SUMMARY OF SAFETY AND EFFECTIVENESS DATA (SSED)

I. GENERAL INFORMATION

Device Generic Name:	Next Generation Sequencing Oncology Panel, Somatic or Germline Variant Detection System
Device Trade Name:	Guardant360® CDx
Device Procode:	PQP
Applicant's Name and Address:	Guardant Health, Inc. 505 Penobscot Drive Redwood City, CA 94306 USA
Date(s) of Panel Recommendation:	None
Premarket Approval Application (PMA) Number:	P200010/S001
Date of FDA Notice of Approval:	May 21, 2021

The original PMA (P200010) for Guardant360® CDx was approved on August 7, 2020 for the detection of genetic alterations in circulating cell-free DNA (cfDNA) from plasma of peripheral whole blood derived from patients who may benefit from one of the FDA-approved therapies for non-small cell lung cancer (NSCLC). The SSED to support the previously approved indication is available on the CDRH website.

The current panel-track supplement was submitted to expand the intended use and indications for use of Guardant360® CDx to include a companion diagnostic indication for the detection of epidermal growth factor receptor (*EGFR*) exon 20 insertion mutations in NSCLC patients who may benefit from treatment with RYBREVANT™ (amivantamab-vmjw).

II. INDICATIONS FOR USE

Guardant360® CDx is a qualitative next generation sequencing-based *in vitro* diagnostic device that uses targeted high throughput hybridization-based capture technology for detection of single nucleotide variants (SNVs), insertions and deletions (indels) in 55 genes, copy number amplifications (CNAs) in two (2) genes, and fusions in four (4) genes. Guardant360® CDx utilizes circulating cell-free DNA (cfDNA) from plasma of peripheral whole blood collected in Streck Cell-Free DNA Blood Collection Tubes (BCTs). The test is intended to be used as a companion diagnostic to identify non-small cell lung cancer (NSCLC) patients who may benefit from treatment with the targeted therapies listed in Table 1 in accordance with the approved therapeutic product labeling.

Table 1. Companion Diagnostic Indications

Indication	Biomarker	Therapy
Non-small cell lung cancer (NSCLC)	*EGFR* exon 19 deletions, L858R, and T790M*	TAGRISSO® (osimertinib)
	EGFR exon 20 insertions	RYBREVANT™ (amivantamab-vmjw)

A negative result from a plasma specimen does not assure that the patient's tumor is negative for genomic findings. NSCLC patients who are negative for the biomarkers listed in Table 1 should be reflexed to tissue biopsy testing for Table 1 biomarkers using an FDA approved tumor tissue test, if feasible.

*The efficacy of TAGRISSO® (osimertinib) has not been established in the *EGFR* T790M plasma-positive, tissue-negative or unknown population and clinical data for T790M plasma-positive patients are limited; therefore, testing using plasma specimens is most appropriate for consideration in patients from whom a tumor biopsy cannot be obtained.

Additionally, the test is intended to provide tumor mutation profiling to be used by qualified health care professionals in accordance with professional guidelines in oncology for cancer patients with any solid malignant neoplasms. The test is for use with patients previously diagnosed with cancer and in conjunction with other laboratory and clinical findings.

Genomic findings other than those listed in Table 1 are not prescriptive or conclusive for labeled use of any specific therapeutic product.

Guardant360® CDx is a single-site assay performed at Guardant Health, Inc.

III.   CONTRAINDICATIONS

There are no known contraindications.

IV.   WARNINGS AND PRECAUTIONS

The warnings/precautions and limitations are included in the Guardant360® CDx assay labeling.

V.   DEVICE DESCRIPTION

Guardant360® CDx is a single-site test performed at Guardant Health, Inc. The test includes reagents, software, and procedures for testing cfDNA from whole blood samples. The test uses 5-30 ng of cfDNA for library construction and next generation sequencing. Sequencing data is processed using a customized bioinformatics pipeline designed to detect several classes of genomic alterations, including nucleotide substitutions, indels, CNA, and genomic fusions / rearrangements. The device is designed to sequence 74 genes, but only report pre-defined and *de novo* alterations within the 55

genes outlined in Table 2. The test's reportable range for SNVs and indels covers approximately 46,000 bases.

Table 2. Genes Containing Alterations Detected by the Guardant360® CDx

Alteration Type	Genes
Single Nucleotide Variants (SNVs)	*AKT1, ALK, APC, AR, ARAF, ATM*, BRAF, BRCA1**, BRCA2**, CCND1, CDH1, CDK4, CDK6, CDK12*, CDKN2A, CTNNB1, EGFR, ERBB2, ESR1, FGFR1, FGFR2, FGFR3, GATA3, GNA11, GNAQ, HRAS, IDH1, IDH2, KIT, KRAS, MAP2K1, MAP2K2, MET, MLH1, MTOR, MYC, NF1, NFE2L2, NRAS, NTRK1, NTRK3, PDGFRA, PIK3CA, PTEN, RAF1, RET, RHEB, ROS1, SMAD4, SMO, STK11, TERT, TSC1, VHL*
Indels	*AKT1, ALK, APC, ATM*, BRAF, BRCA1**, BRCA2**, CDH1, CDK12*, CDKN2A, EGFR, ERBB2, ESR1, FGFR2, GATA3, HNF1A, HRAS, KIT, KRAS, MET, MLH1, NF1, PDGFRA, PIK3CA, PTEN, RET, ROS1, STK11, TSC1, VHL*
Copy Number Amplifications (CNAs)	*ERBB2, MET*
Fusions / Rearrangements	*ALK, NTRK1, RET, ROS1*

*Reporting is enabled for pathogenic germline alterations only. Somatic alterations will not be reported.
** Reporting is enabled for both germline and somatic alterations.

Test Output

The test report includes variants reported in the following categories; see Table 3:

Table 3. Category Definitions

Category	Guardant360® CDx			Comments
	Prescriptive use for a Therapeutic Product	Clinical Performance	Analytical Performance	
Category 1: Companion Diagnostic (CDx)	**Yes**	**Yes**	**Yes**	ctDNA biomarkers linked to the safe and effective use of the corresponding therapeutic product, for which Guardant360® CDx has demonstrated clinical performance shown to support therapeutic efficacy and strong analytical performance for the biomarker.
Category 2: ctDNA Biomarkers with Strong Evidence of Clinical Significance in ctDNA	No	No	**Yes**	ctDNA biomarkers with strong evidence of clinical significance presented by other FDA-approved liquid biopsy companion diagnostics for which Guardant360® CDx has demonstrated analytical reliability but not clinical performance.
Category 3A: Biomarkers with Evidence of Clinical Significance in	No	No	**Yes**	ctDNA biomarkers with evidence of clinical significance presented by tissue-based FDA-approved companion diagnostics or professional guidelines for

Category	Guardant360® CDx			Comments
	Prescriptive use for a Therapeutic Product	Clinical Performance	Analytical Performance	
tissue supported by: strong analytical validation using ctDNA				which Guardant360® CDx has demonstrated analytical performance including analytical accuracy, and concordance of blood-based testing to tissue-based testing for the biomarker.
Category 3B: Biomarkers with Evidence of Clinical Significance in tissue supported by: analytical validation using ctDNA	No	No	**Yes**	ctDNA biomarkers with evidence of clinical significance presented by tissue-based FDA-approved companion diagnostics or professional guidelines for which Guardant360® CDx has demonstrated minimum analytical performance including analytical accuracy.
Category 4: Other Biomarkers with Potential Clinical Significance	No	No	**Yes**	ctDNA biomarkers with emergent evidence based on peer-reviewed publications for genes/variants in tissue, variant information from well-curated public databases, or *in-vitro* pre-clinical models, for which Guardant360® CDx has demonstrated minimum analytical performance.

Test Kit Contents

The test includes the Guardant360® CDx Blood Collection Kit (BCK), which is sent to ordering laboratories. Each BCK contains two blood collection tubes. The BCK also contains supporting packaging materials, instructions for use and a return shipping label. The BCK contains the following components:

- Streck blood collection tubes for specimen collection, stabilization, and transport of cfDNA; 2 per kit.
- Cushioning materials to prevent breakage of the blood collection tubes; 2 per kit
- Foam tray for protection of collection tubes during transport
- Absorbent sheet to be used during specimen shipping
- Biohazard specimen bag for protection during specimen transport
- Return shipping label for return of specimen to Guardant Health
- Barcodes for specimen identification and shipping instructions
- Instructions for Use for blood draw
- Patient welcome brochure which contains an overview of the test
- Test requisition form to complete to order Guardant360® CDx for a patient.

The test also includes the Guardant360® CDx Sample Preparation Kit (SPK), which is used in the Guardant Health Clinical Laboratory. The SPK contains reagents for library preparation, library enhancement, and cfDNA quantification/qualification. The kit is assembled into six (6) different boxes (referred to as box 1, 2, 3, 4a, 4b, and 4c) based on

the usage of the reagents. The division of reagents amongst the boxes reflects different storage conditions and/or locations (e.g. different laboratory spaces).

Instruments

Guardant360® CDx is intended to be performed with serial number-controlled instruments as indicated in Table 4. All instruments are qualified by Guardant Health, Inc. under the Guardant Health Quality System.

Table 4. Serial Number Controlled Instruments for use with the Guardant360® CDx assay

Instrument
Agilent Technologies 4200 TapeStation Instrument
Hamilton Company Microlab STAR
Hamilton Company Microlab STARlet
Illumina NextSeq 550 Sequencer
Veriti 96-Well Thermal Cycler

Test Process

A. Whole Blood Collection and Shipping

The Guardant360® CDx Blood Collection Kit is used by ordering laboratories / physicians to collect whole blood specimens and ship them to the Guardant Health Clinical Laboratory. A minimum of 5 mL whole blood must be received in order to achieve optimal performance for the Guardant360® CDx assay. Underfilling of tubes with less than 5 mL of blood may lead to incorrect analytical results or poor product performance.

B. Plasma Isolation and cfDNA Extraction

Whole blood specimens are processed in the Guardant Health Clinical Laboratory within 7 days of blood collection. Plasma is isolated from both tubes of whole blood via centrifugation. One tube of plasma is stored, while the second tube is used for cfDNA extraction using the QIAGEN QIAsymphony SP Instrument and reagent system. The resulting cfDNA is quantified using the 4200 TapeStation. Input amounts ranging from 5 to 30 ng of cfDNA are further processed for each sample.

C. Library Preparation and Enrichment

Reagents from the Guardant360® CDx Sample Preparation Kit are used during library preparation, enrichment, enrichment wash, and quantitation steps using the Veriti 96-Well Thermal Cycler, Microlab STAR and STARlet, and 4200 TapeStation Instruments. During library preparation, cfDNA fragment ends are repaired and library adapters containing inline barcodes are attached using blunt-end ligation. The resulting DNA is amplified by PCR to create libraries suitable for enrichment.

Amplified libraries are enriched for genes of interest using hybrid target capture with custom biotinylated RNA probes. Each enriched library is amplified by PCR using a unique index primer that also contains a sequencing flow cell attachment sequence. Amplified enriched libraries are pooled in equimolar amounts, denatured, and diluted to appropriate concentration for sequencing.

