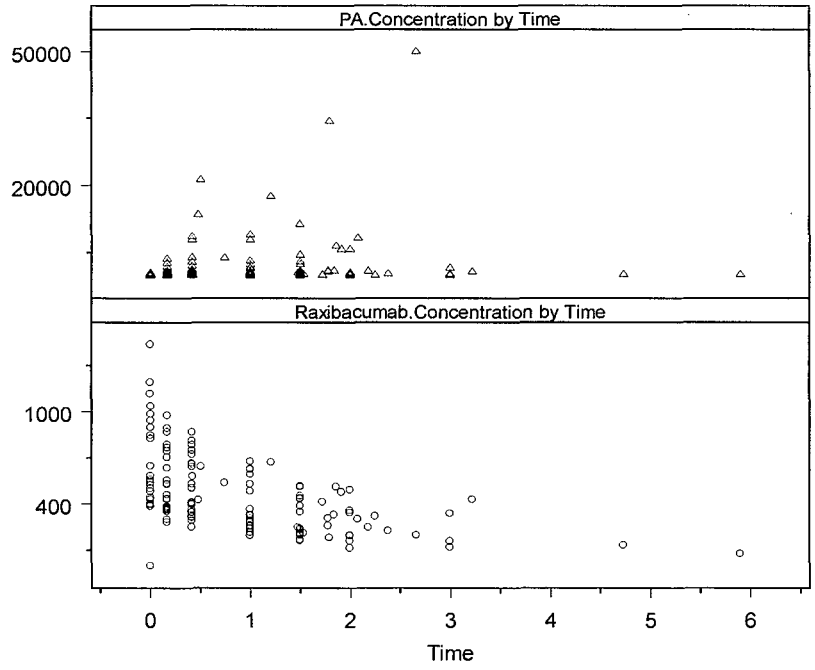
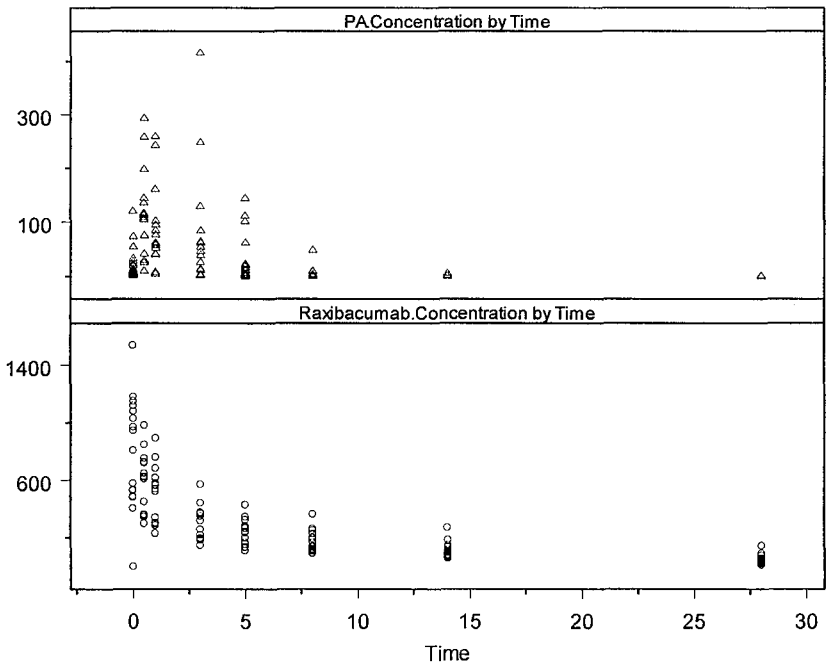
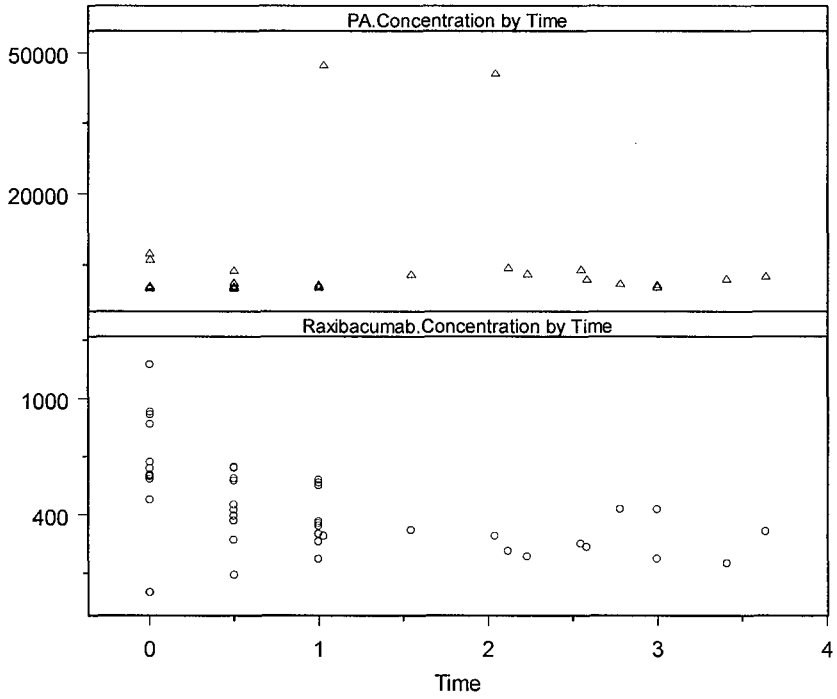


Figure 4.3.4.1-4 **Raxibacumab and PA Concentrations in Monkeys (Study Study 724-G005829)**

Monkeys that Survived



Monkeys that Died



Since an exposure-response relationship between raxibacumab and PA concentrations could not be determined (due to the limitations described above), a reasonable assessment of the inter-relationship between raxibacumab concentrations, PA concentrations and survival for purposes of predicting the acceptability of a proposed dose is a comparison of raxibacumab serum concentrations over time to the concentrations required for 99.0 and 99.9% binding of PA (based on the known mechanism of action and binding kinetics of raxibacumab). Based on in vitro binding kinetics studies, serum concentrations of approximately 40 and 202 µg/mL are required for 99.0 and 99.9% binding of PA, respectively. As presented in Figure 4.3.4.1-6, in human subjects that received 40 mg/kg raxibacumab IV, raxibacumab concentrations remained above 202 µg/mL for 7 days and above 40 µg/mL for 42 days for all human subjects. Thus, in humans a 40 mg/kg dose of raxibacumab would be expected to maintain levels required for virtually complete binding of PA for 7 days.

Despite the theoretical importance of targeting virtually complete binding of PA, the duration of time raxibacumab concentrations remain above the threshold of 202 µg/mL does not appear to impact efficacy, as displayed in Figures 4.3.4.1-7 and 4.3.4.1-8. In both rabbits and monkeys, the amount of time serum concentrations of raxibacumab remained above 202 µg/mL generally did not differ between survivors and non-survivors.

Figure 4.3.4.1-6 **Individual Serum Concentrations of Raxibacumab in Human Subjects Following Administration of 40 mg/kg IV Compared to the Concentrations Required for 99 and 99.9% Binding of PA**

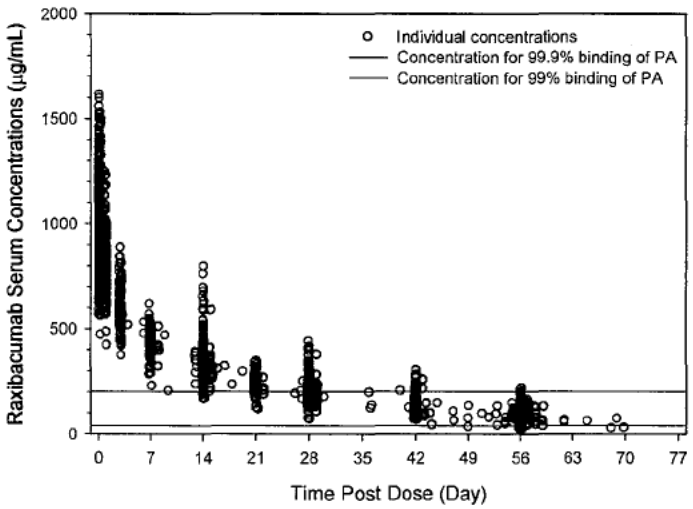


Figure 4.3.4.1-7 **Spaghetti Plot of Serum Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Rabbits**

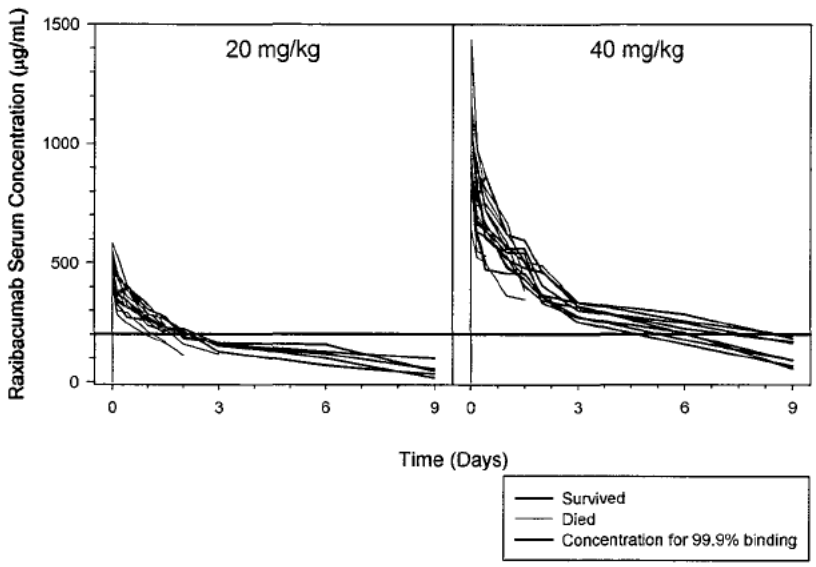
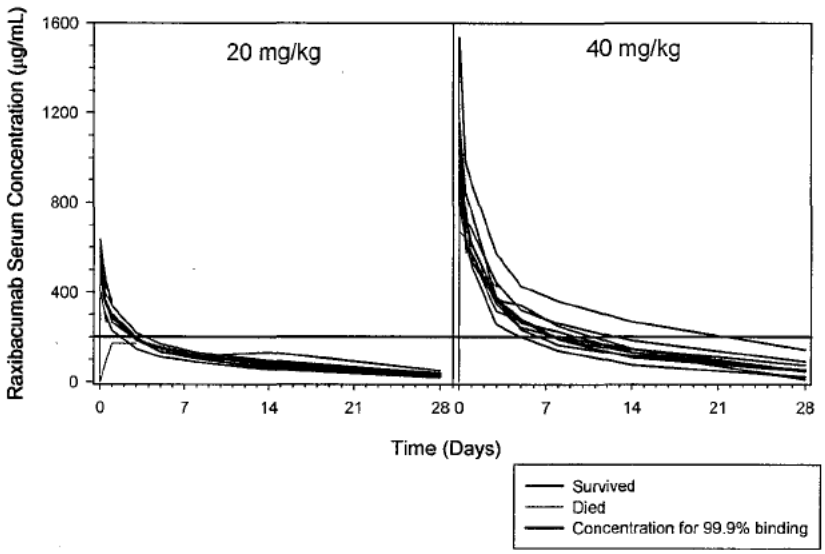


