Title: A PHASE 1, OPEN-LABEL, POSITRON EMISSON TOMOGRAPHY (PET) STUDY WITH [18F]MNI-1054 TO DETERMINE LYSINE-SPECIFIC DEMETHYLASE 1A (LSD1) BRAIN ENZYME OCCUPANCY OF TAK-418 AFTER SINGLE-DOSE ORAL ADMINISTRATION IN HEALTHY SUBJECTS

NCT Number: NCT04202497

SAP Approve Date: 19 February 2020

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TAKEDA DEVELOPMENT CENTER

STATISTICAL ANALYSIS PLAN

STUDY NUMBER: TAK-418_0004

A PHASE 1, OPEN-LABEL, POSITRON EMISSON TOMOGRAPHY (PET) STUDY WITH [^{18}F]MNI-1054 TO DETERMINE LYSINE-SPECIFIC DEMETHYLASE 1A (LSD1) BRAIN ENZYME OCCUPANCY OF TAK-418 AFTER SINGLE-DOSE ORAL ADMINISTRATION IN HEALTHY SUBJECTS

PHASE 1 TAK-418 SINGLE DOSE PET TARGET OCCUPANCY STUDY

Version: Final

Date: 19 February 2020

Prepared by:

PPD

2.0 APPROVAL SIGNATURES

Study Title: **A Phase 1, Open-Label, Positron Emission Tomography (PET) Study with [^{18}F]MNI-1054 to Determine Lysine-specific demethylase 1A (LSD1) Brain Enzyme Occupancy of TAK-418 After Single-Dose Oral Administration in Healthy Subjects**

Takeda Approval:

PPD

Date

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4.0 LIST OF ABBREVIATIONS

LSD1	lysine-specific demethylase 1A, also known as KDM1A
ADaM	Analysis Data Model
AE	adverse event
AUC	area under the plasma concentration-time curve
AUC_{scan}	area under the plasma concentration-time curve for TAK-418 during each PET scan
$AUEC_t$	area under the effect-time curve from time 0 to last sampling time point
BLQ	below the limit of quantification
BMI	body mass index
$C_{ave(scan)}$	average plasma concentration during each PET scan period following TAK-418 dosing
C_{max}	maximum observed plasma concentration
C-SSRS	Columbia-Suicide Severity Rating Scale
CTS	Clinical and Translational Science
DLRM	dose level review meeting
ECG	electrocardiogram
E_{max}	maximum observed effect
MAV	markedly abnormal value
MedDRA	Medical Dictionary for Regulatory Activities
MRI	magnetic resonance imaging
PD	pharmacodynamic
PET	positron emission tomography
PK	pharmacokinetic
PT	preferred term
ROI	region of interest
SAE	serious adverse event
SOC	system organ class
SRD	single rising dose
TEAE	treatment-emergent adverse event

5.0 OBJECTIVES

5.1 Primary Objective

The primary objective of the study is to determine the relationship between the occupancy of LSD-1 by TAK-418, following administration of a single oral dose, and the TAK-418 plasma concentration in healthy subjects using [^{18}F]MNI-1054 PET imaging.

5.2 Secondary Objectives

The secondary objective(s) of the study are:

1. To estimate the LSD1 enzyme turnover rate.
2. To acquire safety data following administration of a single oral dose of TAK-418.
3. To acquire safety data following injection of [^{18}F]MNI-1054.

5.3 Study Design

This is a PET imaging study with a single oral dose of TAK-418 and up to 3 administrations of intravenous microdoses of the LSD1 PET radiotracer [^{18}F]MNI-1054. The primary objective to determine brain LSD1 enzyme occupancy and the relationship of occupancy to TAK-418 dose and plasma exposure after single oral dosing of TAK-418 in healthy subjects.

A maximum of 16 evaluable male or female subjects are planned to participate in this study. Within that total number of subjects, up to 5 dose levels may be evaluated with up to 6 subjects per dose level, although typically there will be 2 to 3 subjects per dose level.

Each subject will receive up to 3 dynamic [^{18}F]MNI-1054 PET scans (up to 180 minutes with intermittent breaks) to assess enzyme occupancy and turnover in humans (1 baseline scan and 2 scans after a single dose of TAK-418). It is anticipated that there will be at least a 3-night stay at the overnight confinement unit. Plasma samples will be taken at prescribed intervals to assess the peripheral pharmacokinetics (PK) of TAK-418. A brain magnetic resonance imaging (MRI) scan without gadolinium contrast will be performed as part of the screening visit and used to delineate the anatomical regions of interest (ROIs) for individual PET images.