D. DNA Sequencing

Paired-end sequencing by synthesis is performed with the Illumina NextSeq 550 Sequencing system. The amplified cfDNA is analyzed by parallel sequencing of amplified target genes to an average depth of coverage of greater than 2,700 unique molecules.

E. Data Analysis and Reporting

The Guardant360® CDx Software uses a custom-developed analysis bioinformatics pipeline (BIP) software module. The BIP software module uses the raw data (output) from the targeted sequencing, partitions the data based on the sample index sequence (barcode) of each read to separate reads originating from individual samples, and executes a proprietary algorithmic reconstruction of the digitized sequencing signals based on molecular barcodes for high-fidelity molecule-based alteration calling downstream. The sequence data then undergoes an alignment process where it is mapped to the human genome (hg19) and an analysis of sequence alteration data is performed.

Alteration detection is conducted according to alteration calling metrics derived from clinical sample analysis. All alterations must pass alteration calling metrics as described in Table 5.

The SNV and indel cut-offs are defined in terms of mutant allele fraction (MAF) estimate, number and type of molecules supporting the alteration, pseudo-gene assessment, and likelihood ratio (LLR) score. The MAF estimate describes the calculated allelic fraction of an SNV or indel. The number of molecules describes the observed number of molecules meeting requirements for a particular alteration call. The LLR score is a calculated number that reflects how much observed support for the mutation exceeds expectations based on PCR and sequencing induced artifacts.

Table 5. Alteration Analytical Calling Threshold/Cut-Off Metrics

SNV Calling Property	Metric
DNA Molecule Support	≥ 2
MAF Estimate	$\geq 0.001\%$
Log Likelihood Ratio	≥ 0
Indel Calling Property	**Metric**
DNA Molecule Support	≥ 2
MAF Estimate	$\geq 0.01\%$
Log Likelihood Ratio	≥ 10

CNA Calling Property	Metric
ERBB2 copy number	≥ 2.18
ERBB2 Z-score	≥ 10
ERBB2 amplification is not associated with chromosome-arm aneuploidy	TRUE
MET copy number	≥ 2.16
MET Z-score	≥ 10
MET amplification is not associated with chromosome-arm aneuploidy	TRUE
Fusion Calling Property	**Metric**
MAPQ score of supporting molecule to fusion sequence	> 30
Number of unique fusion molecules	≥ 2
Number of unique fusion reads	> 2

The laboratory and physician receive a qualitative alteration-level result. A sample will receive an overall "Failed" result when any QC metric is failed. Samples failing any QC metric are automatically held and not released. The laboratory may attempt to rerun a patient sample that has failed a QC metric by using stored plasma or intermediate products.

Results from samples passing all QC metrics are formatted onto an IVD results report with CDx relevant information (Category 1) and all other biomarkers (Categories 2-4) within the LIMS system. The IVD results report will be populated with patient-specific information and may be merged with additional information provided by Guardant Health as a professional service prior to approval and release by the laboratory director or designee.

F. Quality Control Measures

The Guardant360® CDx Sample Preparation Kit includes the Variant Control, which is engineered to contain known positive and negative alterations and is treated as a sample. Additionally, a no template negative control (NTC) is run in parallel with patient samples.

The Variant Control consists of a mixture of cfDNA from multiple human cancer cell lines containing all four alteration types, SNVs, indels, CNAs and fusions. The control is treated as a sample and processed starting from 15 ng cfDNA input through sequencing where it is analyzed for the presence and absence of the specific alterations.

Although the Variant Control does not contain all the alterations that the test is capable of detecting, concordant detection of alterations targeted in the Variant Control indicates that assay is performing as expected across the panel.

In addition to assessing Variant Control performance within a batch, the test is assessing multiple per-sample in-process and post-sequencing analytical metrics for

each of the patient samples tested. These metrics provide in depth analytical QC information that complements Variant Control performance data and is specific and informative to that sample performance.

The NTC samples are absent of a DNA template, so cfDNA extraction, library preparation, and enrichment steps are expected to result in background level metrics.

VI.   ALTERNATIVE PRACTICES AND PROCEDURES

There is an FDA approved companion diagnostic (CDx) alternative for the detection of *EGFR* exon 19 deletions, *EGFR* L858R, and *EGFR* T790M genetic alterations using cfDNA, for the TAGRISSO® (osimertinib) therapeutic. The approved CDx test is detailed below: for additional details see FDA List of Cleared or Approved Companion Diagnostic Devices at
https://www.fda.gov/MedicalDevices/ProductsandMedicalProcedures/InVitroDiagnostics/ucm301431.htm?source=govdelivery

- cobas® EGFR Mutation Test v2 (Roche Molecular Systems, Inc.)
  - o Technology: Polymerase Chain Reaction (PCR)
  - o Therapy: TAGRISSO® (osimertinib)
  - o Indication: Non-small cell lung cancer (NSCLC)
- FoundationOne® Liquid CDx (F1 Liquid CDx) (Foundation Medicine, Inc.)
  - o Technology: Next Generation Sequencing (NGS)
  - o Therapy: TAGRISSO® (osimertinib)
  - o Indication: Non-small cell lung cancer (NSCLC)

Each alternative has its own advantages and disadvantages. A patient should fully discuss these alternatives with his/her physician to select the method that best meets expectations and lifestyle.

There is no FDA approved CDx alternative for the detection of *EGFR* exon 20 insertions in patients with NSCLC using cfDNA for the RYBREVANT™ (amivantamab-vmjw) therapeutic.

VII.   MARKETING HISTORY

Guardant Health, Inc. initially designed and developed the Guardant360 laboratory developed test (Guardant360 LDT), and the first commercial sample was tested in 2012 to detect the presence of genomic alterations in plasma isolated from whole blood. The Guardant360® CDx was FDA-approved on August 7, 2020 and subsequently commercialized in the USA.

Guardant360® CDx has not been marketed in any foreign country.

VIII.   POTENTIAL ADVERSE EFFECTS OF THE DEVICE ON HEALTH

Failure of the device to perform as expected or failure to correctly interpret test results may lead to incorrect test results, and subsequently, may lead to inappropriate patient

management decisions. Patients with false positive results may undergo treatment with the therapies listed in the intended use statement without clinical benefit and may experience adverse reactions associated with the therapies. Patients with false negative results may not be considered for treatment with the indicated therapies. There is also a risk of delayed results, which may lead to delay of treatment with indicated therapy.

For the specific adverse events that occurred in the clinical studies, please see the RYBREVANT™ (amivantamab-vmjw) FDA approved package insert which is available at Drugs@FDA.

IX.   SUMMARY OF NONCLINICAL STUDIES

A. Laboratory Studies

The primary evidence for supporting the performance of Guardant360® CDx in detecting *EGFR* exon 20 insertion mutations was from the data presented using intended use specimens across all validation studies. In addition to the existing platform-level *EGFR* exon 20 insertion validation results (P200010), analytical accuracy/concordance, limit of detection (LoD), and precision at LoD studies were conducted to support the indication for *EGFR* exon 20 insertion mutations. Further, results from exogenous interference and guardbanding studies that were completed to fulfil the conditions of approval for the original PMA P200010 and that utilized samples carrying *EGFR* exon 20 insertion mutations, were also included to support this PMA supplement.

For Guardant360® CDx platform-level validation (P200010), performance characteristics were established using plasma-derived cfDNA samples from a wide range of cancer types. Each study included CDx variants as well as a broad range of representative alteration types (substitution, indels, CNAs, rearrangements) in various genomic contexts across several genes. The platform validation studies included NSCLC samples with *EGFR* exon 20 insertions. These results from the platform-level validation (P200010) have been leveraged to support Guardant360® CDx detection of *EGFR* exon 20 insertions. (please see Section IX.A.10, Table 33 in Summary of Safety and Effectiveness Data P200010).

Additional validation studies to support expansion of the intended use to include *EGFR* exon 20 insertions are described below.

1. Analytical Accuracy/Concordance

An analytical accuracy study was performed with available plasma clinical specimens (56 *EGFR* exon 20 insertion positive patients and 23 *EGFR* exon 20 insertion negative patients) from NSCLC patients enrolled in the CHRYSALIS clinical trial (refer to Section X.A for study details) to demonstrate the concordance between Guardant360® CDx and an externally validated orthogonal NGS assay for the detection of *EGFR* exon 20 insertions. The clinical trial samples accounted for 76% of the *EGFR* exon 20 insertions observed in the CHRYSALIS clinical study. Due to the low prevalence of some *EGFR* exon 20 insertions and small size of biomarker negative samples from the CHRYSALIS clinical trial, samples from Guardant Health's biobank (128 *EGFR* exon 20

insertion positive samples and 70 *EGFR* exon 20 insertion negative samples) were included in the accuracy study.

A total of 293 patient samples were originally selected for the accuracy study. Of the 293 samples, 13 samples were excluded due to insufficient material for testing and 3 samples were excluded due to issues with inclusion criteria to the CHRYSALIS study or since they did not meet the diagnostic study inclusion criteria. Among the remaining 277 samples, five samples were excluded from the study; four samples failed testing with the comparator assay due to sequencing failures, while one sample failed testing with Guardant360® CDx due to enrichment QC metrics.

A summary of positive percent agreement (PPA) and negative percent agreement (NPA) and corresponding 95% two-sided exact confidence intervals (CIs) is provided in Table 6. Since the samples were selected from different sources based on different assays, the unadjusted agreements in Table 6 may be subject to potential bias. Positive agreement was 96.30% while negative agreement was 86.91% for detection of *EGFR* exon 20 insertions in the concordance analysis. It was noted that the discordance was due to sensitivity differences between Guardant360® CDx and the comparator assay.

Table 6. Summary of Concordance Between Guardant360® CDx and NGS Comparator for *EGFR* Exon 20 Insertions

Alteration Type	Guardant360 CDx(+), Comparator #2 (+)	Guardant360 CDx(+), Comparator #2 (-)	Guardant360 CDx(-), Comparator #2 (+)	Guardant360 CDx(-), Comparator #2 (-)	Patients (n)	PPA (95% CI)	NPA (95% CI)	PPV (95% CI)	NPV (95% CI)
EGFR exon 20 insertions	78	25	3	166	272	96.30% (89.56%, 99.23%)	86.91% (81.29%, 91.35%)	75.73% (66.29%, 83.64%)	98.22% (94.9%, 99.63%)

PPA, Positive Percent Agreement; NPA, Negative Percent Agreement; PPV, Positive Predictive Value; NPV, Negative Predictive Value

To further differentiate between comparator assay false negatives and Guardant360® CDx false positives as the origin for the 25 Guardant360® CDx positive comparator negative samples, the agreement between Guardant360® CDx and the comparator assay was calculated for each sample source independently (Table 7). All Guardant360® CDx positive comparator negative samples were found in the Guardant Health (GH) Biobank-Pos and Clinical Validation (CV) Local Test cohorts, which demonstrated challenged NPA values of 83.1% (54/65) and 63.2% (24/38), respectively. As the CV_LocalTest cohort was predominantly composed of samples with tissue testing results, it was expected that some of

these could have lower circulating tumor amounts resulting in more stochastic detections by the comparator reducing the NPA relative to the GH-Biobank-Pos cohort. These NPA values were in marked contrast to the GH-Biobank-Unselected and CV_Sensitivity populations, which comprised no Guardant360® CDx positive comparator negative samples and demonstrated an NPA of 100% (68/68) and 100% (20/20), respectively. The CIs for GH-Biobank-Pos and CV_LocalTest overlapped and were below the CI for GH-Biobank-Unselected. The analysis showed that NPA was similar between the GH-Biobank-Pos and CV_LocalTest cohorts, which had significantly lower NPA than the GH-Biobank-Unselected and CV_Sensitivity cohorts.