Figure 4.3.4.1-8 **Spaghetti Plot of Serum Concentration-Time Profiles for Raxibacumab Following Single Intravenous Administration of 20 mg/kg and 40 mg/kg Raxibacumab in Spore-Challenged Monkeys**



In summary, an evaluation of the relationship between PA concentrations, raxibacumab concentrations, and outcome (survival or death) in monkeys and rabbits suggests the following:

1. The exposure-response data suggest a relationship between raxibacumab dose, concentration, and survival. Although the applicant proposes a sufficiently high Cmax optimizes the likelihood of efficacy, a threshold of Cmax was not identified.
2. The definition of an exposure-response relationship between raxibacumab and PA concentrations is confounded by the following factors: 1) for both analytes, only total serum/plasma concentrations were measured, 2) the measurement of PA concentrations is affected by the presence of raxibacumab in serum and plasma, and 3) significant inherent intra- and inter-individual pharmcodynamic variability of PA data.
3. Based on in vitro binding kinetics studies, the proposed dose of 40 mg/kg raxibacumab in humans would be expected to maintain levels required for virtually complete binding (99.9%) of PA for 7 days and those required for 99% binding for up to 42 days. Despite the theoretical importance of targeting virtually complete binding of PA, the duration of time raxibacumab concentrations remain above the threshold of 202 µg/mL does not appear to impact efficacy in inhalational anthrax models.

4.3.4.2 Relationship Between Raxibacumab PK and CNS Findings

In the pivotal animal studies, animals that were treated with raxibacumab and died exhibited a higher incidence of histopathologic findings in the brain versus placebo treated animals, as presented in Figure 4.3.4.2-1. Histopathologic findings were also of higher severity in raxibacumab treated animals versus animals receiving placebo treatment, as presented in Figure 4.3.4.2-2. The raxibacumab 20 mg/kg dose group had a higher incidence and severity of CNS findings versus the 40 mg/kg group, suggesting an absence of clear dose-response relationship for brain histopathology.

Figure 4.3.4.2-1 Histopathology Findings by Treatment in Animals that Died

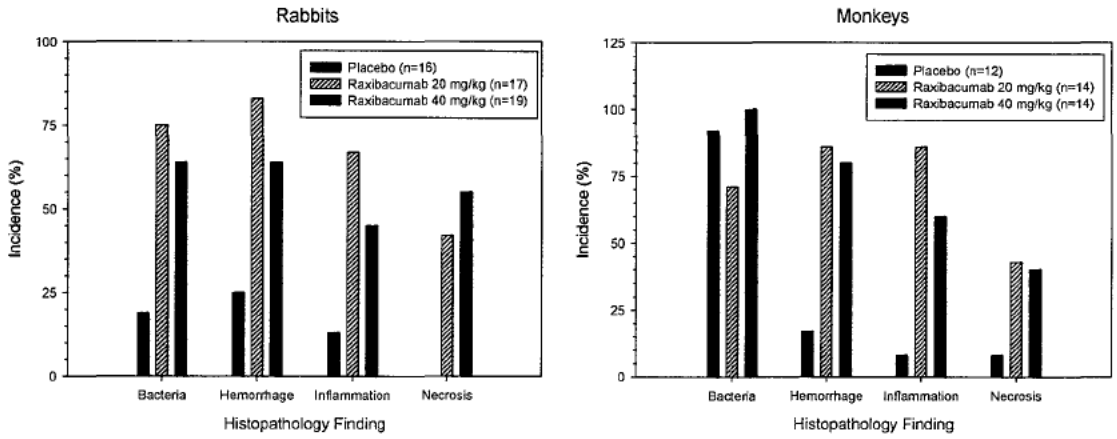
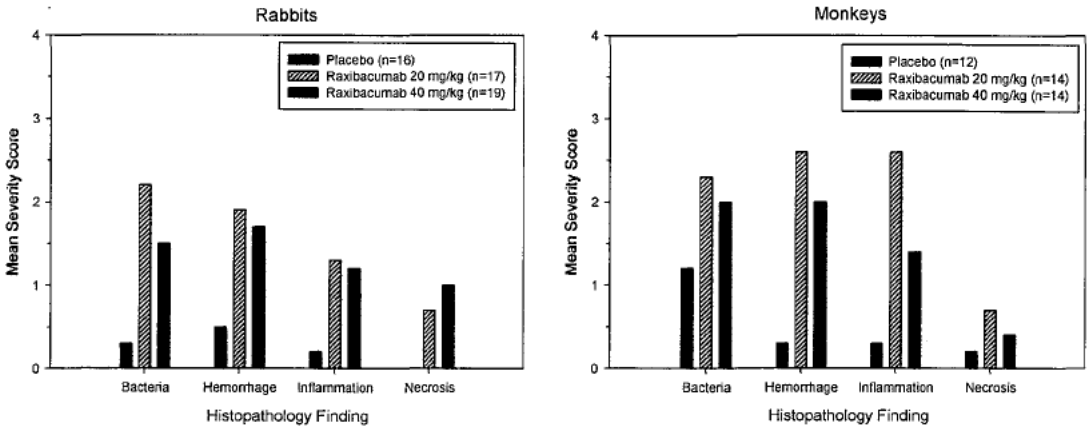


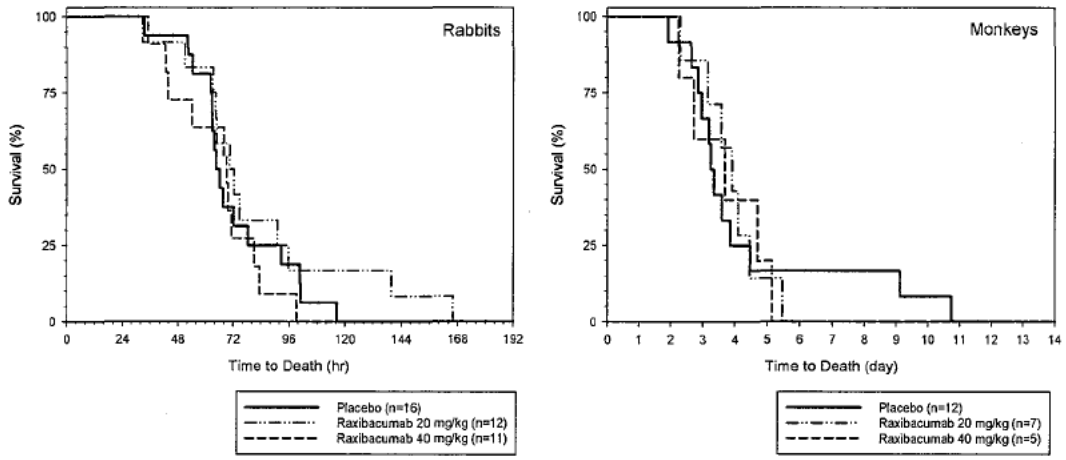
Figure 4.3.4.2-2 **Severity of Histopathology Findings by Treatment in Rabbits and Monkeys that Died**



Grading scale is as follows: Grade 1, minimal representing the least detectable lesion; Grade 2, mild representing an easily discernable lesion unlikely to have biological relevance; Grade 3, moderate representing a change affecting a large area of the represented tissue that had the potential to be of some relevance; and Grade 4, marked representing a lesion that approached maximal.

A potential hypothesis proposed by the applicant to explain the finding that adverse CNS findings were more prevalent in animals that received raxibacumab and died versus animals that received placebo and died is that animals that received raxibacumab survived longer and brain lesions increased over time. Figure 4.3.4.2-3 shows percent survival and survival time by raxibacumab dose for rabbits and monkeys that died in the pivotal studies. The survival plots show that in rabbits that died, placebo animals have comparable survival times versus animals that received 40 mg/kg of raxibacumab. In monkeys that died, animals that received placebo lived longer than those receiving raxibacumab. Thus, the potential for raxibacumab treatment to result in adverse CNS findings is not likely to be related to prolonged survival time, and the applicant's assertion that animals receiving raxibacumab do not die more rapidly than placebo animals is not appropriate.

Figure 4.3.4.2-3 Time to Death in Rabbits and Monkeys That Died



In addition, time to death by exposure quantile was examined to determine if in animals that died, higher raxibacumab exposure led to longer survival times. Figures 4.3.4.2-4 and 4.3.4.2-5 show percent survival and survival time by quantiles of exposure (Cmax and AUC) for rabbits and monkeys that died, respectively. In rabbits that died, animals in the lower Cmax and AUC quantiles appeared to live longer than the other groups and placebo, but in monkeys there was no discernable difference between survival times for the higher and lower quantiles of exposure. The placebo group exhibited a longer survival time compared to the raxibacumab groups. This suggests that in animals that died, the magnitude of raxibacumab exposure did not affect survival time.