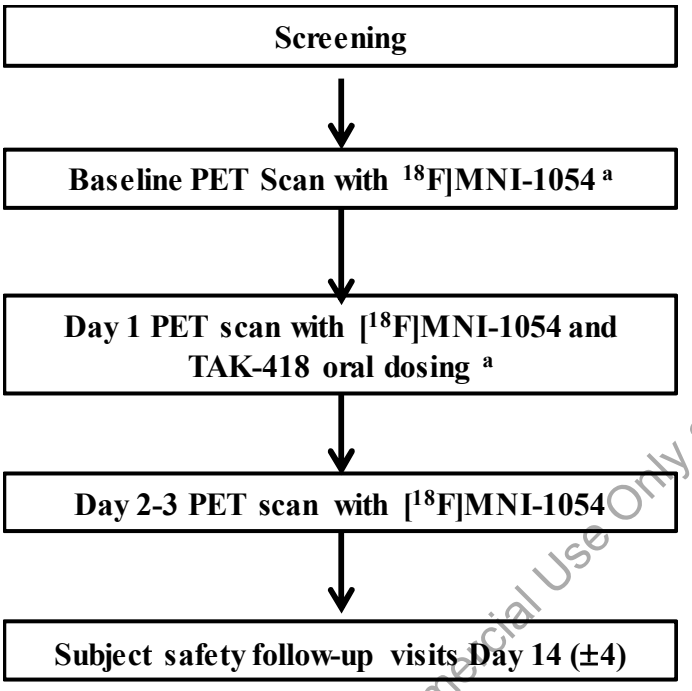
This study will have an adaptive design such that the TAK-418 dose and timing of postdose imaging for subsequent subjects will be based on the data from the previous subjects and determined with input from the sponsor and the clinical site team. For the first 2 subjects, the postdose PET scans will be performed at approximately 6 and 26.5 hours after TAK-418 dosing. Flexibility of approximately ±1 hour is permissible in scan timing. Deviations of greater than this (eg, due to logistical issues such as radiotracer production delays) are permissible if agreed with the sponsor.

5.4 Dose Selection

The starting dose in this study will be 1.5 mg. Dose levels for subsequent subjects may be lower or higher than this and will be selected based on review of the imaging and PK data available at

that point to enable an accurate determination of the exposure-occupancy relationship. Only doses lower than or equal to those that have been tested in the phase 1 safety studies and determined to be safe and well-tolerated will be assessed in this study. The maximum dose of TAK-418 evaluated in phase 1 safety study was 160 mg.

Figure 5.a Schematic of Study Design



PET: positron emission tomography.
[a] Baseline and Day 1 visit may occur over 1 or 2 calendar days.

A schedule of assessments and their timing during the study is listed in Protocol TAK-418-0004 [1].

6.0 ANALYSIS ENDPOINTS

6.1 Primary endpoints

The primary endpoints of the study are:

- Quantitative estimates of binding of [^{18}F]MNI-1054 based on appropriate PET radiotracer kinetic models (eg, K_i from irreversible 2-tissue compartmental model).
- Percent enzyme occupancy calculated from quantitative estimates of binding (= 100 × [baseline – postdose]/baseline).
- Plasma PK parameters including, if feasible, but not limited to:
  - C_{max}.
  - Area under the concentration-time curve from time 0 to time t (AUC_t).
  - AUC_∞.

6.2 Secondary endpoints

Secondary endpoints include:

1. The relationship between percent receptor occupancy calculated from first and second postdose scans.
2. Summary of safety observations, including but not limited to:
   - Number and percentage of participants with 1 or more AEs.
   - Number and percentage of participants with 1 or more SAEs.
   - Number and percentage of participants with clinically defined abnormal laboratory values.
   - Number and percentage of participants with clinically defined abnormal vital signs.

7.0 DETERMINATION OF SAMPLE SIZE

No formal statistical sample size calculation was performed. The sample size of up to 5 dose levels and up to 6 subjects per dose level within the limit of 16 total evaluable subjects is based on precedents of other PET occupancy studies and is considered to be sufficient for evaluation of target occupancy, duration of occupancy, safety, tolerability, and the relationship between occupancy and TAK-418 plasma exposure.