Table 7. Summary of Concordance Between Guardant360® CDx and NGS Comparator for EGFR Exon 20 Insertions by Data Cohort

Sample Cohort	Guardant360 CDx(+), Comparator #2(+)	Guardant360 CDx(+), Comparator #2 (-)	Guardant360 CDx(-), Comparator #2 (+)	Guardant360 CDx(-), Comparator #2(-)	Patients (n)	PPA (95% CI)	NPA (95% CI)	PPV (95% CI)	NPV (95% CI)
Guardant Health Biobank-Pos	61	11	1	54	127	98.39% (91.34%, 99.96%)	83.08% (71.73%, 91.24%)	84.72% (74.31%, 92.12%)	98.18% (90.28%, 99.95%)
CV_Local Test	16	14	2	24	56	88.89% (65.29%, 98.62%)	63.16% (45.99%, 78.19%)	53.33% (34.33%, 71.66%)	92.31% (74.87%, 99.05%)
CV_Sensitivity	1	0	0	20	21	100% (2.5%, 100%)	100% (83.16%, 100%)	100% (2.5%, 100%)	100% (83.16%, 100%)
Guardant Health Biobank-Unselected	0	0	0	68	68	NaN% (0%, 100%)	100% (94.72%, 100%)	NaN% (0%, 100%)	100% (94.72%, 100%)

NaN, Not a Number

2. Analytical Sensitivity

a. Limit of Blank (LoB)

Please refer to the Summary of Safety and Effectiveness Data P200010 (Section IX.A.3.a) for Guardant360® CDx platform-level analytical sensitivity data for LoB. There were no false positives for *EGFR* exon 20 insertions.

b. Limit of Detection (LoD)

The LoD for *EGFR* exon 20 insertions was established panel-wide as 0.8% and 0.2% MAF for 5 ng and 30 ng cfDNA input levels, respectively, using a single *EGFR* exon 20 insertions variant, p.A767_V769dup. This result was included in P200010 for Guardant360® CDx (refer to P200010 SSED Section IX.A.3 for details). Because the LoD for *EGFR* exon 20 insertion, A767_V769dup was established using pools of cfDNA from clinical plasma samples from multiple cancer types, the LoD was confirmed in NSCLC patient samples for this indication.

To supplement the existing LoD study, Guardant Health, Inc. performed additional LoD establishment and confirmation studies using NSCLC patient samples that were representative of varying prevalence and insertions lengths of *EGFR* exon 20 insertions as observed in the CHRYSALIS clinical study (refer to Section X for clinical study details). Three sample pools harboring three *EGFR* exon 20 insertions; *EGFR* p.A767_V769dup (9 bp insertion, Pool 1), *EGFR* p.N771_H773dup (9 bp insertion, Pool 2), and *EGFR* p.H773dup (3 bp insertion, Pool 3) were prepared from NSCLC clinical sample cfDNA identified from Guardant Health's Biobank. These variants accounted for 39% of the *EGFR* exon 20 insertions observed in the CHRYSALIS clinical trial. Pools 2 and 3 were used for LoD establishment, while Pool 1 was used for the LoD confirmation study. The presence of *EGFR* exon 20 insertions in each source pool and corresponding mutant allele frequencies (MAFs) were confirmed by a validated NGS comparator method. Patient cfDNA samples positive for selected *EGFR* exon 20 insertions were diluted with mutation-negative cfDNA derived from NSCLC clinical samples ("WT cfDNA") to target the highest MAF level of the source pool. Pool 2 was subsequently diluted serially to form a titration series consisting of 5 different MAF levels at 5 and 30 ng cfDNA inputs. Pool 3 LoD estimation was also done similar to Pool 2 except the study included only 5 ng cfDNA input level. The LoDs were established using 21 replicates for 5 ng cfDNA input and 14 replicates for 30 ng cfDNA input at each titration point. LoD confirmation for *EGFR* p.A767_V769dup was performed using 21 and 14 replicates at target MAFs of 1.4% for 5 ng input and 0.3% for 30 ng input, respectively.

The LoD results for selected *EGFR* exon 20 insertions varied between 0.8% to 1.8% MAF (median 1.2% MAF) for 5 ng cfDNA input, while they were between 0.2% to 0.3% MAF (median 0.3% MAF) for 30 ng cfDNA input level. LoD establishment and confirmation results are summarized in Table 8.

Table 8. Summary of Established and Confirmed LoDs for *EGFR* Exon 20 Insertions in NSCLC Clinical Samples

Alteration	Alteration Type	LoD, 5ng input (MAF)	LoD, 30 ng input (MAF)
EGFR exon 20 insertions	H773dup (3 bp)	0.9% MAF	N.D.
	A767_V769dup (9 bp)*	1.4% MAF	0.3% MAF
	N771_H773dup (9 bp)	1.8% MAF	0.3% MAF
	A767_V769dup (9 bp)**	0.8% MAF	0.2% MAF

N.D.: not determined, * Confirmed LoD, ** Established LoD from mixed cancer types.

Given that not all representative *EGFR* exon 20 insertion mutations that had a high prevalence in the clinical study were evaluated in the LoD study, a post-market study is planned with additional samples harboring 6 bp and 12 bp *EGFR* exon 20 insertions (see section XIII).

3. **Analytical Specificity**

Please refer to the Summary of Safety and Effectiveness Data P200010 (Section IX.A.3) for platform-level validation of analytical specificity, including endogenous and microbial interfering substance and *in silico* hybrid capture bait specificity for Guardant360® CDx.

a. **Exogenous Interfering Substances**

The purpose of the study was to evaluate the effect of cfDNA extraction kit wash buffer carry-over on the performance of Guardant360® CDx assay.

Two cfDNA clinical sample pools derived from a total of 74 clinical cfDNA samples were used in the study. The pools were prepared using proportions for 5 ng cfDNA input. The sample pools were comprised of clinical cfDNA (pool 1) and a mixture of clinical cfDNA and cell line-derived DNA (pool 2) with 10 known variants diluted in WT clinical cfDNA to 1 – 2x LoD (Table 9). The sample pools were spiked with 10% (v/v) of the wash buffer. In the study, a total of 24 replicates (6 replicates/pool x 2 experimental combinations x 2 sample pools), 1 reagent lot, 3 instrument combinations, and 2 operators were used.

Table 9. The characteristics of the 10 variants used in the Exogenous Interference study

Pool	Variant Type	Variant	Adequacy of MAF / CN level in relationship to the LoD (5ng), as assessed by Guardant360 CDx		
			Mean of Reference Condition (MAF% or copy number)	LoD (MAF% or copy number)	Fold over LoD
Pool 1	SNVs	*EGFR* L858R	3.00	1.5	2.0
		EGFR T790M	2.03	1.4	1.4
		KRAS G12C	2.88	1.8	1.6
	Indels	*EGFR* E746_A750del	2.43	1.5	1.6
		EGFR A767_V769dup	0.79	0.8	1.0
	CNA	*MET*	2.87	2.4	2.2
Pool 2	SNVs	*PIK3CA* E545A	4.11	2.4	1.7
		PIK3CA H1047R	3.10	1.7	1.8
	Fusions	*EML4-ALK*	2.77	1.4	2.0
		TPM3-NTRK1	2.86	0.9	3.2

The study result was considered valid if the pre-specific acceptance criteria for Qualitative Detection Rate (QDR) and NPA were met for test and reference conditions. For each test condition, equivalency was concluded if either QDR for the test condition was not significantly lower than the corresponding reference condition by a two-sample test for equality of proportions using a chi-square statistic (one-sided, alpha = 0.05) or the lower limit of the Exact 95% CI for QDR exceeded 80%. For each condition (test and reference), the acceptance criteria for NPA at targeted variant sites was 100%.

Reference condition showed a QDR of 98.3%, with one variant (*EGFR* exon 20, A767_V769dup) missing in one sample. The test condition showed a 100% QDR. For each condition, the lower limits of the 95% confidence interval were higher than 80% meeting the acceptance criteria. The chi-square test also showed a non-significant difference of QDRs (p-value = 0.49) between the test condition and reference condition. Per-sample NPA was 100% for both conditions. All acceptance criteria were met. The exogenous interfering substance study results are summarized in Tables 10 and 11.

Table 10. Summary of QDR analysis results for exogenous interfering substance study for Guardant360® CDx

QDR*		Reference	Test
Exogenous Interference (Wash Buffer)	Positive detected variants across samples	59	64
	QDR	59/60 = 98.33%	64/64 = 100%
	95% CI	[91.06%, 99.96%]	[94.40%, 100%]

*The Quantitative Detection Rate (QDR) is defined as the number of positively detected targeted variants tested across eligible samples (D) divided by the total number of targeted variants tested across eligible samples (N) expressed as a percentage (100 * D / N).

Table 11. Per-sample NPA analysis results for exogenous interfering substance study for Guardant360® CDx

Study	Condition	per-sample NPA
Exogenous Interference (Wash Buffer)	Reference	12/12 = 100%
	Test	13/13 = 100%

The exogenous interfering substance study results fulfilled the condition of approval #3 in Section XIII of P200010.

4. Precision

Please see the Summary of Safety and Effectiveness Data for P200010 (Section IX.A.5) for Guardant360® CDx platform-level validation of precision, including precision from cfDNA pools and precision from plasma extraction.

a. Precision from mutation positive samples

The purpose of the precision study was to demonstrate the repeatability and within-site reproducibility of Guardant360® CDx through closeness of agreement between measured qualitative output obtained in replicate testing using different combinations of three reagent lots, two instruments, four operators, and more than three testing days for detection of *EGFR* exon 20 insertions using NSCLC clinical sample pools. Variant source pools were prepared by diluting NSCLC patient cfDNA samples positive for selected *EGFR* exon 20 insertions with mutation-negative cfDNA derived from NSCLC clinical samples. The *EGFR* exon 20 insertions used in the precision study are listed in Table 12. Each insertion was tested across six precision combinations at 5 ng input at MAF levels indicated in Table 12.

All CDx alterations demonstrated acceptable precision with PPA ranging from 97.6% to 100.0%. The combined precision for *EGFR* exon 20 insertion variant category was 98.4% with 95% CI of 94.3% - 99.8% (Table 13).