Figure 4.3.4.2-4 Comparison of Time to Death in Rabbits by Quantile of Raxibacumab Exposure

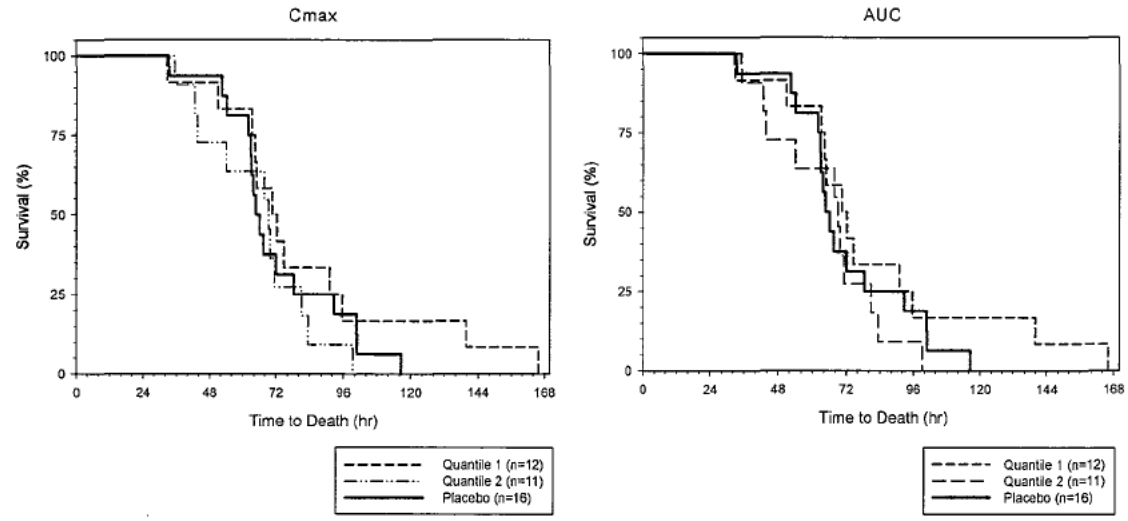
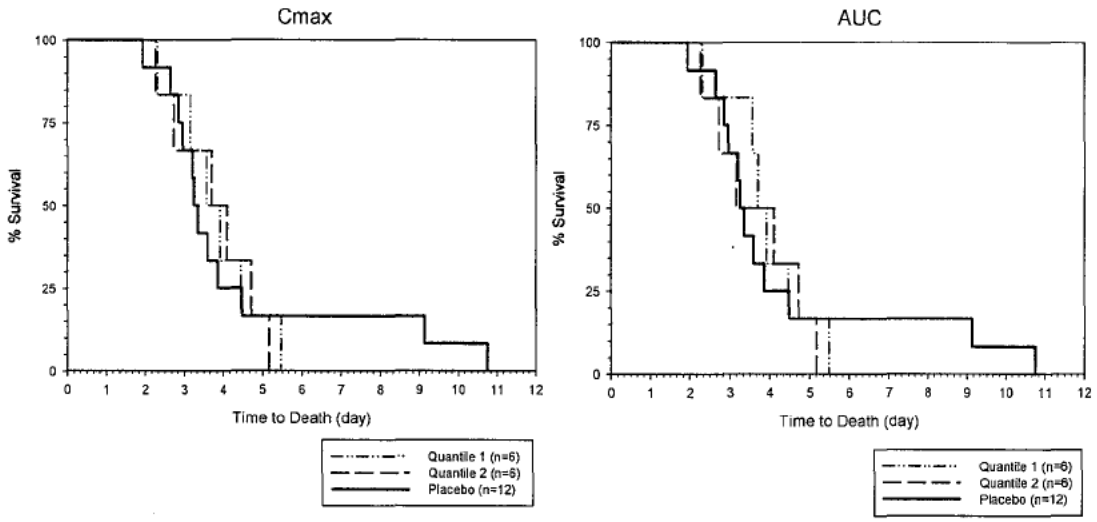
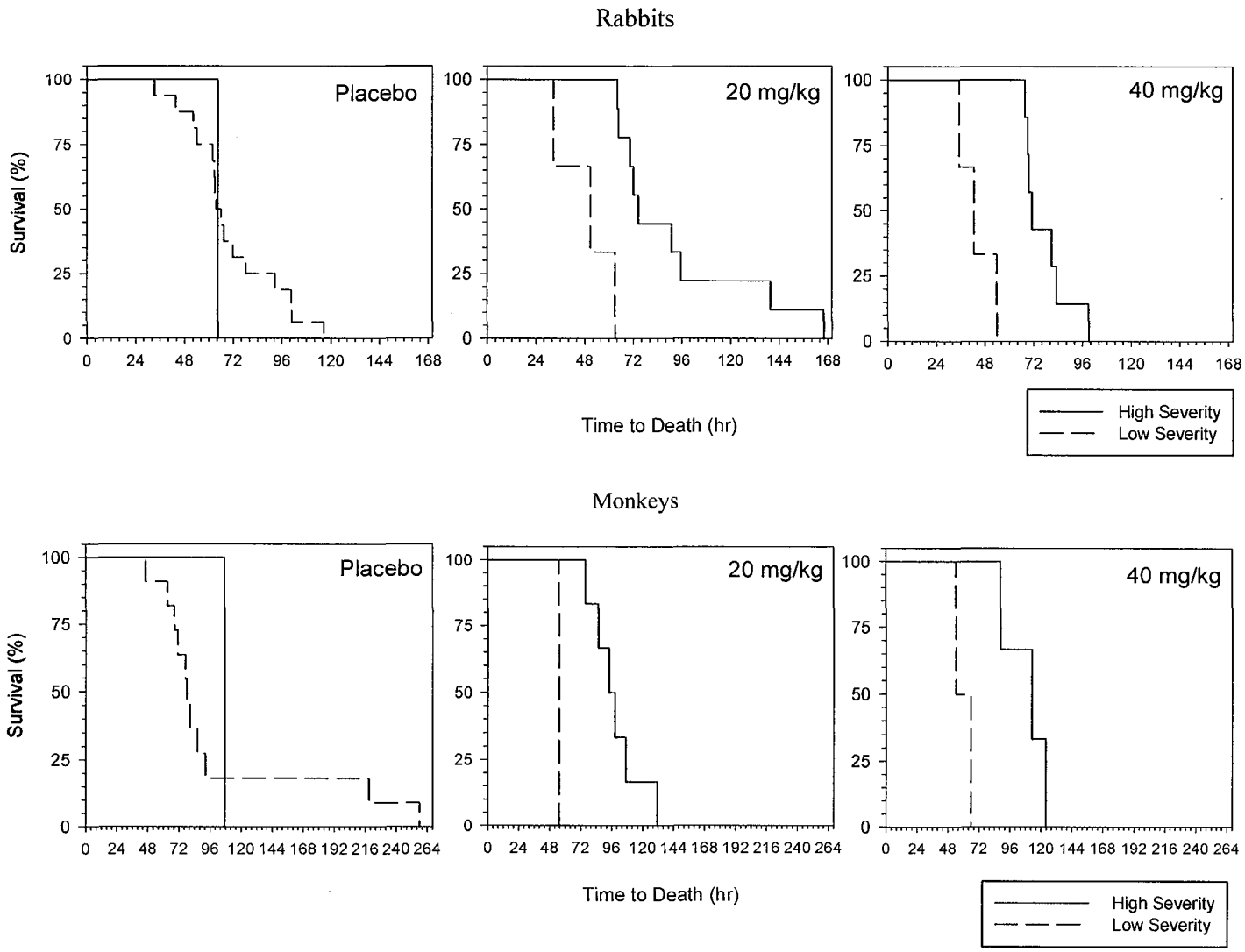


Figure 4.3.4.2-5 Comparison of Time to Death in Monkeys by Quantile of Raxibacumab Exposure



The applicant also contends that time to death in animals with moderate to marked inflammation was longer than that for animals without moderate to marked inflammation, i.e. that more severe inflammatory brain findings are associated with a longer time to death. Figure 4.3.4.2-6 shows percent survival and survival time in both rabbits and monkeys by treatment group and CNS pathology severity. In rabbits, this appears to be true only for the raxibacumab treated animals; in animals that received placebo, rabbits with low grade CNS findings lived longer than those with high grade pathology. Similar findings were observed in monkeys. These observations suggest that animals that received raxibacumab and exhibited high grade CNS pathology generally lived longer than animals exhibiting low grade pathology. The opposite was found for placebo animals.

Figure 4.3.4.2-6 Comparison of Time to Death in Animals by Severity of CNS Pathology



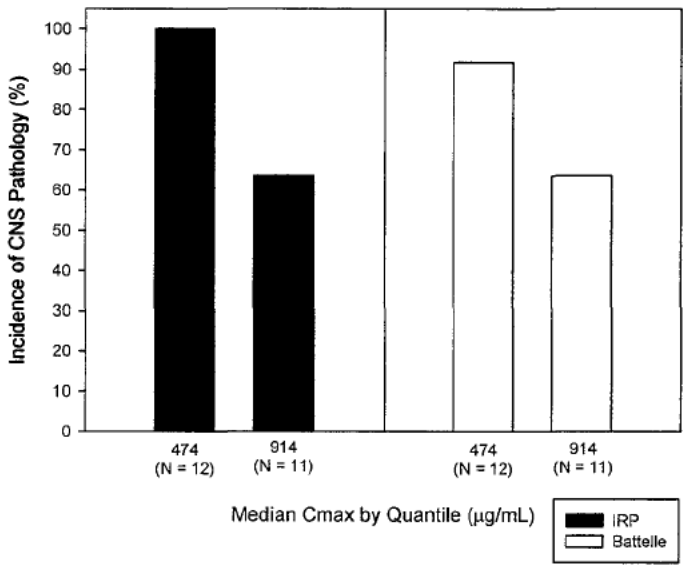
Gross necroscopy and microscopic histopathology data from the pivotal efficacy trials in both rabbits and monkeys (Studies 682-G005758 and 724-G005829) were reviewed separately by a board-certified pathologist at the study site (Battelle) and by independent, blinded microscopic histology review (IRP). To evaluate a potential relationship between these CNS findings and exposure to raxibacumab, an exposure-response analysis was conducted.