8.0 METHODS OF ANALYSIS AND PRESENTATION

8.1 General Principles

All study-related raw data, including derived data, will be presented in data listings. Continuous data will be summarized using: number of subjects (N), mean, SD, median, minimum, and maximum, where appropriate. Where indicated, coefficient of variation (%CV) and geometric mean will also be included in the summary of continuous data. Categorical data will be summarized using the number and percentage of subjects for each category where appropriate.

8.1.1 Missing Data

There will be no imputation of incomplete or missing data. Decisions regarding inclusion or exclusion of data from an analysis for subjects who are noncompliant with the dose schedule, or who have incomplete data, will be made on a case-by-case basis, but the data will be presented in data listings regardless.

Plasma concentrations that are below the limit of quantification (BLQ) will be given a value of 0 in the summarized concentration values and derived PK parameters. These values will be flagged in the data listings, and deviations from this convention may be considered on a case-by-case basis as deemed appropriate.

8.1.2 Derived Datasets and Variables

Derived datasets will be generated according to CDISC guidance documents: Analysis Data Model (ADaM) Implementation Guide, Version 1.0 (17 Dec 2009); ADaM Data Structure for Adverse Event Analysis, Version 1.0 (10 May 2012).

8.1.3 Definition of Study Days and Baseline

For all safety endpoints, Baseline will be defined as the last observation before the first dose of any study drug unless otherwise specified. Study day prior to the first dose of treatment will be calculated as: date of assessment/event – date of treatment; study day on or after the date of first dose of treatment will be calculated as: date of assessment/event – date of treatment + 1.

8.2 Analysis Sets

Safety Set	The Safety Analysis Set consists of all enrolled subjects who receive [^{18}F]MNI-1054 or TAK-418 as part of this study. Subjects in this analysis set will be used for demographic, baseline characteristics, and safety summaries.
Pharmacokinetic (PK) Set	The PK Analysis Set will consist of all subjects who receive study drug (TAK-418) and have at least 1 measurable plasma concentration for TAK-418.
Pharmacodynamic (PD) Set	The PD Analysis Set will consist of all subjects who receive study drug (TAK-418) and have at least 1 analyzable PET scan following TAK-418 administration.

If any subjects are found with incomplete data, a decision will be made on a case-by-case basis as to their inclusion in the analysis but will be presented in the subject listings.

8.3 Disposition of Subjects

The number and percentage of subjects who complete study drug and study visits, and those who prematurely discontinue study drug and study visits will be summarized by Baseline [^{18}F]MNI-1054, each TAK-418 dose level, and TAK-418 overall for subjects in the Safety Set. In addition, the number and percentage of subjects will be summarized for reasons of study drug discontinuation and study visits by Baseline [^{18}F]MNI-1054, each TAK-418 dose level, and TAK-418 overall. Subjects' study completion data, including reasons for premature termination, will be listed.

The number and percentage of subjects comprising each analysis set will be summarized by Baseline [^{18}F]MNI-1054, each TAK-418 dose level, and TAK-418 overall.

8.4 Demographic and Baseline Characteristics

Demographic characteristics will be summarized and listed. Summary statistics (N, mean, SD, median, minimum, and maximum) will be generated for continuous variables (eg, age, height, weight and BMI) for Baseline [^{18}F]MNI-1054, each TAK-418 dose level, and TAK-418 overall, and the number and percentage of subjects within each category will be presented for categorical variables (eg, gender, race, and ethnicity).

Individual demographic characteristics, date of informed consent, any available safety data will also be presented in the data listings.

8.5 Medical History and Concurrent Medical Conditions

Medical history includes any significant conditions or diseases relevant to the disease under study that stopped at or prior to informed consent. Concurrent medical conditions are those significant ongoing conditions or diseases that are present at signing of informed consent.

Medical history and concurrent medical condition verbatim reported terms will be coded using the Medical Dictionary for Regulatory Activities (MedDRA). No summary statistics for medical history and concurrent medical conditions will be provided. All medical history and concurrent medical conditions will be listed.

8.6 Medication History and Concomitant Medications

Medication history information obtained includes any medication stopped at or within 30 days prior to signing of informed consent. Medications used from signing of informed consent through the end of study will be considered as concomitant medications.