Table 12. *EGFR* Exon 20 Insertions with LoD Values Represented in Precision Study

EGFR Variant	Insertion Length	LoD at 5 ng	Precision (fold change to target)
A767_V769dup	9 bp	1.4% MAF	1.4% MAF (1X)
H773dup	3 bp	0.9% MAF	0.9% MAF (1X)
N771_H773dup	9 bp	1.8% MAF	1.8% MAF (1X)
		1.8% MAF	1.25% MAF (0.7X)

Table 13. Precision Study Summary

EGFR Exon 20 Insertion	Number Positive / Number Expected	PPA (95% CI)
A767_V769dup	41 / 42	97.6% [87.4% - 99.9%]
N771_H773dup	41 / 41	100% [91.4% - 100%]
H773dup	41 / 42	97.6% [87.4% - 99.9%]
Overall [95% CI]	**123 / 125**	**98.4% [94.3% - 99.8%]**

b. Precision from mutation negative samples

Samples from healthy donors were pre-screened by an externally validated orthogonal method. Mutation negative samples by the orthogonal method were tested by Guardant360® CDx in three reproducibility conditions (i.e., different reagent lots, operators, instruments, and days). Four replicates from each donor were tested with Guardant360® CDx across the different reproducibility conditions, with a total of 240 replicates evaluated for the study. Within-condition and between-condition Average Negative Agreement (ANA) values were 100.0% for *EGFR* exon 20 insertions.

5. Carryover/Cross-Contamination

Please refer to the Summary of Safety and Effectiveness Data of P200010 (Section IX.A.6) for platform-level carryover/cross-contamination data for Guardant360® CDx.

6. **Reagent Lot Interchangeability**

   Please refer to the Summary of Safety and Effectiveness Data P200010 (Section IX.A.7) for platform-level reagent lot interchangeability data for Guardant360® CDx.

7. **Stability**

   Please refer to the Summary of Safety and Effectiveness Data of P200010 (Section IX.A.8) for platform-level stability data including reagent stability, whole blood stability, plasma stability, cfDNA stability, and intermediate sample stability for Guardant360® CDx.

8. **Guardbanding/Robustness**

   The study established the tolerance of Guardant360® CDx to variation in critical parameters; namely, cfDNA input, adapter volume, hybridization time, and hybridization wash buffer temperature guardbanding (Table 14). A total of 75 clinical cfDNA samples and two cancer cell lines were used to create the 2 sample pools. The sample pools included clinical cfDNA (Pool 1) and a mixture of clinical cfDNA and cell line-derived cfDNA (Pool 2) with 10 known variants diluted in WT clinical cfDNA targeted at 1 – 2x LoD for 5ng and 30ng input cfDNA amounts (Table 15). The study included a total of 126 replicates (6 replicates/pool x 8 experimental combinations x 2 sample pools and 5 replicates/pool x 3 experimental combinations x 2 sample pools), 2 reagent lots, 7 instrument combinations, and 3 operators. The QDR and NPA for test and reference conditions were estimated.

   Guardbanding conditions were evaluated based on the rate of positive agreement for detection of variants in Pool 1 and 2, expressed as the QDR. The study results were considered valid if the pre-specific acceptance criteria for QDR and NPA were met for test and reference conditions. The pre-specified QDR was 95.0% (95% CI: 83.0% - 99.4%). The QDR analysis was performed using a chi-square statistic (one-sided, alpha = 0.05) or the lower limit of the Exact 95% CI for QDR greater than 80%. The NPA at targeted variant sites was set as 100%.

Table 14. Guardbanding Study Overview for Guardant360® CDx

Guardbanding Condition	Reference condition	Condition 1	Condition 2
cfDNA Input amount	5 ng	2.5 ng	4 ng
cfDNA Input amount	30 ng	36 ng	45 ng
Adapter volume	18.0 µL	16.2 µL	19.8 µL
Hybridization Time	12 hours	24 hours	N/A

Wash Buffer Temperature	71°C	70°C	72°C

N/A, Not Applicable.

Table 15. The Characteristics of the 10 Variants Used in the Guardbanding Study for Guardant360® CDx

Pool	Variant Type	Variant	Adequacy of MAF / CN level in relationship to the LoD (5ng), as assessed by Guardant360 CDx			Adequacy of MAF / CN level in relationship to the LoD (30ng), as assessed by Guardant360 CDx		
			Observed Mean (MAF% or copy number) in Reference	LoD (MAF% or copy number)	Fold change over LoD	Observed Mean (MAF% or copy number) in Reference	LoD (MAF% or copy number)	Fold change over LoD
Pool 1	SNVs	*EGFR* L858R	3.27	1.5	2.2	0.31	0.2	1.5
		EGFR T790M	2.40	1.4	1.7	0.49	0.2	2.4
		KRAS G12C	3.56	1.8	2.0	0.84	0.5	1.7
	Indels	*EGFR* E746_A750del	2.96	1.5	2.0	0.36	0.2	1.8
		EGFR A767_V769dup	1.05	0.8	1.3	0.28	0.2	1.4
	CNA	*MET*	3.10	2.4	2.8	3.16	2.4	2.9
Pool 2	SNVs	*PIK3CA* E545A	3.45	2.4	1.4	0.33	0.4	0.8
		PIK3CA H1047R	3.26	1.7	1.9	0.42	0.3	1.4
	Fusions	*EML4-ALK*	3.00	1.4	2.1	0.46	0.2	2.3
		TPM3-NTRK1	1.88	0.9	2.1	0.5	0.2	2.5

For each tested guardbanding condition, all the lower limits of 95% CI were higher than 80%, meeting the acceptance criteria. The chi-square test was performed only between the 4 ng condition and reference condition regarding guard banding testing of cfDNA input at 5 ng, and it showed a non-significant difference of QDRs (p-value = 0.59) between the 4 ng input condition and reference condition (Table 16). The per-sample NPA at targeted variants across all conditions were 100% (Table 17).

Table 16. Summary of Guardbanding Study using QDR Analysis

	QDR	Reference (18.0 µL)	Guardbanding Condition 1 (16.2 µL)	Guardbanding Condition 2 (19.8 µL)
Adaptor Volume	Positive detected variants across samples	56	60	50
	QDR	56/56 = 100%	60/60 = 100%	50/50 = 100%
	95% CI	[93.62%, 100%]	[94.04%, 100%]	[92.89%, 100%]
	QDR	**Reference (30 ng)**	**Guardbanding Condition 1 (36 ng)**	**Guardbanding Condition 2 (45 ng)**
cfDNA input at 30 ng	Positive detected variants across samples	50	46	50
	QDR	50/50 = 100%	46/46 = 100%	50/50 = 100%
	95% CI	[92.89%, 100%]	[92.29%, 100%]	[92.89%, 100%]
	QDR	**Reference (5ng)**	**Guardbanding Condition 1 (4 ng)**	
cfDNA input at 5 ng	Positive detected variants across samples	56	35	N/A
	QDR	56/56 = 100%	35/36 = 97.22%	N/A
	95% CI	[93.62%, 100%]	[85.47%, 99.93%]	N/A
	QDR	**Reference (12 hours)**	**Guardbanding Condition 1 (24hours)**	
Hybridization Time	Positive detected variants across samples	56	60	N/A
	QDR	56/56 = 100%	60/60 = 100%	N/A
	95% CI	[93.62%, 100%]	[94.04%, 100%]	N/A
	QDR	**Reference (71°C)**	**Guardbanding Condition 1 (70°C)**	**Guardbanding Condition 2 (72°C)**
Wash Buffer Temperature	Positive detected variants across samples	56	60	60
	QDR	56/56 = 100%	60/60 = 100%	60/60 = 100%
	95% CI	[93.62%, 100%]	[94.04%, 100%]	[94.04%, 100%]

N/A, Not Applicable

Table 17. Summary of Guardbanding Study using NPA Analysis per sample

Study	Condition	per-sample NPA
Adaptor Volume	Reference (18.0 μL)	11/11 = 100%
	Condition 1 (16.2 μL)	12/12 = 100%
	Condition 2 (19.8 μL)	10/10 = 100%
cfDNA input at 30 ng	Reference (30 ng)	10/10 = 100%
	Condition 1 (36 ng)	9/9 = 100%
	Condition 2 (45 ng)	10/10 = 100%
cfDNA input at 5ng	Reference (5 ng)	11/11 = 100%
	Condition (4 ng)	6/6 = 100%
Hybridization Time	Reference (12 hours)	11/11 = 100%
	Test condition (24 hours)	12/12 = 100%
Wash Buffer Temperature	Reference (71°C)	11/11 = 100%
	Condition 1 (70°C)	12/12 = 100%
	Condition 2 (72°C)	12/12 = 100%

The results of the guardbanding study showed robustness of Guardant360® CDx to variations in the device's workflow and therefore fulfilled the condition of approval #2 in Section XIII for P200010.

9. **General Lab Equipment and Reagent Evaluation**

   a. **cfDNA Extraction**

   Please refer to the Summary of Safety and Effectiveness Data of P200010 (Section IX.A.9.a) for cfDNA extraction performance data for Guardant360® CDx.

   b. **Other Instruments and Reagents**

   Please refer to the Summary of Safety and Effectiveness Data of P200010 (Section IX.A.9.b) for Other Instruments and Reagents data for Guardant360® CDx.

B. Animal Studies

No animal studies were conducted using Guardant360® CDx.

C. Additional Studies

None.

X. SUMMARY OF PRIMARY CLINICAL STUDIES

The clinical performance of Guardant360® CDx for detecting *EGFR* exon 20 insertions in NSCLC patients who may benefit from treatment with RYBREVANT™ (amivantamab-vmjw) (Table 1), was demonstrated through a clinical bridging study using specimens from patients screened for enrollment into the CHRYSALIS (Janssen EDI1001 or 61186372EDI1001 or NCT02609776) study.

A. Guardant360 CDx Clinical Bridging Study for *EGFR* Exon 20 Insertions

The safety and effectiveness of Guardant360® CDx for detecting *EGFR* exon 20 insertions in NSCLC patients who may benefit from treatment with amivantamab-vmjw was demonstrated in a retrospective analysis of plasma specimens from patients enrolled in the CHRYSALIS study. A bridging study was conducted to assess: 1) the concordance between *EGFR* exon 20 insertion status (biomarker positive or negative) tested with the clinical trial enrollment assays and Guardant360® CDx in the intent-to-test population and 2) the clinical efficacy of Guardant360® CDx in identifying *EGFR* exon 20 insertion positive patients for treatment with amivantamab-vmjw monotherapy.

1. Therapeutic Study Design

The CHRYSALIS clinical study is a first-in-human, open-label, multicenter, 2-part, Phase 1 study in patients with metastatic NSCLC having an in-frame base pair insertion mutation in *EGFR* exon 20 whose disease has progressed on or after platinum-based chemotherapy. The primary amivantamab-vmjw registration population comprised *EGFR* exon 20 insertion mutation-positive subjects from the CHRYSALIS clinical study whose disease progressed on or after platinum-based chemotherapy and who were treated with the recommended phase 2 dose (RP2D) of amivantamab-vmjw. Patients were enrolled based on the presence of *EGFR* exon 20 insertions in their tumor or plasma specimens as determined by CLIA-certified local laboratory testing. This clinical study was used to support the approval of RYBREVANT™ (amivantamab-vmjw) under BLA 761210.