4.3.4.2.1 CNS Findings in Rabbits

Comparisons of Cmax and AUCinf values and the incidence of CNS pathology in rabbits are presented by quantiles in Figure 4.3.4.2.1-1. A lower incidence of CNS pathology was observed with higher quantiles of exposure. Comparisons of Cmax values and the incidence of specific pathological findings (bacteria, hemorrhage, inflammation, necrosis) in rabbits are presented by quantiles in Figure 4.3.4.2.1-2. A comparison of AUCinf values by quantile showed results identical to the results for Cmax (data not shown). The quantile data suggest a lower incidence of bacteria, hemorrhage, and inflammation, but not necrosis, with higher exposures (both Cmax and AUC).

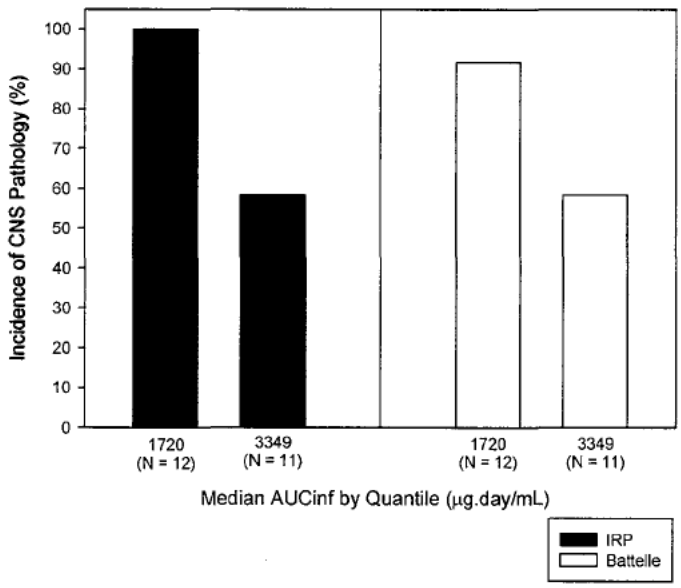
Figure 4.3.4.2.1-1 **Comparison of Incidence of CNS Pathology in Rabbits that Died by Quantile of Raxibacumab Exposure**

Cmax



Quantile ranges: Quantile 1 (342 – 543), Quantile 2 (624 – 1166)

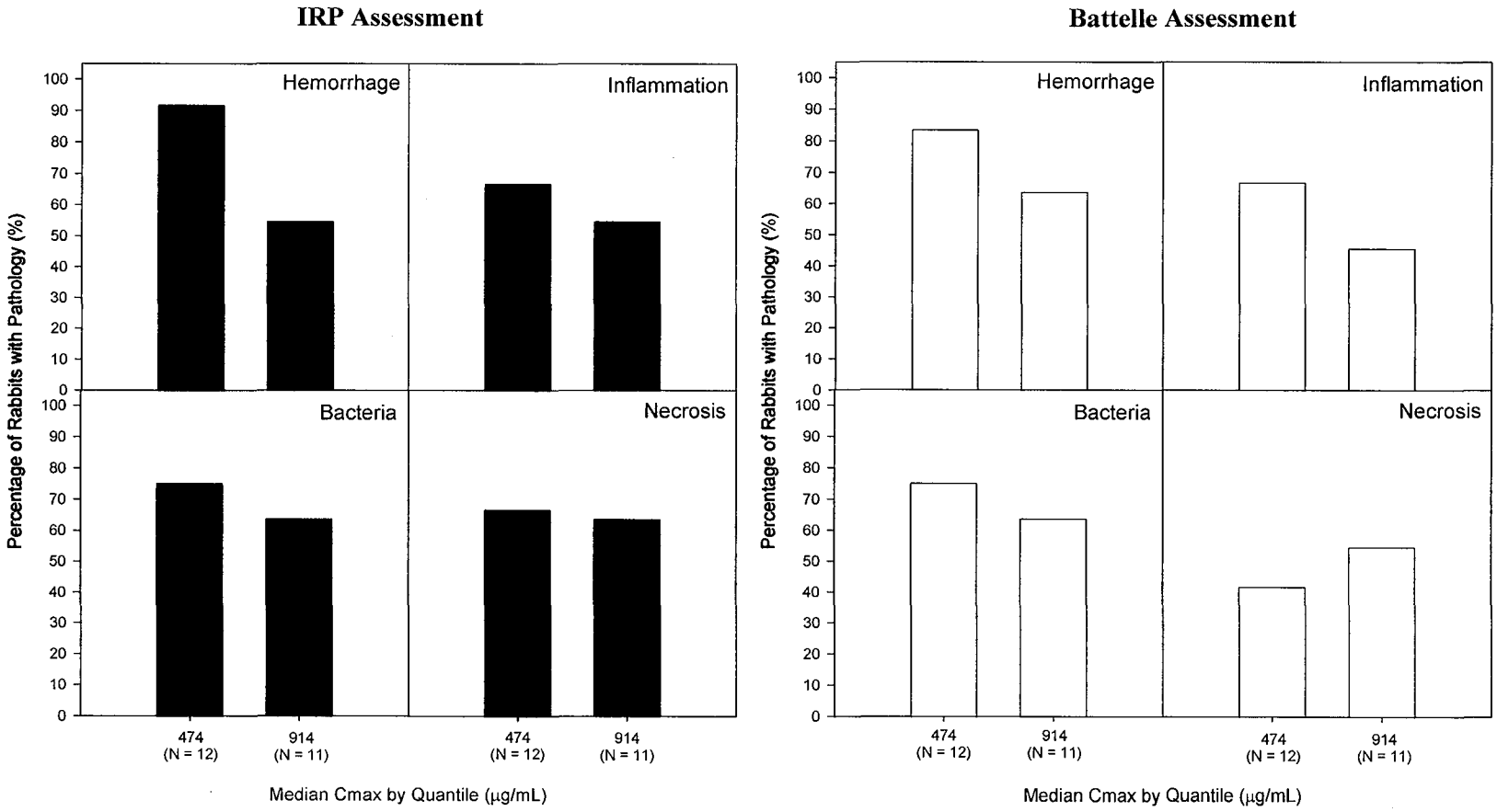
AUC



Quantile ranges: Quantile 1 (1380 – 2144), Quantile 2 (2971 – 3978)

IRP, readings performed by independent pathologist review
Battelle, readings performed by board-certified pathologist at study site

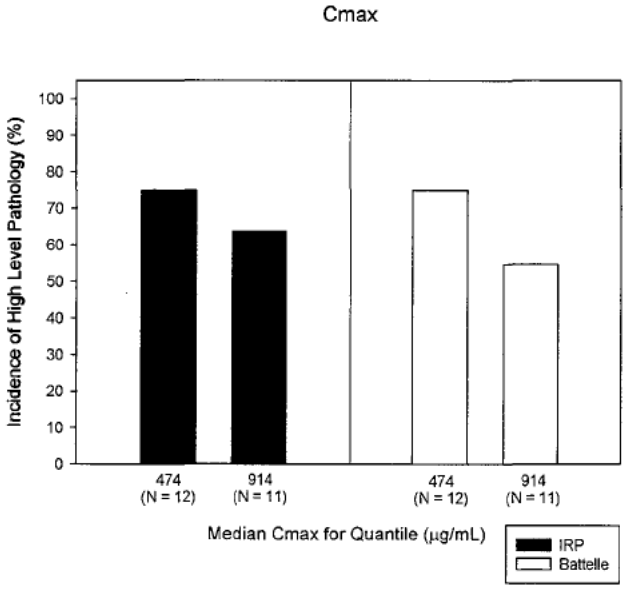
Figure 4.3.4.2.1-2 **Comparison of Incidence of Specific CNS Pathology Findings in Rabbits that Died by Quantile of Raxibacumab Cmax**



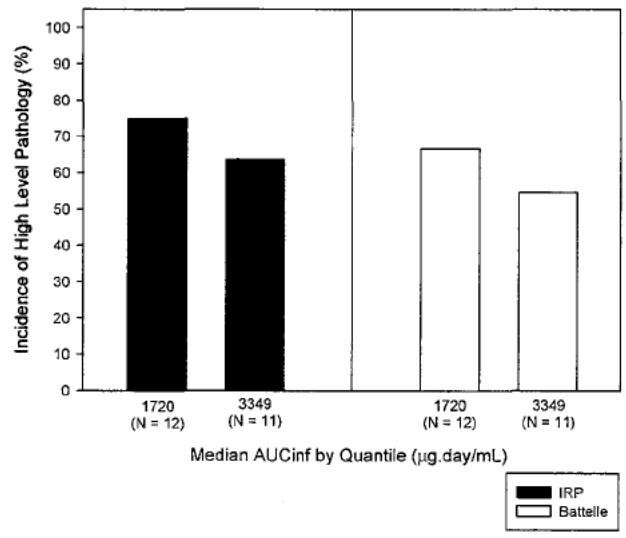
Quantile ranges: Quantile 1 (342 – 543), Quantile 2 (624 – 1166)
IRP, readings performed by independent pathologist review
Battelle, readings performed by board-certified pathologist at study site

Comparisons of Cmax and AUCinf values and the incidence of high level CNS pathology (defined as pathology Grades ≥ 3) in rabbits are presented by quantiles in Figure 4.3.4.2.1-3. A lower incidence of high level CNS pathology was observed in the higher quantiles of exposure, but the number of animals resulting in this difference are small.