Medication history and concomitant medications will be coded using World Health Organization Drug Dictionary (WHO Drug). No summary statistics for medication history and concomitant medications will be provided. All medication history and concomitant medications data will be listed.

8.7 Study Drug Exposure and Compliance

The date and time of each dose for each subject will be reported in the data listing. No summary statistics for the extent of exposure to study drug or compliance calculations will be performed for this study.

8.8 Efficacy Analysis

Not applicable.

8.9 Pharmacokinetic/Pharmacodynamic Analysis

Serial blood samples for determination of TAK-418 will be collected at approximately 0.5, 1, and 3 hours after TAK-418 administration, immediately before and after the Day 1 PET scan, and within 30 minutes before and within 30 minutes after the Day 2 (or Day 3) PET scan.

Analyte	Matrix	Day	Scheduled Time (hours)
TAK-418	Plasma	1	At approximately 0.5, 1, and 3 hours after TAK-418 administration, immediately before and immediately after the Day 1 PET scan
TAK-418	Plasma	2 (or 3)	within 30 minutes before and within 30 minutes after the follow-up PET scan

The plasma concentration of TAK-418 will be summarized by TAK-418 dose level and PET scan period over each scheduled sampling time using descriptive statistics (N, mean, SD, %CV, median, minimum, and maximum) where appropriate.

Individual plasma concentration data versus time will be presented in a data listing.

8.9.1 Analysis of Pharmacokinetic Parameters

The PK parameters of plasma TAK-418 will be determined from the concentration-time profiles for all evaluable subjects. Actual sampling times, rather than scheduled sampling times, will be used in all computations. The following PK parameters will be calculated if feasible.

Symbol/Term	Definition
Plasma	
AUC_t	Area under the plasma concentration-time curve from time 0 to t
C_{max}	Maximum observed plasma concentration
AUC_∞	Area under the plasma concentration-time curve from time 0 to infinity
Cavg	Average plasma concentrations
Cavg(scan)	Average plasma concentration for TAK-418 during PET scan period
AUC(scan)	AUC of TAK-418 during the PET scan period
AUC_{last}	Area under the plasma concentration-time curve from time 0 to time of the last quantifiable concentration

Descriptive statistics (N, mean, SD, %CV, median, minimum, and maximum) will be used to summarize the plasma PK parameters for TAK-418 by TAK-418 dose level and PET scan period where appropriate.

All plasma PK parameters will be presented in a data listing.

Additional parameters may be calculated if necessary.

8.9.2 Pharmacodynamic Analysis

LSD1 binding parameter (Ki) data obtained from each PET scan before (Day -1) and after TAK-418 dosing (Day 1, and Day 2 or 3) will be summarized by relative time to the TAK-418 dosing, TAK-418 dose level and brain regions of interest using descriptive statistics (N, mean, SD, CV%, minimum, maximum, median) where appropriate.

Individual LSD1 Ki values will be presented in a data listing.

LSD1 occupancy data obtained after TAK-418 dosing (Day 1, and Day 2 or 3) will be summarized by relative time to the TAK-418 dosing, TAK-418 dose level and brain regions of interest using descriptive statistics (N, mean, SD, CV%, minimum, maximum, median) where appropriate.

Individual LSD1 occupancy results will be presented in a data listing.

The relationships between LSD1 occupancy and TAK-418 dose level and PK parameters will be evaluated using scatter plots and Emax models for:

- LSD1 occupancy (%) vs. TAK-418 dose, AUC, Cavg(scan) and Cmax for the Day 1 PET scans.
- LSD1 occupancy (%) vs. TAK-418 dose, AUC, Cavg(scan) and Cmax for the Day 2 or 3 (if acquired) PET scans, if appropriate to be combined.

The following E_{max} model will be used:

$$E = \frac{Exposure \times E_{max}}{Exposure + EC_{50}},$$

where:

E=Observed LSD1 occupancy (%) in a brain region of interest;

Exposure = Observed TAK-418 dose, TAK-418 AUC, or TAK-418 Cavg(scan);

E_{max}=Maximum LSD1 Occupancy in the brain region of interest;

EC_{50}=Exposure or TAK-418 dose level which related to the 50% of the Maximum LSD1 occupancy in the brain region of interest.