The primary efficacy endpoint of the study was overall response rate (ORR) with 95% 2-sided exact CI using Response Evaluation Criteria in Solid Tumors (RECIST v1.1), based on Investigator and Blinded Independent Central Review (BICR) assessments. The secondary endpoints were defined as clinical benefit rate (CBR; confirmed complete response (CR) + confirmed partial response (PR) + stable disease (SD) for at least 11 weeks), duration of response (DOR), progression-free survival (PFS), time to treatment failure (TTF), and overall survival (OS).

The primary efficacy population set to support the BLA 761210 application included 81 subjects. All ongoing BICR assessed responders were followed up for at least 6 months with a median of 9.7 months from the onset of response.

2. <u>Guardant360® CDx *EGFR* Exon 20 Insertions Bridging Study Design</u>

The aim of this bridging study was to determine the concordance between *EGFR* exon 20 insertion results from the clinical trial enrollment assays generated at the time of patient screening for CHRYSALIS and the *EGFR* exon 20 insertion results generated using Guardant360® CDx. The study was also conducted to establish the clinical utility of the Guardant360® CDx assay in identifying *EGFR* exon 20 insertion positive patients for treatment with amivantamab-vmjw monotherapy.

Pre-treatment plasma samples from 78 CHRYSALIS clinical study patients (78/81, 96.3% of the primary registration population) were retrospectively tested with Guardant360® CDx (see sPMA cohort accountability, below). No plasma from the CHRYSALIS clinical study patients negative for *EGFR* exon 20 insertions by local testing was available to represent the local test-negative portion of the Guardant360® CDx-positive intended use population. Therefore, supplemental matched tissue and plasma samples from the CHRYSALIS clinical study screen fail population (non-*EGFR* exon 20 insertion cohorts) and the Noninvasive vs. Invasive Lung Evaluation clinical study (the NILE study, NCT03615443) were used to estimate the prevalence of biomarker-negative patients by local testing to evaluate the potential impact of this population on clinical efficacy.

2.1. Clinical Bridging Study Inclusion and Exclusion Criteria

The criteria for inclusion into the clinical bridging study are summarized below:

- Inclusion criteria for plasma samples from the CHRYSALIS clinical study
  - Subject enrolled in the CHRYSALIS clinical study with informed consent for blood sample use for further research.
  - Subject part of the primary amivantamab-vmjw registration population.
  - Adequate pre-treatment plasma sample available for Guardant360® CDx testing or a previously generated Guardant360® CDx test result from the CHRYSALIS clinical study.
- Inclusion criteria for plasma samples from the CHRYSALIS clinical study for the diagnostic study sensitivity analysis prevalence sub-study
  - Subject failed screening for the CHRYSALIS clinical study with informed consent for blood sample use for further research.
  - Pre-treatment plasma sample available for testing with Guardant360® CDx or a Guardant360® CDx test result previously generated under the Guardant Health CHRYSALIS clinical study protocol.

- Availability of previously generated CHRYSALIS clinical study central tissue testing results.
- Inclusion criteria for samples from the NILE clinical study
  - Subjects enrolled in the NILE clinical study with documented informed consent.
  - A valid Guardant360® CDx test result previously generated from a pre-treatment plasma sample.
  - Previously generated valid test result from cobas® EGFR Mutation Test v2 testing on tissue slides and/or a tissue block of formalin-fixed paraffin-embedded tissue with sufficient tumor content and quantity for testing as defined by central laboratory testing requirements.

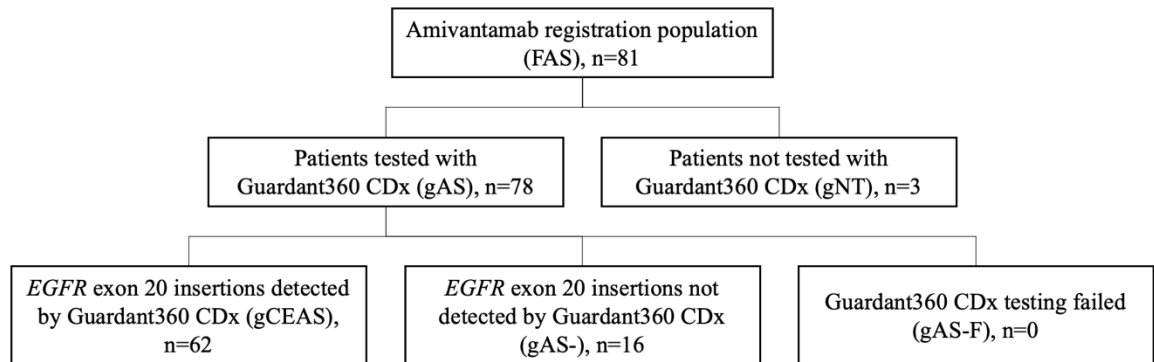2.2. Diagnostic Study Objective and Endpoints

The primary objective of the diagnostic study was to demonstrate the comparability of single-agent amivantamab-vmjw efficacy in primary amivantamab-vmjw registration population subjects who are positive for *EGFR* exon 20 insertions by Guardant360® CDx to the size-adjusted null hypothesis efficacy cited in the CHRYSALIS clinical study protocol. The primary endpoint was ORR by RECIST 1.1 as assessed by BICR.

The possible influence of local test-negative Guardant360® CDx-positive patients not represented in the CHRYSALIS clinical study was assessed through sensitivity analysis. As no plasma samples from CHRYSALIS clinical study patients negative for *EGFR* mutations by local testing were available to represent the local test-negative portion of the Guardant360® CDx-positive intended use population, samples from non-*EGFR* exon 20 cohorts of CHRYSALIS clinical study screen fail population and the NILE clinical study were tested with Guardant360® CDx and either central tissue test (CHRYSALIS clinical study screen fail population) or cobas® EGFR Mutation Test v2, using tissue samples (NILE clinical study population) to calculate the prevalence of this population for the sensitivity analysis.

B. **Accountability of PMA Cohort for the Guardant360® CDx Clinical Bridging Study for *EGFR* Exon 20 Insertions**

The Guardant360® CDx diagnostic study efficacy analysis included 78 of the total 81 (96.3%) patients from the primary amivantamab-vmjw registration population (Figure 1). Of these, 62 patients (76.5% of the primary amivantamab-vmjw registration population) tested positive by Guardant360® CDx while 16 (19.8%) tested negative. Three (3/81, 3.7%) were not tested due to unavailability of plasma specimen for testing, and none failed testing.

Figure 1. Guardant360® CDx *EGFR* Exon 20 Insertions Bridging Study Efficacy Analysis Patient Accountability and Analysis Set Definitions

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                    ┌──────────────────────────────────┐
                    │  Amivantamab registration         │
                    │  population (FAS), n=81           │
                    └──────────────────────────────────┘
                                   │
              ┌────────────────────┴────────────────────┐
   ┌───────────────────────────┐         ┌───────────────────────────┐
   │  Patients tested with     │         │  Patients not tested with │
   │  Guardant360 CDx (gAS),   │         │  Guardant360 CDx (gNT),   │
   │  n=78                     │         │  n=3                      │
   └───────────────────────────┘         └───────────────────────────┘
              │
   ┌──────────┴──────────────────────────────┐
┌──────────────────┐ ┌──────────────────┐ ┌──────────────────────┐
│ EGFR exon 20     │ │ EGFR exon 20     │ │ Guardant360 CDx      │
│ insertions       │ │ insertions not   │ │ testing failed       │
│ detected by      │ │ detected by      │ │ (gAS-F), n=0         │
│ Guardant360 CDx  │ │ Guardant360 CDx  │ │                      │
│ (gCEAS), n=62    │ │ (gAS-), n=16     │ │                      │
└──────────────────┘ └──────────────────┘ └──────────────────────┘
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The Guardant360® CDx diagnostic study assay agreement analysis originally included 268 patients tested with Guardant360® CDx and other test results from both the CHRYSALIS and NILE clinical studies (Figure 2). The agreement analysis set included 97 patients with local test results (9 with plasma testing results, 87 with tissue testing results, 1 with test results using an unknown analyte), 83 screen-fail patients with central tissue test results from other cohorts of CHRYSALIS, and 88 with cobas® EGFR Mutation v2 PCR tissue test results from the NILE study. The additional 19 samples (19/97) included in the positive agreement analysis had the same inclusion criteria as the primary registration population except that these began treatment after the clinical cutoff date and therefore did not have 3 post-baseline disease assessment at the clinical cutoff. The negative agreement analysis cohort did not include samples from the primary registration population, but the 83 samples were screen fails from other arms of the clinical study (non-*EGFR* exon 20 insertion cohorts of CHRYSALIS). Of the 83 screen-fail samples and the 88 samples from the NILE study, 4 and 3 samples, respectively, had *EGFR* exon 20 insertion mutations identified; and, therefore excluded from the negative agreement analysis. The remaining 164 samples were used for negative agreement analysis. The final number of samples used in the agreement analysis was 261.

Central testing for the screen fail samples utilized two different tissue-based NGS tests (69% with FoundationOne® CDx and 31% with Oncomine Dx Target Test) while samples from the NILE study were selected using the tissue-based cobas® EGFR Mutation v2 PCR Test. Overall, the combination of the NILE clinical study and CHRYSALIS non-registration cohorts closely represents the local testing distribution used to enroll the registration population, both in terms of general test methodology (i.e. the registration population 40% PCR, 55% NGS; the supplemental cohorts 51% PCR, 49% NGS) and specific test methodology (i.e. the registration population enrolled by NGS with 35% Oncomine Dx Target Test, 65% FoundationOne® CDx; the supplemental cohorts with 31% and 69% respectively).