Figure 4.3.4.2.1-6 Comparison of Incidence of High Level (Grades ≥ 3) CNS Pathology in Rabbits that Died by Quantile of Raxibacumab Exposure



Quantile ranges: Quantile 1 (342 – 543), Quantile 2 (624 – 1166)



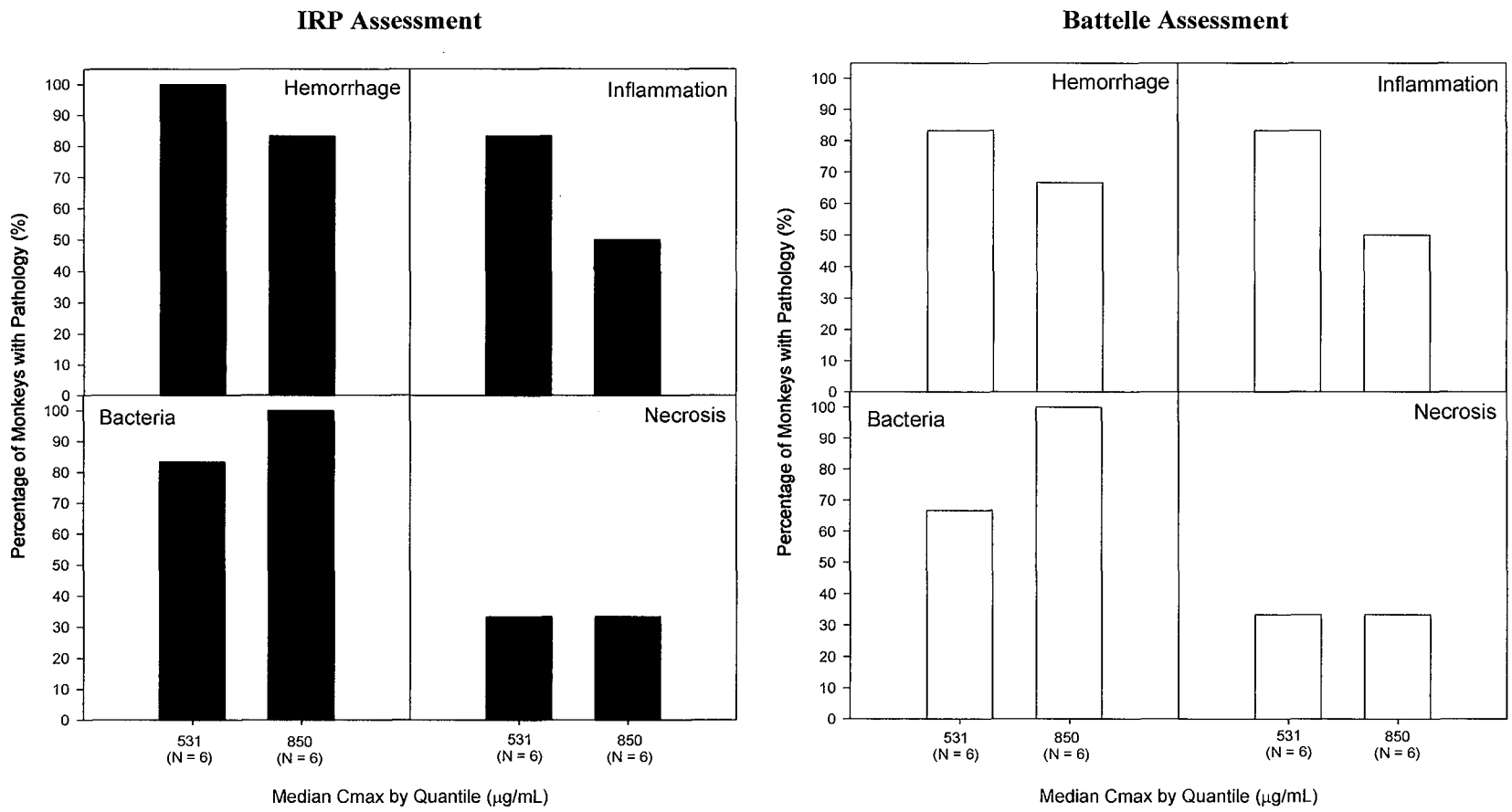
Quantile ranges: Quantile 1 (1380 – 2144), Quantile 2 (2971 – 3978)

IRP, readings performed by independent pathologist review
Battelle, readings performed by board-certified pathologist at study site

4.3.4.2.2 CNS Findings in Monkeys

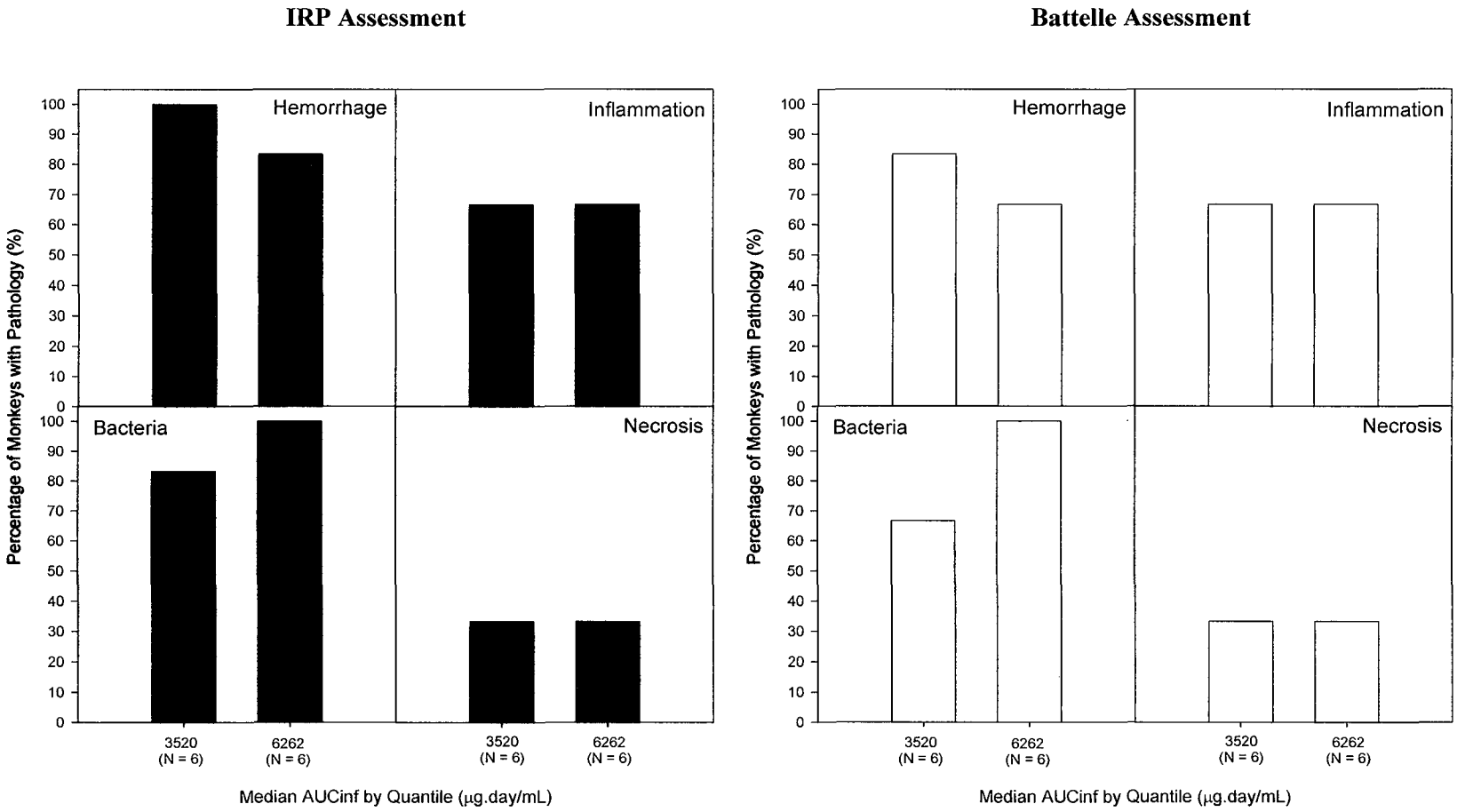
Based on the Battelle assessment, all but one monkey (11/12, 91.7%) that received raxibacumab treatment exhibited CNS findings on necroscopy. Based on the independent pathologist's assessment (IRP), all raxibacumab monkeys that received raxibacumab and died had CNS pathology. Comparisons of Cmax and AUCinf values and the incidence of specific pathological findings (bacteria, hemorrhage, inflammation, necrosis) in monkeys are presented by quantiles in Figures 4.3.4.2.2-1 and 4.3.4.2.2-2. The quantile data suggest a lower incidence of hemorrhage and inflammation with higher exposures (primarily for Cmax). Conversely, the higher quantiles of exposure had a higher incidence of bacteria in the CNS. The incidence of necrosis did not differ between the two quantiles of exposure.