When modeling Day 1 and Day 2 data separately, E_{max} and EC_{50} will be estimated as fixed parameters. When modeling Day 1 and Day 2 together, random effect model will be explored. Model parameter estimates (E_{max} and EC_{50}) and their 95% confidence intervals will be provided. Results from a simple E_{max} model in which E_{max} set to be 100% , will also be presented.

Other models will also be explored if deemed appropriate.

Enzyme turnover will be quantified for each subject by modeling the recovery in PET signal between Day 2 or 3 and Day 1, from to the relationship.

$$S_2 = S_1 + (S_0 - S_1)\left(1 - e^{-(t_2 - t_1)/TR}\right)$$

where:

S_0 = the PET signal (e.g., Ki) in the region of interest at baseline

S_1 = the PET signal (e.g., Ki) in the region of interest from the Day 1 scan

S_2 = the PET signal (e.g., Ki) in the region of interest from the Day 2 or 3 scan

t_1 = the time in hours post-TAK-418-dose of the Day 1 scan

t_2 = the time in hours post-TAK-418-dose of the Day 2 or 3 scan

TR = the turnover rate (half-life) in hours.

Accordingly, the enzyme turnover rate (half-life) can be estimated for each subject as:

$$TR = -\frac{(t_2 - t_1)}{\ln\left(\frac{S_0 - S_2}{S_0 - S_1}\right)}$$

Other models will also be explored if deemed appropriate.

Individual turnover rates will be presented in a data listing and graphed in scatterplots vs. TAK-418 dose, exposure measures and Day 1 occupancy. If appropriate, enzyme turnover rates will be summarized using descriptive statistics (N, mean, SD, CV%, minimum, maximum, median).

8.10 Other Outcomes

PET data collected on the eCRF will be included in listings.

The imaging analysis will be conducted by an outside vendor and are not included in this SAP.

8.11 Safety Analysis

The safety endpoints including AEs, clinical laboratory parameters, vital sign parameters, 12-lead ECG parameters will be summarized and described in data listings. The Safety Set will be used for all summaries of safety parameters. Physical examination and C-SSRS will be described in data listings.

8.11.1 Adverse Events

A treatment-emergent AE (TEAE) is defined as an AE or a SAE that occurs or gets worse after receiving the first dose of study drug (either Baseline [^{18}F]MNI-1054 or TAK-418), and within 30 days (onset date – last date of dose + 1 $\leq$30) after the last dose of study drug. A TEAE may also be a pretreatment adverse event or a concurrent medical condition diagnosed prior to the date of first dose of study drug that increases in severity after the start of dosing. Adverse events data with onset occurring more than 30 days after last dose of study drug (AE start date – last dose date >30) will be listed, but not included in the summary tables.

Adverse event verbatim reported terms will be coded by system organ class (SOC), high-level term and preferred term (PT) using MedDRA.

The TEAE summary tables will present the number and percentages of subjects experiencing at least 1 TEAE by SOC and PT and will be tabulated for Baseline [^{18}F]MNI-1054, each TAK-418 dose level, and TAK-418 overall. TEAEs tabulated under Baseline [^{18}F]MNI-1054 are defined as TEAEs starting after first [^{18}F]MNI-1054 injection during Baseline Imagining Period and prior to TAK-418 dosing whereas TEAEs tabulated under TAK-418 dose levels are defined as TEAEs starting after TAK-418 dosing.

The following is a list of AE summary tables to be generated:

- Overview of TEAEs.
- TEAEs by SOC and PT at subject and event level.
- Subject Mappings for TEAEs.
- TEAEs by PT.

- Non-Serious TEAEs by PT.
- Relationship of TEAEs to Study Drug by SOC and PT.
- Drug-Related TEAEs by SOC and PT.
- Intensity of TEAEs by SOC and PT.
- Intensity of Drug-Related TEAEs by SOC and PT.
- Pretreatment Events by SOC and PT.

Additional AE summary tables may be added as appropriate.

A subject with 2 or more different AEs within the same level of MedDRA term and regimen will be counted only once in that level using the most extreme intensity for the intensity tables, and relationship to study drug for the causality tables.

Data listings will be provided for all AEs (including pretreatment events for enrolled subjects), and for AEs leading to study drug discontinuation and leading to study visit discontinuation, SAEs, and AEs resulting in death.