Figure 2. Guardant360® CDx *EGFR* Exon 20 Insertions Bridging Study Assay Agreement Analysis Patient Accountability and Analysis Set Definitions. Assay agreement subgroups (AAAS-L, AAAS-C, and AAAS-P) shaded in green.
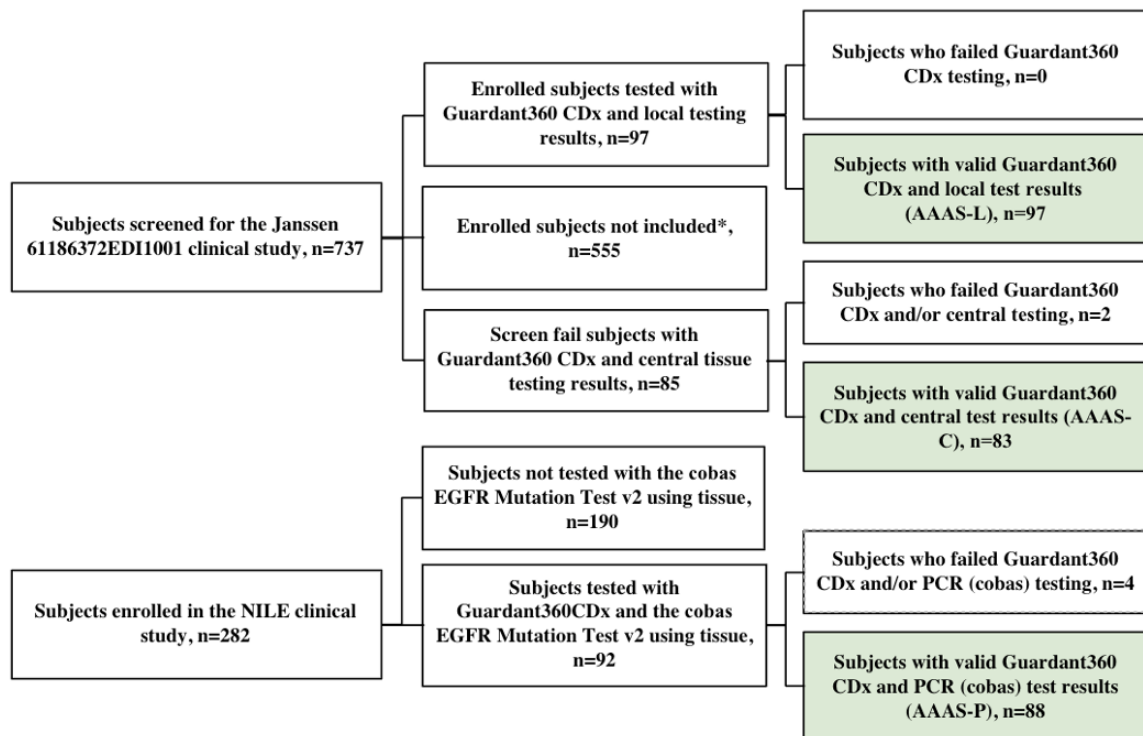
*The CHRYSALIS subjects not included in the assay agreement analysis refers to subjects without the requisite central testing data, from the dose escalation series (non-R2PD-treated), without previous chemotherapy exposure, from additional expansion cohorts, and/or without adequate treatment and/or follow-up.

Abbreviations: AAAS-L, Assay agreement analysis set – Local testing; AAAS-C, Assay agreement analysis set – Central NGS tissue testing; AAAS-P, Assay agreement analysis set – PCR testing

C. **Study Population Demographics and Baseline Parameters for the Guardant360®**
   CDx Clinical Bridging Study for *EGFR* Exon 20 Insertions

Demographics and baseline clinical characteristics of subjects enrolled in CHRYSALIS clinical study were categorized relative to the diagnostic study populations as defined by Guardant360® CDx results. As shown in Tables 18 and 19, the diagnostic study efficacy population (gCEAS) demographics and baseline clinical characteristics closely resemble those of the overall primary amivantamab-vmjw registration population (FAS).

Table 18. Baseline Demographics

	FAS	gAS	gNT	gCEAS	gAS-	gAS-F	gAS-Unk (gAS-F +gNT)	p Value gAS vs gAS-Unk
Analysis set:	81	78	3	62	16	-	3	
Age, years								
N	81	78	3	62	16	0	3	0.914
Mean (SD)	62.3 (9.96)	62.3 (10.04)	61.7 (9.29)	62.5 (10.03)	61.6 (10.40)	-	61.7 (9.29)	
Median	62.0	62.0	59.0	62.0	62.0	-	59.0	
Range	(42; 84)	(42; 84)	(54; 72)	(42; 84)	(46; 76)	-	(54; 72)	

	FAS	gAS	gNT	gCEAS	gAS-	gAS-F	gAS-Unk (gAS-F +gNT)	p Value gAS vs gAS-Unk
<65	48 (59.3%)	46 (59.0%)	2 (66.7%)	38 (61.3%)	8 (50.0%)	-	2 (66.7%)	
>=65	33 (40.7%)	32 (41.0%)	1 (33.3%)	24 (38.7%)	8 (50.0%)	-	1 (33.3%)	
<75	74 (91.4%)	71 (91.0%)	3 (100.0%)	56 (90.3%)	15 (93.8%)	-	3 (100.0%)	
>=75	7 (8.6%)	7 (9.0%)	0	6 (9.7%)	1 (6.3%)	-	0	
Sex								
N	81	78	3	62	16	0	3	1.000
Female	48 (59.3%)	46 (59.0%)	2 (66.7%)	40 (64.5%)	6 (37.5%)	-	2 (66.7%)	
Male	33 (40.7%)	32 (41.0%)	1 (33.3%)	22 (35.5%)	10 (62.5%)	-	1 (33.3%)	
Race								
N	81	78	3	62	16	0	3	0.104
Asian	40 (49.4%)	39 (50.0%)	1 (33.3%)	34 (54.8%)	5 (31.3%)	-	1 (33.3%)	
Black or African American	2 (2.5%)	1 (1.3%)	1 (33.3%)	1 (1.6%)	0	-	1 (33.3%)	
White	30 (37.0%)	29 (37.2%)	1 (33.3%)	21 (33.9%)	8 (50.0%)	-	1 (33.3%)	
Not reported	9 (11.1%)	9 (11.5%)	0	6 (9.7%)	3 (18.8%)	-	0	
Ethnicity								
N	81	78	3	62	16	0	3	1.000
Hispanic or Latino	3 (3.7%)	3 (3.8%)	0	3 (4.8%)	0	-	0	
Not Hispanic or Latino	68 (84.0%)	65 (83.3%)	3 (100.0%)	53 (85.5%)	12 (75.0%)	-	3 (100.0%)	
Not reported	10 (12.3%)	10 (12.8%)	0	6 (9.7%)	4 (25.0%)	-	0	
Weight, kg								
N	81	78	3	62	16	0	3	0.563
Mean (SD)	67.49 (16.784)	67.28 (16.407)	73.03 (29.258)	65.20 (16.149)	75.34 (15.297)	-	73.03 (29.258)	
Median	62.50	62.95	57.10	61.60	73.60	-	57.10	
Range	(35.4; 115.0)	(35.4; 115.0)	(55.2; 106.8)	(35.4; 106.2)	(52.0; 115.0)	-	(55.2; 106.8)	
Height, cm								
N	81	78	3	62	16	0	3	0.504
Mean (SD)	163.71 (9.020)	163.84 (9.044)	160.27 (9.295)	163.12 (9.406)	166.66 (7.034)	-	160.27 (9.295)	
Median	162.60	162.75	154.90	160.05	165.65	-	154.90	
Range	(144.5; 192.0)	(144.5; 192.0)	(154.9; 171.0)	(144.5; 192.0)	(150.0; 176.6)	-	(154.9; 171.0)	
Body mass index, kg/m²								
N	81	78	3	62	16	0	3	0.320
Mean (SD)	24.993 (4.9047)	24.886 (4.8151)	27.776 (7.5866)	24.330 (4.7289)	27.043 (4.6727)	-	27.776 (7.5866)	
Median	24.250	24.508	23.798	23.455	25.858	-	23.798	
Range	(14.00; 36.87)	(14.00; 36.87)	(23.01; 36.52)	(14.00; 36.72)	(19.57; 36.87)	-	(23.01; 36.52)	
Underweight <18.5	4 (4.9%)	4 (5.1%)	0	4 (6.5%)	0	-	0	
Normal 18.5-<25	43 (53.1%)	41 (52.6%)	2 (66.7%)	35 (56.5%)	6 (37.5%)	-	2 (66.7%)	
Overweight 25-<30	21 (25.9%)	21 (26.9%)	0	16 (25.8%)	5 (31.3%)	-	0	
Obese >=30	13 (16.0%)	12 (15.4%)	1 (33.3%)	7 (11.3%)	5 (31.3%)	-	1 (33.3%)	
Local Test Type*								
N	81	78	3	62	16	0	3	0.803
NGS (Blood)	4 (4.9%)	4 (5.1%)	0	3 (4.8%)	1 (6.3%)	-	0	
NGS (Tissue)	34 (42.0%)	33 (42.3%)	1 (33.3%)	24 (38.7%)	9 (56.3%)	-	1 (33.3%)	

	FAS	gAS	gNT	gCEAS	gAS-	gAS-F	gAS-Unk (gAS-F +gNT)	p Value gAS vs gAS-Unk
OTHER (Blood)	1 (1.2%)	1 (1.3%)	0	1 (1.6%)	0	-	0	
OTHER (Tissue)	7 (8.6%)	7 (9.0%)	0	7 (11.3%)	0	-	0	
PCR (Blood)	1 (1.2%)	1 (1.3%)	0	1 (1.6%)	0	-	0	
PCR (Tissue)	30 (37.0%)	28 (35.9%)	2 (66.7%)	23 (37.1%)	5 (31.3%)	-	2 (66.7%)	
UNKNOWN (Tissue)	4 (4.9%)	4 (5.1%)	0	3 (4.8%)	1 (6.3%)	-	0	

Abbreviations: FAS, Full Analysis Set; gAS, Guardant360® CDx analysis set; gNT, Guardant360® CDx not tested set; gCEAS, Guardant360® CDx primary clinical efficacy analysis set; gAS-, Guardant360® CDx analysis set- negative; gAS-F, Guardant360® CDx analysis set- failed; gAS-Unk, Guardant360® CDx unknown set. SD, standard deviation. All percentages calculated using N as denominator.

Table 19. Baseline Clinical Characteristics

	FAS	gAS	gNT	gCEAS	gAS-	gAS-F	gAS-Unk	p Value gAS vs gAS-Unk
Analysis set:	81	78	3	62	16	-	3	
Initial diagnosis NSCLC subtype								
N	81	78	3	62	16	0	3	0.922
Adeno-carcinoma	77 (95.1%)	74 (94.9%)	3 (100.0%)	59 (95.2%)	15 (93.8%)	-	3 (100.0%)	
Large cell carcinoma	0	0	0	0	0	-	0	
Squamous cell carcinoma	3 (3.7%)	3 (3.8%)	0	2 (3.2%)	1 (6.3%)	-	0	
Other	1 (1.2%)	1 (1.3%)	0	1 (1.6%)	0	-	0	
Not reported	0	0	0	0	0	-	0	
Histology grade at initial diagnosis								
N	81	78	3	62	16	0	3	0.708
Moderately differentiated	18 (22.2%)	17 (21.8%)	1 (33.3%)	16 (25.8%)	1 (6.3%)	-	1 (33.3%)	
Poorly differentiated	12 (14.8%)	11 (14.1%)	1 (33.3%)	8 (12.9%)	3 (18.8%)	-	1 (33.3%)	
Well differentiated	5 (6.2%)	5 (6.4%)	0	5 (8.1%)	0	-	0	
Other	46 (56.8%)	45 (57.7%)	1 (33.3%)	33 (53.2%)	12 (75.0%)	-	1 (33.3%)	
Not reported	0	0	0	0	0	-	0	
Cancer stage at initial diagnosis								
N	81	78	3	62	16	0	3	0.078
0	0	0	0	0	0	-	0	
IA	6 (7.4%)	6 (7.7%)	0	4 (6.5%)	2 (12.5%)	-	0	
IB	1 (1.2%)	1 (1.3%)	0	1 (1.6%)	0	-	0	
IIA	1 (1.2%)	1 (1.3%)	0	1 (1.6%)	0	-	0	
IIB	4 (4.9%)	3 (3.8%)	1 (33.3%)	3 (4.8%)	0	-	1 (33.3%)	
IIIA	4 (4.9%)	3 (3.8%)	1 (33.3%)	2 (3.2%)	1 (6.3%)	-	1 (33.3%)	
IIIB	4 (4.9%)	4 (5.1%)	0	3 (4.8%)	1 (6.3%)	-	0	
IV	61 (75.3%)	60 (76.9%)	1 (33.3%)	48 (77.4%)	12 (75.0%)	-	1 (33.3%)	
Not reported	0	0	0	0	0	-	0	