Figure 4.3.4.2.2-1 **Comparison of Incidence of Specific CNS Pathology Findings in Monkeys that Died by Quantile of Raxibacumab Cmax**



Quantile ranges: Quantile 1 (276 – 600), Quantile 2 (613 – 1097)
IRP, readings performed by independent pathologist review
Battelle, readings performed by board-certified pathologist at study site

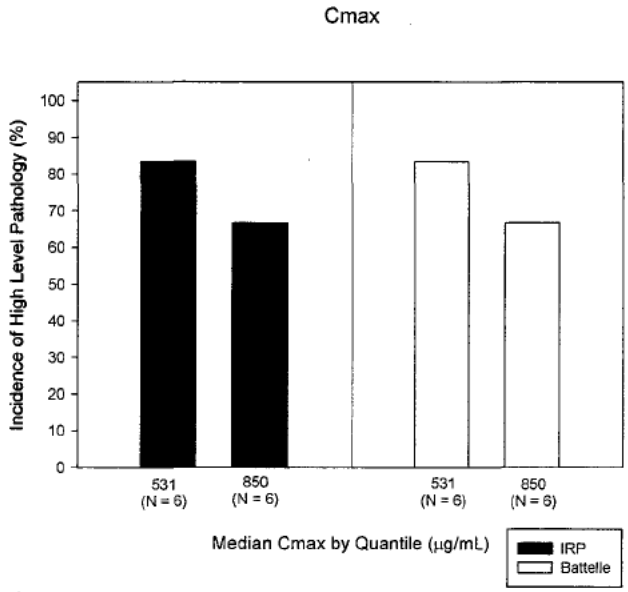
Figure 4.3.4.2.2-2 Comparison of Incidence of Specific CNS Pathology Findings in Monkeys that Died by Quantile of Raxibacumab AUC



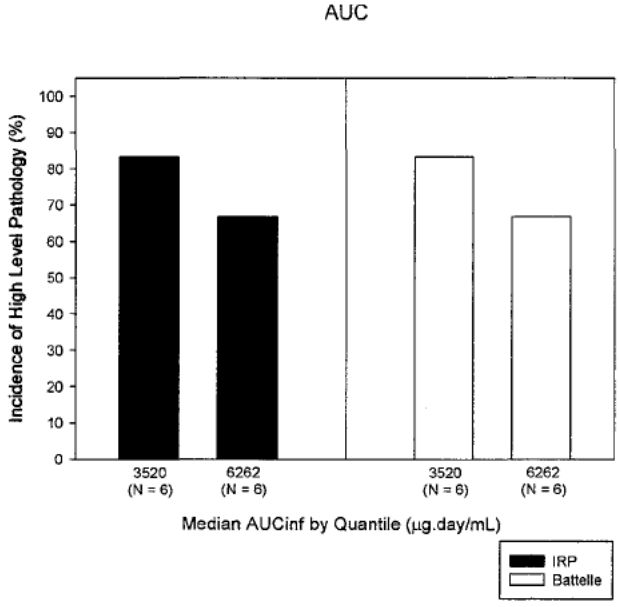
Quantile ranges: Quantile 1 (2597 – 4080), Quantile 2 (4780 – 8653)
IRP, readings performed by independent pathologist review
Battelle, readings performed by board-certified pathologist at study site

Comparisons of Cmax and AUCinf values and the incidence of high level CNS pathology (defined as pathology Grades ≥ 3) in monkeys are presented by quantiles in Figure 4.3.4.2.2-3 (both the Battelle and independent pathologist assessments were identical). A lower incidence of high level CNS pathology was observed in the higher quantiles of exposure, but this finding can be attributed to one animal (N = 4 in the lower quantile versus N = 5 in the higher quantile).

Figure 4.3.4.2.2-3 Comparison of Incidence of High Level (Grades ≥ 3) CNS Pathology in Monkeys that Died by Quantile of Raxibacumab Exposure



Quantile ranges: Quantile 1 (276 – 600), Quantile 2 (613 – 1097)



Quantile ranges: Quantile 1 (2597 – 4080), Quantile 2 (4780 – 8653)

IRP, readings performed by independent pathologist review
Battelle, readings performed by board-certified pathologist at study site

4.3.4.2.3 Reviewer's Conclusions on CNS findings

In conclusion, no clear relationship between exposure and CNS pathology can be discerned based on the limited amount of data in animals that died. Overall, the raxibacumab 20 mg/kg dose group had a higher incidence and severity of CNS findings than the 40 mg/kg group in both rabbits and monkeys. Due to the small numbers of animals that exhibited specific pathology findings, no definitive conclusions about exposure-response for bacteria, hemorrhage, inflammation, or necrosis could be made.

The potential hypothesis proposed by the applicant that animals that received raxibacumab survived longer (and thus had more brain lesions that increased over time) does not appear to be a likely explanation for the higher CNS pathology rates in animals treated with raxibacumab. In rabbits that died, placebo animals have comparable survival times versus animals that received the higher raxibacumab dose (40 mg/kg). In monkeys that died, animals that received placebo lived longer than those receiving raxibacumab.

In rabbits that died, animals in the lower Cmax and AUC quantiles appeared to live longer than the other groups and placebo, but in monkeys there was no discernable difference between survival times for the higher and lower quantiles of exposure. The placebo group exhibited a longer survival time compared to the raxibacumab groups. This suggests that in animals that died, the magnitude of raxibacumab exposure did not affect survival time.

The applicant also contends that time to death in animals with moderate to marked inflammation was longer than that for animals without moderate to marked inflammation, i.e. that more severe inflammatory brain findings are associated with a longer time to death. In rabbits, this appears to be true only for the raxibacumab treated animals; in animals that received placebo, rabbits with low grade CNS findings lived longer than those with high grade pathology. Similar findings were observed in monkeys. These observations suggest that animals that received raxibacumab and exhibited high grade CNS pathology generally lived longer than animals exhibiting low grade pathology. The opposite was found for placebo animals.

5 LISTING OF ANALYSIS CODES AND OUTPUT FILES

File Name	Description	Location in \\cdsnas\pharmacometrics\
Survival Plots.JNB	SigmaPlot file for all survival box plots and time to event Kaplan Meier Plots	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Graphs Group1\
CNS Plots.JNB	SigmaPlot file for all quantile comparisons for CNS pathology E/R analysis	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Graphs Group1\
rsPKPA.sdd	SPlus PK and PA data file for rabbits that survived	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Graphs Group2\
rdPKPA.sdd	Splus PK and PA data file for rabbits that died	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Graphs Group2\
msPKPA.sdd	SPlus PK and PA data file for monkeys that survived	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Graphs Group2\
mdPKPA.sdd	Splus PK and PA data file for monkeys that died	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Graphs Group2\
rs PLOT.sgr	SPlus scatter plot for rabbits that survived	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Graphs Group2\
rd PLOT.sgr	SPlus scatter plot for rabbits that died	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Graphs Group2\
ms PLOT.sgr	SPlus scatter plot for monkeys that survived	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Graphs Group2\
rmd PLOT.sgr	SPlus scatter plot for monkeys that died	\\cdsnas\pharmacometrics\BLA125349_Raxibacumab_KB\ER Analyses\Graphs Group2\

6 APPENDICES

Appendix 1 Clinical Pharmacology Reviewer's Population PK Analysis

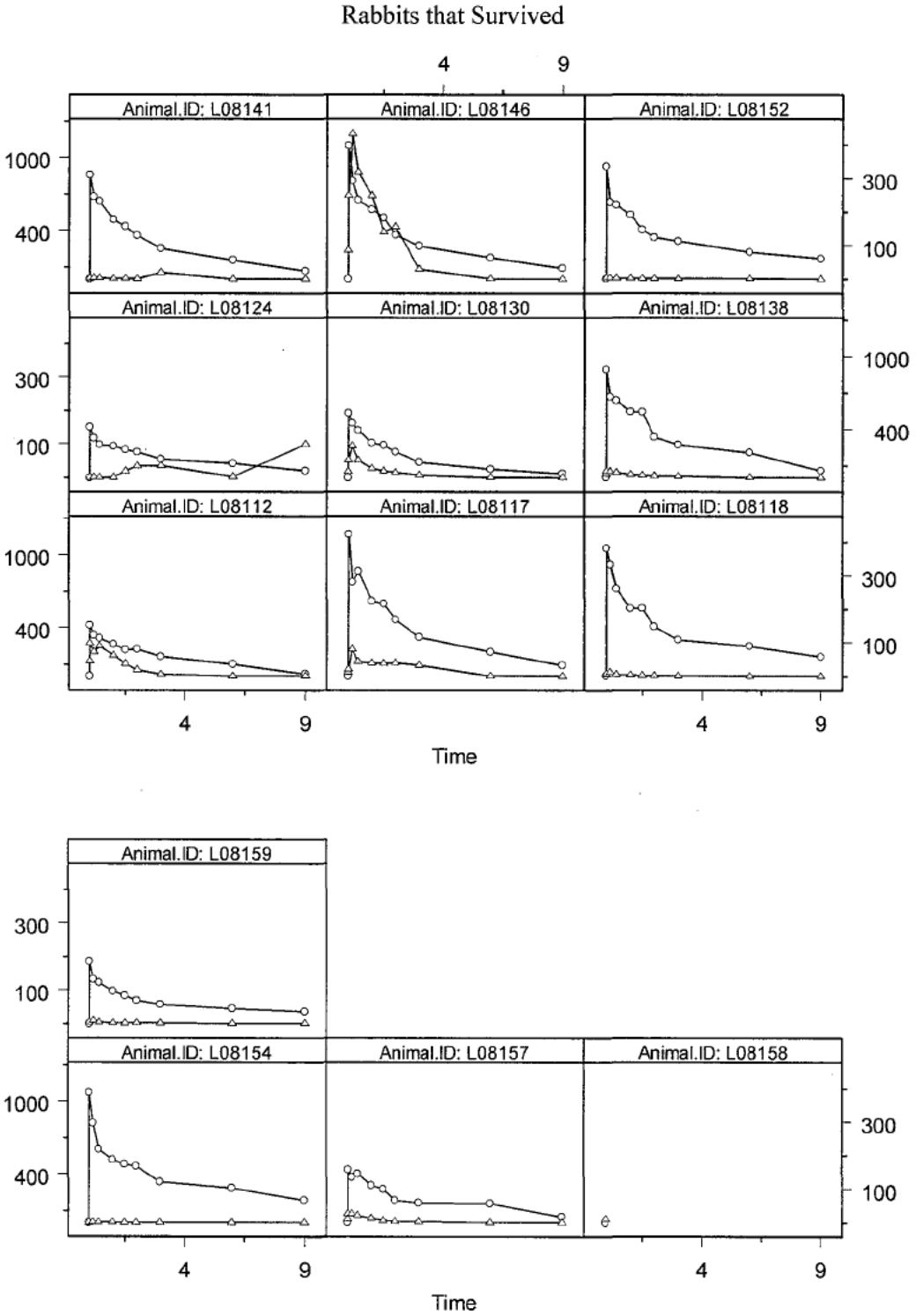
As mentioned in Section 3.3.1 in the Pharmacometric Review, the choice of proportional error model for V2 was found to be questionable. The reviewer recreated the population PK analysis from the basic two compartment model with the following changes during each run. (Link: \\cdsnas\PHARMACOMETRICS\BLA125349_Raxibacumab_KB\PPK Analyses\Structure Model)