8.11.2 Clinical Laboratory Evaluations

Clinical safety laboratory tests include clinical chemistry, hematology, and urinalysis. The samples for these tests will be collected following a minimum 8-hour overnight fast (for details of hematology, chemistry and urinalysis assessments). For visits that include PET scans, labs are to be performed within approximately 1 hour after each PET scan.

Individual results for hematology and chemistry laboratory tests will be evaluated against the Takeda predefined laboratory MAV criteria (Appendix A) using the result and criteria in SI units. All subjects with at least 1 post dose laboratory result that meet the MAV criteria will be presented in a data listing. The number and percentage of subjects with at least 1 postdose markedly abnormal laboratory test result will also be summarized for Baseline [^{18}F]MNI-1054 and each TAK-418 dose level. Subjects who meet the MAV criteria will be mapped to their respective qualifying laboratory result. All post dose clinical lab MAV results, including scheduled and unscheduled measurements will be included in the MAV summaries.

All clinical laboratory data will be presented in both SI and conventional units in the listings.

8.11.3 Vital Signs

Vital signs will include oral temperature, pulse rate, respiratory rate, and systolic and diastolic blood pressure. These vital signs will be obtained 1 hour before each [^{18}F]MNI-1054 injection, within approximately 1 hour after each PET scan, within approximately 1 hour before TAK-418 administration, at approximately 1, 2, and 24 hours after TAK-418 administration.

All individual vital signs that meet Takeda's predefined criteria for MAVs (Appendix B) will be listed. The number and percentage of subjects with at least 1 postdose markedly abnormal vital sign measurement will be summarized for Baseline [^{18}F]MNI-1054 and each TAK-418 dose

level. Subjects who meet the MAV criteria will be mapped to their respective qualifying vital sign result. All post dose MAV vital signs, including both scheduled and unscheduled measurements, will be included in the MAV summaries.

Individual subject vital signs will be presented in data listings.

8.11.4 Physical Examinations

Physical examination is performed at Screening, Check-in (Day -15 to Day-2), Baseline PET Scan (Day-14 to Day -1), Day 1, Study Exit (Day 3) or Early Termination and Follow-up (Day 14 ±4).

The physical examination findings will be presented in data listings. No summary tables will be provided.

8.11.5 12-Lead ECGs

ECG measurements include heart rate, PR interval, QRS interval, QT interval, QTcF interval, and the interpretation of the ECG profile by the principal investigator. The samples for these tests are collected at the following timepoints, at 1 hour before each [^{18}F]MNI-1054 injection, within approximately 3 hours after each PET scan, within approximately 1 hour before TAK-418 administration, at approximately 1, 2, and 24 hours after TAK-418 administration. The predose ECG will obtained within approximately 1 hour before study drug dosing. This measurement will be used as the baseline assessment.

All individual ECGs that meet Takeda's predefined criteria for MAV (Appendix B) will be listed. The number and percentage of subjects with at least 1postdose markedly abnormal ECG measurement will be summarized for Baseline [^{18}F]MNI-1054 and each TAK-418 dose level. Subjects who meet the MAV criteria will be mapped to their respective qualifying ECG result. All post dose MAV ECGs, including both scheduled and unscheduled measurements, will be included in the MAV summaries.

Individual subject ECGs will be presented in data listings.

8.11.6 Other Observations Related to Safety

All results for the neurological monitoring and the C-SSRS will be listed.

8.12 Interim Analysis

Not applicable.

8.13 Changes in the Statistical Analysis Plan

Any changes/modifications of the statistical analyses outlined in this SAP, will be presented and justified in the relevant section of the CSR.

9.0 REFERENCES

1. Protocol: A Phase 1, Open-label, Positron Emission Tomography Study With [^{18}F]MNI-1054 to Determine Lysine-Specific Demethylase 1A Brain Enzyme Occupancy of TAK-418 After Single-Dose Oral Administration in Healthy Subjects. 27 September 2019.

Appendix A Criteria for Identification of Markedly Abnormal Laboratory Values

CCI

[redacted]

CCI

[redacted]

CCI

[redacted]

CCI

ELECTRONIC SIGNATURES

Signed by	Meaning of Signature	Server Date (dd-MMM-yyyy HH:mm 'UTC')
PPD	Biostatistics Approval	20-Feb-2020 17:37 UTC