	FAS	gAS	gNT	gCEAS	gAS-	gAS-F	gAS-Unk	p Value gAS vs gAS-Unk
Location of metastasis [a]								
N	81	78	3	62	16	0	3	0.598
Bone	34 (42.0%)	33 (42.3%)	1 (33.3%)	30 (48.4%)	3 (18.8%)	-	1 (33.3%)	
Liver	7 (8.6%)	7 (9.0%)	0	5 (8.1%)	2 (12.5%)	-	0	
Brain	18 (22.2%)	17 (21.8%)	1 (33.3%)	14 (22.6%)	3 (18.8%)	-	1 (33.3%)	
Lymph Node	43 (53.1%)	43 (55.1%)	0	38 (61.3%)	5 (31.3%)	-	0	
Adrenal Gland	3 (3.7%)	3 (3.8%)	0	3 (4.8%)	0	-	0	
Other	45 (55.6%)	42 (53.8%)	3 (100.0%)	31 (50.0%)	11 (68.8%)	-	3 (100.0%)	
Not reported	0	0	0	0	0	-	0	
Time from initial diagnosis of cancer to first dose (months)								
N	81	78	3	62	16	0	3	0.881
Mean (SD)	22.905 (21.1901)	22.835 (21.3828)	24.717 (18.7773)	23.972 (22.8978)	18.427 (13.7407)	-	24.717 (18.7773)	
Median	17.018	16.986	26.021	16.789	18.431	-	26.021	
Range	(1.45; 130.10)	(1.45; 130.10)	(5.32; 42.81)	(2.86; 130.10)	(1.45; 45.37)	-	(5.32; 42.81)	
Time from metastatic disease diagnosis to first dose (months)								
N	81	78	3	62	16	0	3	0.401
Mean (SD)	18.071 (16.4424)	18.374 (16.6647)	10.185 (5.0347)	18.886 (17.4686)	16.388 (13.3918)	-	10.185 (5.0347)	
Median	14.160	14.883	9.856	14.883	14.850	-	9.856	
Range	(0.69; 116.40)	(0.69; 116.40)	(5.32; 15.38)	(0.69; 116.40)	(1.35; 45.37)	-	(5.32; 15.38)	
Number of prior lines of therapy								
N	81	78	3	62	16	0	3	0.614
Mean (SD)	2.3 (1.41)	2.2 (1.40)	2.7 (2.08)	2.3 (1.47)	1.9 (1.06)	-	2.7 (2.08)	
Median	2.0	2.0	2.0	2.0	2.0	-	2.0	
Range	(1; 7)	(1; 7)	(1; 5)	(1; 7)	(1; 4)	-	(1; 5)	
ECOG performance status								
N	81	78	3	62	16	0	3	0.980
0	26 (32.1%)	25 (32.1%)	1 (33.3%)	19 (30.6%)	6 (37.5%)	-	1 (33.3%)	
1	54 (66.7%)	52 (66.7%)	2 (66.7%)	42 (67.7%)	10 (62.5%)	-	2 (66.7%)	
2	1 (1.2%)	1 (1.3%)	0	1 (1.6%)	0	-	0	
>2	0	0	0	0	0	-	0	
Not reported	0	0	0	0	0	-	0	
History of smoking								
N	81	78	3	62	16	0	3	0.631
Yes	38 (46.9%)	37 (47.4%)	1 (33.3%)	25 (40.3%)	12 (75.0%)	-	1 (33.3%)	
No	43 (53.1%)	41 (52.6%)	2 (66.7%)	37 (59.7%)	4 (25.0%)	-	2 (66.7%)	
Unknown	0	0	0	0	0	-	0	

Abbreviations: FAS, Full Analysis Set; gAS, Guardant360 CDx analysis set; gNT, Guardant360® CDx not tested set; gCEAS, Guardant360® CDx primary clinical efficacy analysis set; gAS-, Guardant360® CDx

analysis set- negative; gAS-F, Guardant360® CDx analysis set- failed; gAS-Unk, Guardant360® CDx unknown set; ECOG, Eastern Cooperative Oncology Group; SD, standard deviation. All percentages calculated using N as denominator.

D. Safety and Effectiveness Results for the Guardant360® CDx Clinical Bridging Study for *EGFR* Exon 20 Insertions

1. Safety Results

The safety with respect to treatment with amivantamab-vmjw was addressed during the review of the BLA and is not addressed in this SSED. Please refer to Drugs@FDA for complete safety information on RYBREVANT™ (amivantamab-vmjw). No adverse events were reported in the conduct of the diagnostic studies used to support this PMA supplement as these involved retrospective testing of banked specimens only.

2. Effectiveness Results

a. Concordance Results

Concordance between Guardant360® CDx and predominantly tissue testing in the assay analysis agreement set (AAAS) population, which included subjects with CHRYSALIS clinical study local enrolling test results (for biomarker-positives) to determine the positive percent agreement (PPA) and with central tissue NGS test results and NILE clinical study central tissue PCR test results (representing biomarker-negatives) to determine the negative percent agreement (NPA), is shown in Table 20. Guardant360® CDx demonstrated NPA of 100% (95% CI, 97.7% – 100%), PPA of 80.4, (95% CI, 71.4% – 87.1%) relative to local testing results.

Table 20. Unadjusted Agreement Between Guardant360® CDx and CHRYSALIS Enrollment Testing, CHRYSALIS Central Testing, or NILE clinical study cohort cobas® EGFR Testing

	CHRYSALIS Clinical Study Enrolling or Central Testing or NILE Clinical Study cobas® EGFR Mutation v2 Testing		
	EGFR exon 20 Insertion+	*EGFR* exon 20 Insertion-	Total
Guardant360® CDx			
EGFR exon 20 Insertion+	78	0	78
EGFR exon 20 Insertion-	19	164	183
Total	97	164	261
PPA (95% CI)	80.4% (71.4% - 87.1%)		
NPA (95% CI)	100.0% (97.7% - 100.0%)		

Due to the enrichment of the AAAS-L population for subjects positive for *EGFR* exon 20 insertions, adjusted agreement was assessed using the PPV = P(local test+ | Guardant360® CDx+) and NPV = P(local test– | Guardant360® CDx–) for the total AAAS population. In this analysis, Guardant360® CDx demonstrated high adjusted PPV of 100% (95% CI, 95.6% - 100%) and NPV of 99.6% (95% CI, 99.5% - 99.8%) relative to local testing. The prevalence estimate P(local test+) used in the adjusted agreement was 1.8%.

b. Clinical Efficacy Results in the CHRYSALIS *EGFR* Exon 20 insertion Cohort

The BICR-assessed confirmed ORR as of the 8 October 2020 clinical cutoff date in the *EGFR* exon 20 insertion plus prior chemotherapy at RP2D primary efficacy population was 39.5% (95% CI: 28.8%, 51.0%). Sixty-four (79%) subjects experienced tumor shrinkage of greater than 10% and the clinical activity was observed with amivantamab-vmjw independent of *EGFR* exon 20 insertion subtype. In the primary efficacy population, the BICR-assessed median DOR was 11.1 months (95% CI: 6.90, not estimable [NE]) as of the clinical cutoff date, with the longest response reported as 21.7 months. Amivantamab-vmjw efficacy results are summarized in Table 21.

Table 21. Efficacy Results for CHRYSALIS Clinical Study

	Prior Platinum-based Chemotherapy Treated (N=81)
Overall Response Rate (95% CI)	39.5% (28.8%, 51.0%)
Complete response (CR)	3.7%
Partial response (PR)	35.8%
Duration of Response (DOR)	
Median, months (95% CI) [b]	11.1 (6.9, NE)
Patients with DOR ≥6 months,	63%

[b] Based on Kaplan-Meier estimates.
NE=Not Estimable, CI=confidence interval

As shown in Table 22, in the primary efficacy population, a total of 31 different *EGFR* Exon 20 insertion variants with varying insertion lengths was observed. The most prevalent insertion was *EGFR* A767_V769dup (29.9%) followed by *EGFR* S768_D770dup (16.9%) and *EGFR* H773dup (5.2%).

Table 22. Prevalence of *EGFR* Exon 20 Insertions in CHRYSALIS Primary Efficacy Population

EGFR exon 20 insertion	Prevalence in CHRYSALIS (%)
A767_V769dup	29.9
S768_D770dup	16.9
H773dup	5.2
D770delinsGY	3.9
N771_H773dup	3.9
A763_Y764insFQEA	2.6
D770_N771insGF	2.6
H773_V774insAH	2.6
H773_V774insNPH	2.6
P772_H773dup	2.6
D770_N771delinsASVDN	1.3
D770_N771insG	1.3
D770_N771insGD	1.3
D770_N771insKD	1.3
D770_N771insY	1.3
D770_P772dup	1.3
D771_H773dup	1.3
H773_V774dup	1.3
H773_V774insPH	1.3
H773_V774insPHPH	1.3
H773delinsNPY	1.3
N771_P772insH	1.3
N771_P772insT	1.3
N771_P772insV	1.3
N771delinsGF	1.3
N771delinsGY	1.3
N771delinsKG	1.3
P772_H773insDNP	1.3
P772_H773insPNP	1.3
S768_V769delinsIL	1.3
V769_D770insGVV	1.3

c. ORR in Patients Positive by Guardant360® for *EGFR* Exon 20 Insertions

The clinical validity of Guardant360® CDx for the selection of NSCLC patients with *EGFR* exon 20 insertions for treatment with amivantamab-vmjw was demonstrated by comparing the efficacy of amivantamab-vmjw monotherapy in the BLA registration population who are positive for *EGFR* exon 20 insertions by Guardant360® CDx to the null hypothesis efficacy cited in the CHRYSALIS study protocol. Guardant Health's primary objective analysis set included 62 (76.5% of the CHYRSALIS primary efficacy population) clinical trial samples with valid Guardant360® CDx positive results. The efficacy of amivantamab-vmjw as measured by the ORR observed in the primary objective analysis set (gCEAS) by BICR was 38.7% (95% CI, 26.6% – 51.9%, Table 23). The lower limit of the 95% CI of 26.6% showed statistically significant amivantamab-vmjw efficacy relative to the size-adjusted benchmark ORR of 14% (unadjusted benchmark 15% for chemotherapies in second line treatment setting) from the CHRYSALIS clinical study in the

Guardant360® CDx-positive, local test-positive portion of the intended use population. The study met the prespecified efficacy acceptance criterion. The gCEAS ORR point estimate was also similar to the FAS ORR of 39.5% (95% CI, 28.8% – 51.0%, Table 23). CHRYSALIS efficacy was also assessed by DOR by BICR for the FAS and gCEAS populations. As shown in Table 24, no meaningful differences in DOR by BICR were observed between the populations.