Run #	Description	Objective function
Run 1	Applicant's Model	20195.49
	The model was recreated due to questionable choice of error model, evidence of shrinkage in interindividual variability on V2 (34%) and Q (60%) and the lack of covariance step. At the same time, the use of "weight-74" as centering method versus the allometric method was evaluated.	
Run 2	Base model (two compartment model; log-normal distribution for all interindividual variability parameters; combined additive and proportional model for residual error; No covariates)	20683.00
Run 3	Based on diagnostic plots, allometric model for CL, V1 and V2	20256.309
Run 4	Based on large SE on interindividual variability on Q and V2 and shrinkage in post hoc Q, variability on Q fixed to 0	20276.355
Run 5	**Based on shrinkage in post hoc V2 and imprecise estimation, variability on V2 fixed to zero. Based on diagnostics, added correlation parameter between CL and V1**	**20311.57**
Run 6	Fix allometric coefficients	20375.01
Run 8	Add sex as a covariate on V1	20376.09
Run 9	Add sex as a covariate on V1	20376.718

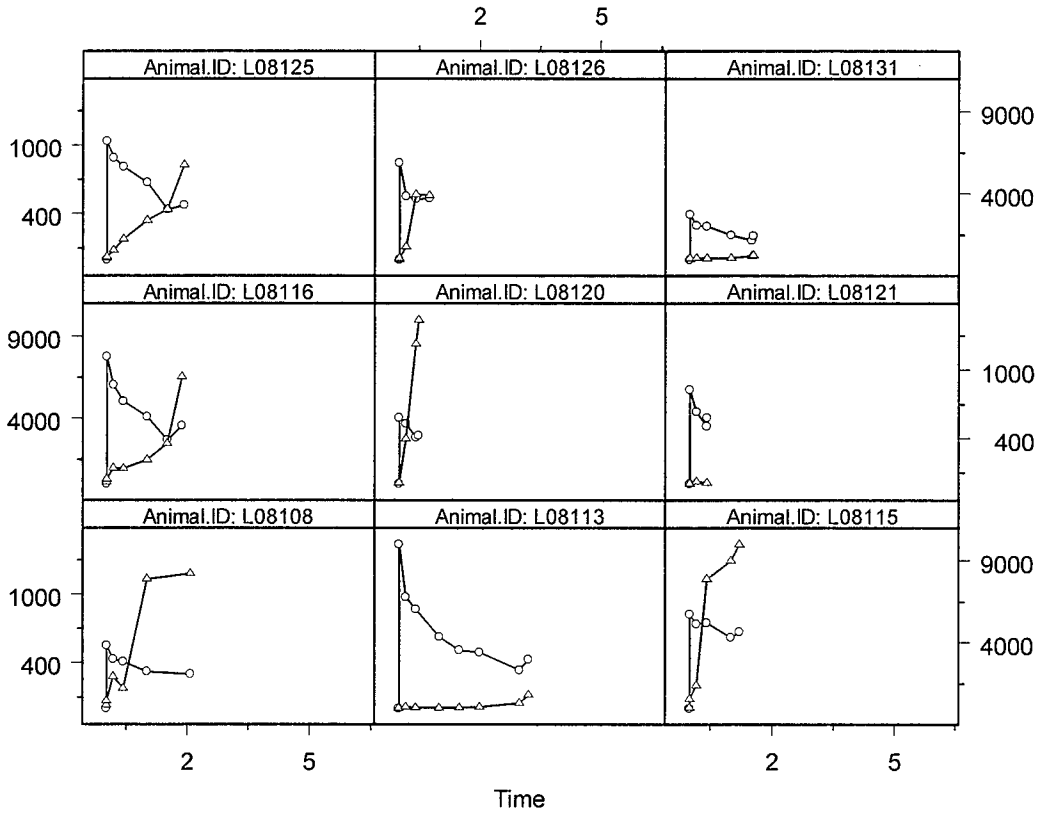
Using Run 5 as the final model, the following parameters were derived.

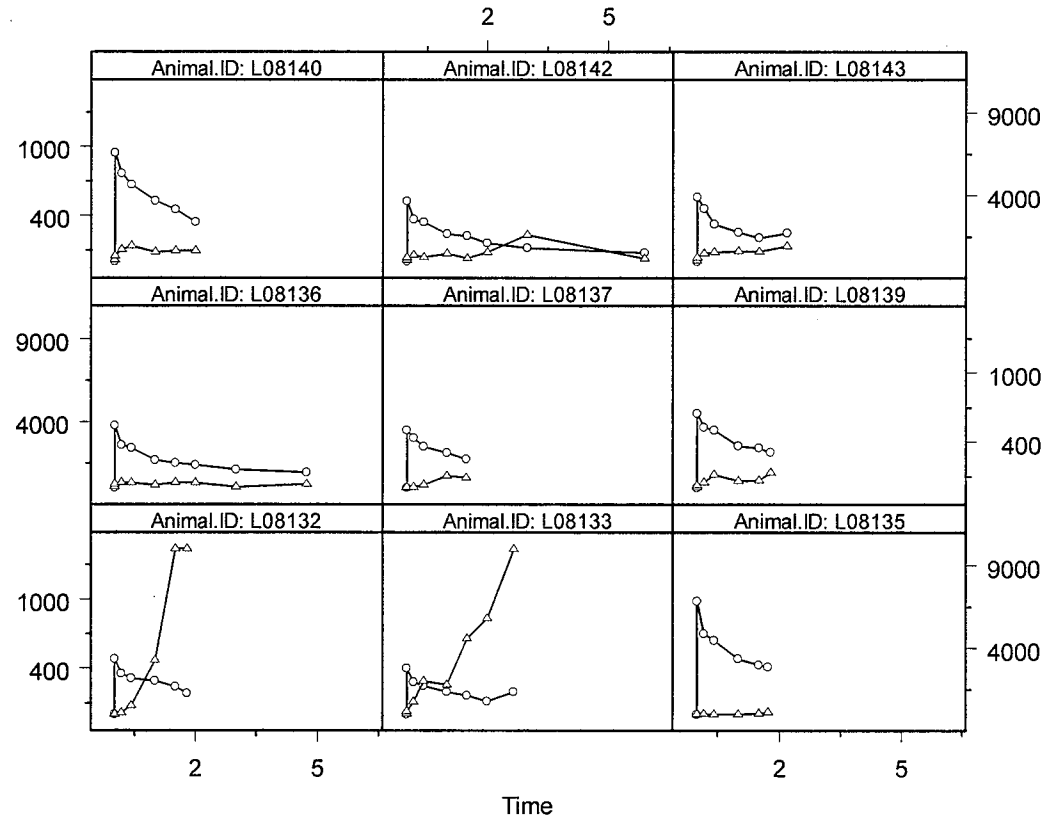
Primary Parameter	Value (RSE [%])	CV%
Volume of distribution (Central) (mL)	3040 (1.04)	17.0 (9.25)
Effect of weight on V1	V1 = 3040 * (weight /70)^0.738 (6.02)	
At 45 kg	2194.1	
At 62 kg	2779.6	
At 86 kg	3538.8	
At 156 kg	5491.8	
Clearance (mL/day)	173 (1.24)	20.0 (10.4)
Effect of weight on CL	CL = 173 * (weight /70)^0.773 (6.47)	
At 45 kg	122.9	
At 62 kg	157.5	
At 86 kg	202.8	
At 156 kg	321.4	
Volume of distribution (Peripheral) (mL)	2030 (2.16)	0 FIX
Effect of weight on V2	V2 = 2030* (weight /70)^0.664 (12.8)	
At 45 kg	1513.9	
At 62 kg	1872.8	
At 86 kg	2327.3	
At 156 kg	3456.1	
CLD2 (mL/day)	420 (6.33)	0 FIX

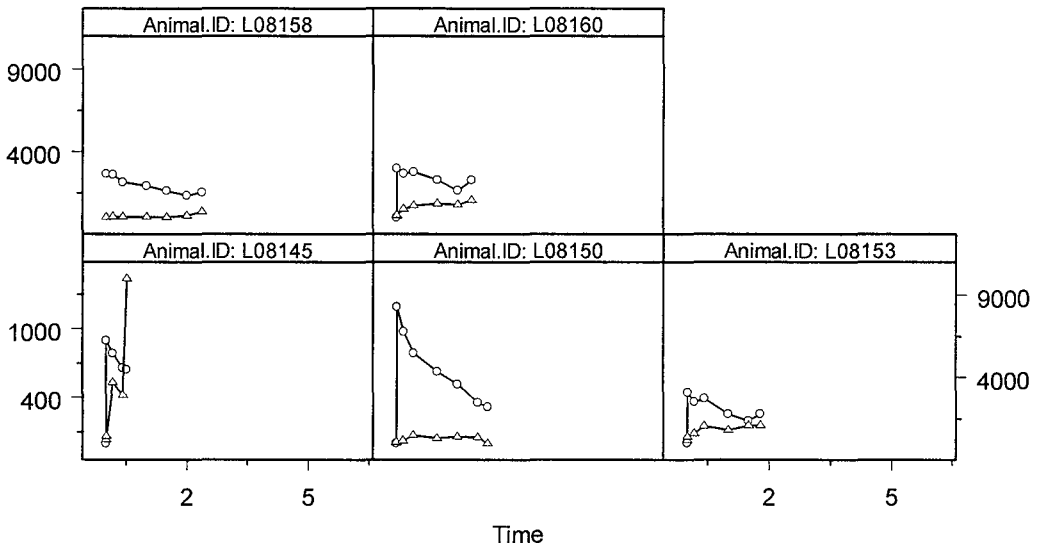
Appendix 2 Serum Raxibacumab Concentrations and PA Concentrations for Individual Rabbits in Study 682-G005758