Table 23. Summary of ORR in the gCEAS and FAS by BICR

Analysis set: Efficacy	gCEAS	FAS
Best Overall Response (N)	62	81
Complete Response (CR)	2 (3.2%)	3 (3.7%)
Partial Response (PR)	22 (35.5%)	29 (35.8%)
Stable Disease (SD)	29 (46.8%)	39 (48.1%)
Progressive Disease (PD)	7 (11.3%)	8 (9.9%)
Not Evaluable/Unknown	2 (3.2%)	2 (2.5%)
Overall Response Rate (Confirmed CR + Confirmed PR) (95% CI)	24 (38.7%) (26.6%, 51.9%)	32 (39.5%) (28.8%, 51.0%)
Clinical Benefit Rate (Confirmed CR + Confirmed PR+SD) (95% CI)	43 (69.4%) (56.3%, 80.4%)	60 (74.1%) (63.1%, 83.2%)

Table 24. Summary of DOR in the gCEAS and FAS by BICR

	gCEAS	FAS
Analysis set: Efficacy (N)	62	81
Responders	24	32
Event	12 (50.0%)	14 (43.8%)
Censored	12 (50.0%)	18 (56.3%)
Time to event (months) 25th percentile (95% CI) Median (95% CI) 75th percentile (95% CI)	5.55 (3.94, 8.31) 8.31 (5.55, NE) 21.65 (8.31, NE)	5.55 (4.17, 10.84) 11.14 (6.90, NE) 21.65 (11.14, NE)
Range (months)	(1.3+, 21.7)	(1.3+, 21.7)

d. Sensitivity Analysis

The primary objective analysis above demonstrated amivantamab-vmjw efficacy in the Guardant360® CDx-positive, local test-positive subset of the Guardant360® CDx intended use population. Sensitivity analysis was performed to evaluate the potential impact of samples not available for testing and the hypothetical Guardant360® CDx-positive enrolling test-negative population. The sensitivity analysis was done using the lower

bound estimate of the 95% CI for the Pr(local test+|CDx+), which was 95.6%. Sensitivity analysis modeling efficacy across the entire Guardant360® CDx intended use population using ORR by BICR assessment showed robustness to the contribution of the unrepresented Guardant360® CDx-positive, local test-negative subjects, with estimated ORRs for the overall Guardant360® CDx intended use population highly similar to those observed for both the gCEAS and FAS due to the low observed prevalence (0%) of the Guardant360® CDx-positive, local test-negative population. The lower limits of the 95% CI for the estimated ORRs across all modeled conditions exceeded the size-adjusted benchmark ORR of 14%, which demonstrates statistically-significant amivantamab-vmjw efficacy across the entire Guardant360® CDx intended use population, irrespective of amivantamab efficacy in the modeled hypothetical Guardant360® CDx-positive, local test-negative sub-population.

E. **Pediatric Extrapolation**

In this premarket application, existing clinical data was not leveraged to support approval of a pediatric patient population.

F. **Financial Disclosure**

The Financial Disclosure by Clinical Investigators regulation (21 CFR 54) requires applicants who submit a marketing application to include certain information concerning the compensation to, and financial interests and arrangement of, any clinical investigator conducting clinical studies covered by the regulation. The pivotal clinical study included one investigator who was a full-time of the sponsor and had disclosable financial interests/arrangements as defined in 21 CFR 54.2(a), (b), (c) and (f) and described below:

- Compensation to the investigator for conducting the study where the value could be influenced by the outcome of the study: [0]
- Significant payment of other sorts: [0]
- Proprietary interest in the product tested held by the investigator: [0]
- Significant equity interest held by investigator in sponsor of covered study: [1]

The applicant has adequately disclosed the financial interest/arrangements with clinical investigators. Statistical analyses were conducted by FDA to determine whether the financial interests/arrangements had any impact on the clinical study outcome. The information provided does not raise any questions about the reliability of the data.

XI. **SUMMARY OF SUPPLEMENTAL CLINICAL INFORMATION**

Not applicable.

XII.   PANEL MEETING RECOMMENDATION AND FDA'S POST-PANEL ACTION

In accordance with the provisions of section 515(c)(3) of the act as amended by the Safe Medical Devices Act of 1990, this PMA was not referred to the Molecular and Clinical Genetics Panel, an FDA advisory committee, for review and recommendation because the information in the PMA substantially duplicates information previously reviewed by this panel.

XIII.   CONCLUSIONS DRAWN FROM PRECLINICAL AND CLINICAL STUDIES

A. Effectiveness Conclusions

For the intended use to identify *EGFR* exon 20 insertions in NSCLC patients to be treated with amivantamab-vmjw, the effectiveness of the Guardant360® CDx assay was demonstrated through a clinical bridging study using plasma specimens from patients screened for enrollment into the CHRYSALIS study. The data from the analytical validation and clinical bridging studies support the reasonable assurance of safety and effectiveness of the Guardant360® CDx assay when used in accordance with the indications for use. Data from the CHRYSALIS study show that patients who had qualifying *EGFR* exon 20 insertions received benefit from treatment with amivantamab-vmjw and support the addition of the CDx indication to Guardant360® CDx.

B. Safety Conclusions

The risks of the device are based on nonclinical laboratory studies as well as data collected in clinical studies conducted to support PMA approval as described above. As an *in vitro* diagnostic test for the detection of *EGFR* mutations (Table 1) that can inform treatment selection in a NSCLC patient's blood sample, failure of Guardant360® CDx to perform as expected or incorrect interpretations results may lead to inappropriate patient management decisions in NSCLC treatment. Since a patient with a negative result (including a false negative result) from the Guardant360® CDx will be reflexed to having their *EGFR* status determined from an formalin-fixed paraffin-embedded (FFPE) tissue specimen, the risks of the Guardant360® CDx are largely associated with a false positive result in a patient, who may then undergo treatment with one of the therapies listed in Table 1 of the intended use statement without clinical benefit and may experience adverse reactions associated with the therapy. There is also a risk of delayed results, which may lead to delay of treatment with the indicated therapy.

C. Benefit-Risk Determination

For the *EGFR* exon 20 insertion indication, the probable benefits of Guardant360® CDx are based on data collected in the CHRYSALIS study and the bridging study, which was conducted to support PMA approval. The clinical benefit of the Guardant360® CDx assay for the selection of NSCLC patients with *EGFR* exon 20

insertions was demonstrated in a retrospective analysis of efficacy and safety data obtained from the first-in human, open-label, single-arm Phase I CHRYSALIS study.

Treatment with RYBREVANT™ (amivantamab-vmjw) provides meaningful clinical benefit to patients with NSCLC harboring *EGFR* exon 20 insertions whose disease progressed on or after platinum-based chemotherapy. The benefit of Guardant360® CDx in this indication was demonstrated using archived plasma samples from the CHRYSALIS clinical study. Patients positive for *EGFR* exon 20 insertions by Guardant360® CDx demonstrated an ORR similar to that observed in the overall primary amivantamab-vmjw registration population (38.7% vs. 39.5%) with a lower limit of the 95% confidence interval that exceeded the benchmark ORR of 14%, providing evidence of probable benefit.

There is potential risk associated with the use of this device, mainly due to 1) false positives, false negatives, or failure to provide a result and 2) incorrect interpretation of test results by the user.

The risks of Guardant360® CDx for the selection of NSCLC patients with *EGFR* exon 20 insertions for treatment with RYBREVANT™ (amivantamab-vmjw) are associated with the potential mismanagement of patient's treatment resulting from false results of the test. Patients who are determined to be false positive by the test may be exposed to a drug combination that is not beneficial and may lead to adverse events or may have delayed access to other treatments that could be more beneficial. A false negative result may prevent a patient from accessing a potentially beneficial therapeutic regimen. However, this risk is partially mitigated by reflex testing recommendation for negative results with an FDA-approved tumor tissue test on *EGFR* exon 20 insertion biomarker.

The risks of false results are partially mitigated by the analytical validation results summarized above. In addition, the risks of false negative results are partially mitigated by a recommendation that those patients whose plasma generates a negative result for those alterations included in Table 1 should have their tumor mutation status for Table 1 alterations verified by using an FDA-approved tumor tissue test, if feasible. Though the Guardant360® CDx assay has been analytically validated as summarized above, multiple post-market studies are also planned. The overall clinical and analytical data support that for the Guardant360® CDx assay, and the indications noted in the intended use statement, the probable benefits outweigh the probable risks.

  1. Patient Perspectives
  This submission did not include specific information on patient perspectives for this device.

In conclusion, given the available information above, the data support that for the selection of NSCLC patients with *EGFR* exon 20 insertions for treatment with RYBREVANT™ (amivantamab-vmjw) the probable benefits outweigh the probable risks.

D. Overall Conclusions

The data in this application support the reasonable assurance of safety and effectiveness of this device when used in accordance with the indications for use. Data from the clinical studies support the use of Guardant360® CDx in the identification of patients for whom treatment with the therapies listed in the Intended Use Statement may be indicated.

XIV. CDRH DECISION

CDRH issued an approval order on May 21, 2021. The final conditions of approval cited in the approval order are described below.

Guardant Health, Inc. must provide detailed protocols for the studies that are noted below as conditions of approval. These studies must be adequate to confirm the safety and effectiveness of the Guardant360® CDx device and must include a detailed description of the data and sample sets including sample size and tumor type to be tested, the complete testing protocol, acceptance criteria, and a data analysis plan, as applicable. These protocols must be submitted to FDA no later than 60 days after approval.

1. The limit of detection (LoD) study provided in P200010/S001 included a limited number of CDx positive samples covering the range of prevalent *EGFR* exon 20 insertions. Therefore, to provide a more robust assessment of LoD, Guardant Health, Inc. must provide additional LoD data (LoDs for 6 and 12 base pair insertion variants of *EGFR* exon 20) using clinical samples.

2. Guardant Health, Inc. must provide results from regression tests and associated software documentation for the report module software to confirm that the upgrade associated with *EGFR* exon 20 insertion reporting has no impact on the reporting of the other variants (CDx and tumor profiling) specified in the device intended use.

The final study data, study conclusions, and labeling revisions should be submitted within one (1) year of the PMA approval date.

The applicant's manufacturing facilities have been inspected and found to be in compliance with the device Quality System (QS) regulation (21 CFR 820).

XV. APPROVAL SPECIFICATIONS

Directions for use: See device labeling.

Hazards to Health from Use of the Device: See Indications, Contraindications, Warnings, Precautions, and Adverse Events in the device labeling.

Post-approval Requirements and Restrictions: See approval order.

XVI. REFERENCES

None.