Rabbits that Died



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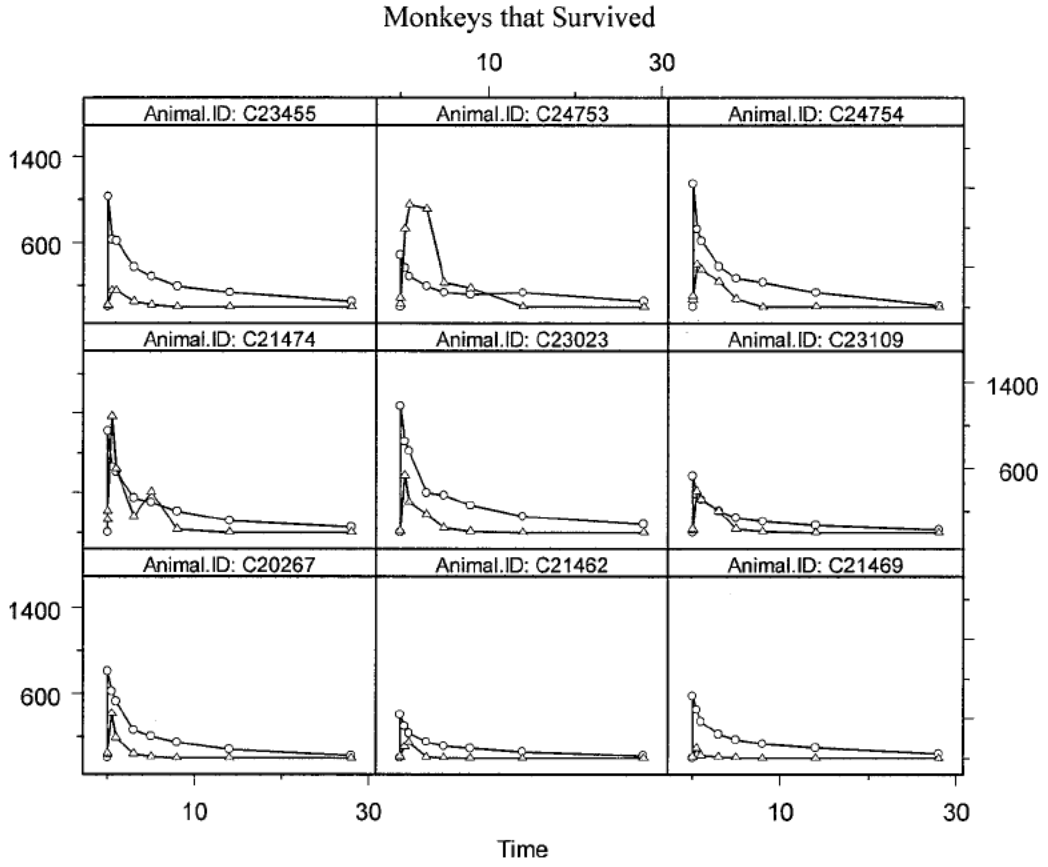
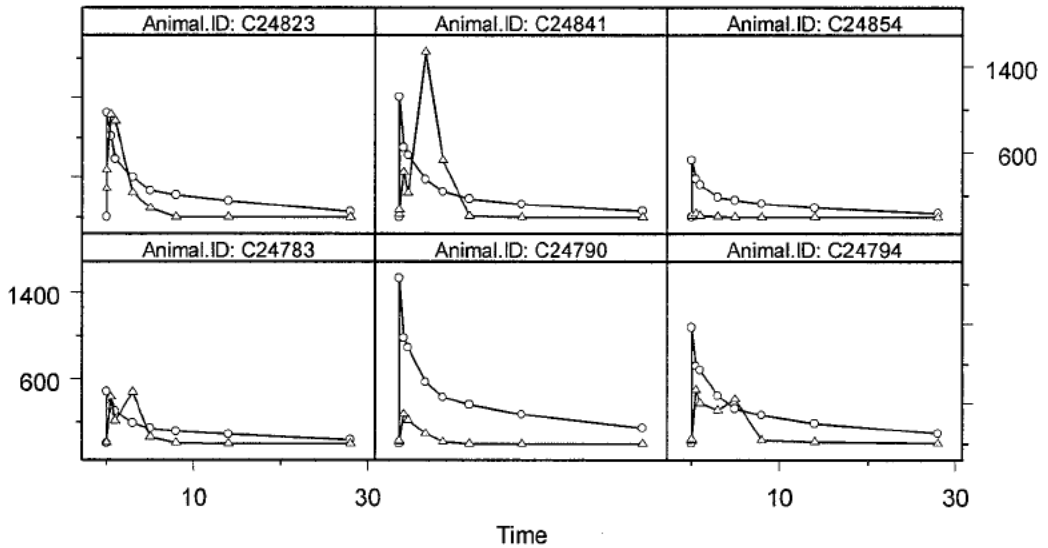
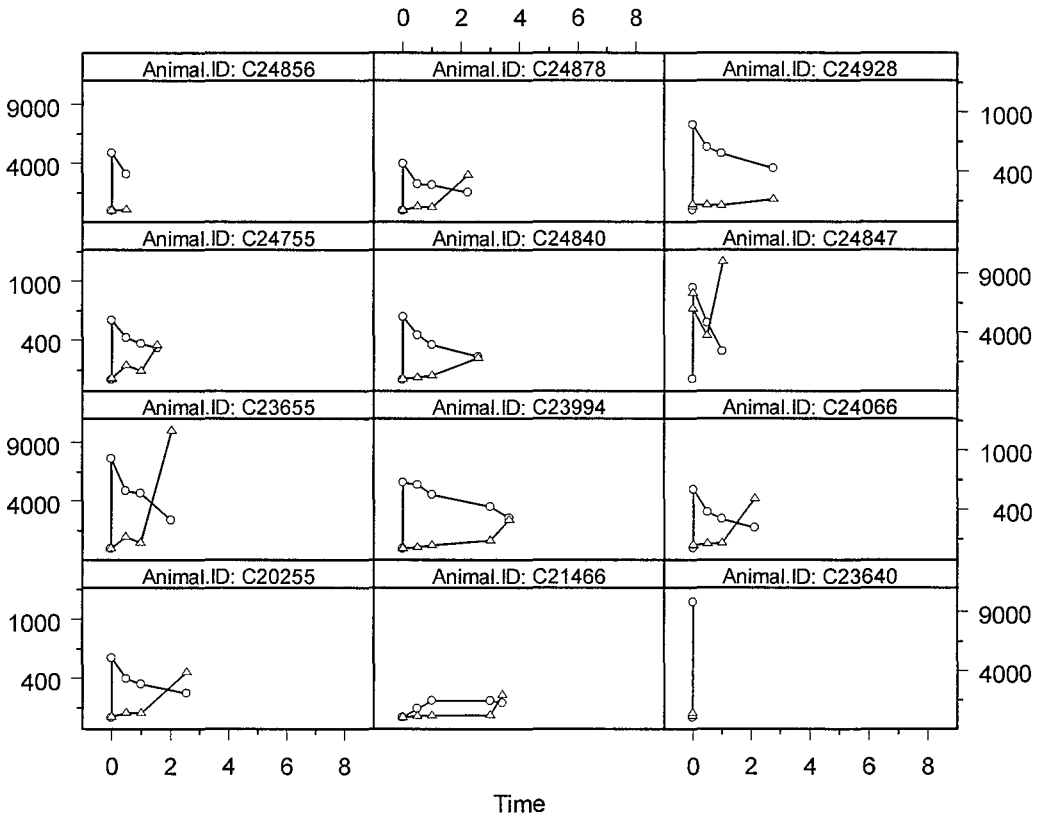
Animal.ID: L08160
Animal.ID: L08145
Animal.ID: L08150
Animal.ID: L08153
9000
4000
1000
400
9000
4000
2
5
2
5
Time

Appendix 3 Serum Raxibacumab Concentrations and PA Concentrations for Individual Monkeys in Study 724-G005829





Monkeys that Died



CLINICAL PHARMACOLOGY NDA FILEABILITY CHECKLIST

BLA:	125349
Drug Name:	Raxibacumab
Applicant:	Human Genome Sciences
Submission Date:	14MAY2009
Filing Date:	10JUN2009
PDUFA Date:	14NOV2009
OCP Primary Reviewer:	Kimberly L. Bergman, PharmD
OCP Team Leader:	Philip M. Colangelo, PharmD, PhD

QUESTION	***YES***	***NO***	***NA***	***COMMENTS***
Fileability: ***Is the Clinical Pharmacology section of the application fileable?*** ***(if 'NO', please comment as to why it is not fileable)***	***X***			
Fileability Review Components				
1. Is the clinical pharmacology section of the NDA organized in a manner to allow substantive review to begin (including a table of contents, proper pagination, reference links, etc.)?	☒	☐	☐	Of note: information request to be sent to applicant regarding the location of summary of changes to amended clinical study reports.
2. Are the clinical pharmacology studies of appropriate design and breadth of investigation to meet the basic requirements for approvability of this product?	☒	☐	☐	
3. If multiple formulations were used in the clinical development of the product, does the NDA contain appropriate biopharmaceutics information to allow comparison between the clinical development and to-be-marketed product(s) (i.e. pivotal BE)?	☒	☐	☐	PK information (Study HGS1021-C1064) has been obtained with the final commercial formulation (21-A) manufactured (b) (4) for commercial production (M11); this formulation was used in the pivotal animal efficacy and human safety studies
4. If unapproved products or altered approved products were used as active controls, was bioequivalence to the approved product demonstrated?	☐	☐	☒	
5. Are complete and relevant bioanalytical reports included in the NDA submission?	☒	☐	☐	
6. If applicable, was the sponsor's request for a waiver of the requirement for submission of in vivo bioavailability data included in the NDA submission?	☐	☐	☒	
7. Are complete datasets supporting the clinical pharmacology studies included in the NDA submission?	☒	☐	☐	

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7. Are complete datasets supporting the clinical pharmacology studies included in the NDA submission?	☒	☐	☐	

OCP Primary Reviewer

6/10/09
Date

OCP Team Leader

6/10/09
Date