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

Protocol Approve Date: 5 March 2020

Certain information within this protocol has been redacted (ie, specific content is masked irreversibly from view with a black/blue bar) to protect either personally identifiable information (PPD) or company confidential information (CCI).

This may include, but is not limited to, redaction of the following:

- Named persons or organizations associated with the study.
- Proprietary information, such as scales or coding systems, which are considered confidential information under prior agreements with license holder.
- Other information as needed to protect confidentiality of Takeda or partners, personal information, or to otherwise protect the integrity of the clinical study.

TAKEDA PHARMACEUTICALS

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

Study Identifier: TAK-418-0004

Compound: TAK-418

Date: 05 March 2020 **Amendment Number:** 2

Amendment History:

Date	Amendment Number	Region
27 September 2019	Initial version	United States
25 October 2019	1	United States
05 March 2020	2	United States

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LIST OF APPENDICES

1.0 STUDY SUMMARY

Name of Sponsor: Takeda Pharmaceuticals USA (TPUSA) 95 Hayden Avenue Lexington, MA 02421	**Compound:** TAK-418
Study Identifier: TAK-418-0004	**Phase:** 1

Protocol Title: 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

Study Design:

This is a positron emission tomography (PET) imaging study with a single oral dose of TAK-418 and up to 3 administrations of intravenous microdoses of the lysine-specific demethylase 1 (LSD1) PET radiotracer [^{18}F]MNI-1054. The primary objective is 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 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). There will be 2 confinement periods: 1 for the baseline PET scan and 1 for the treatment period PET scans. 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 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 hours after TAK-418 dosing, and at approximately 26.5 hours after TAK-418 dosing.

Study Primary Objective:

The primary objective of the study is to determine the relationship between the occupancy of LSD1 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.

Secondary Objectives:

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

Study Subject Population: Healthy male or female subjects.

Planned Number of Subjects: Up to 16 evaluable subjects	**Planned Number of Sites:** 1
Dose Levels: Up to 5	**Route of Administration:** Oral (TAK-418); intravenous ([^{18}F]MNI-1054)
Duration of Treatment: Single dose	**Planned Study Duration:** Approximately 62 days (Screening: ~30 days + Baseline Period Check-in and PET scan: ~15 days + Treatment Period: 3 days + Follow-up: ~14 days)

Main Criteria for Inclusion:

To be eligible for study participation:

- The subject must understand the study procedures and agree to participate by providing written informed consent.
- The subject must be willing and able to comply with all study procedures and restrictions.
- The subject is male or female and aged 18 to 65 years, inclusive, at the screening visit.
- The subject must have a body mass index ≥18.5 and ≤30.0 kg/m^2 at the screening visit
- The subject must be judged to be in good health by the investigator, based on clinical evaluations including laboratory safety tests, medical history, physical examination, 12-lead electrocardiogram, and vital sign measurements performed at the screening visit and before the first dose of study drug or first invasive procedure.
- The subject has adequate circulation to both hands for safe placement of arterial lines (as determined by Allen's test).
- A female subject is EITHER of nonchildbearing potential or, if of childbearing potential, is using at least 1 highly effective method of contraception with low user dependency (as defined in the protocol and informed consent) from screening, throughout the entire duration of the study, and for 31 days after study drug administration. In addition, female subjects must agree not to donate ova during this period.
- Male subjects who are non-sterilized and sexually active with a partner of childbearing potential must use adequate contraception (as defined in the protocol and informed consent) from screening, throughout the entire duration of the study, and for 91 days after study drug administration. In addition, male subjects must agree not to donate sperm during this period.

Main Criteria for Exclusion:

The subject must be excluded from participating in the study if he or she:

- Has contraindications to undergoing MRI examination including but not limited to implants such as implanted cardiac pacemakers or defibrillators, insulin pumps, cochlear implants, metallic ocular foreign body, implanted neural stimulators, central nervous system aneurysm clips and other medical implants that have not been certified for MRI, or history of claustrophobia in MRI.
- Has participated in other research protocols or clinical care in the last year in addition to the radiation exposure expected from participation in this clinical study, such that radiation exposure exceeds the effective dose of 50 mSv, which would be above the acceptable annual limit established by the United States Federal Guidelines.
- Has clinically significant abnormal findings on brain MRI that in the opinion of the investigator may interfere with the interpretation of the PET imaging.
- Has a risk of suicide according to the investigator's clinical judgment per the Columbia-Suicide Severity Rating Scale at screening or has made a suicide attempt in the 12 months before screening.
- Has luteinizing hormone, follicle-stimulating hormone, or estradiol levels that are clinically abnormal.
- Is in the opinion of the investigator, unsuitable in any other way to participate in this study.

Main Criteria for Evaluation and Analyses:

The primary endpoints of the study are:

- Quantitative estimates of binding of [^{18}F]MNI-1054 based on appropriate PET radiotracer kinetic models (eg, inhibitory constant (K_i) from irreversible 2-tissue compartmental model). The modeling approach will be informed by the data from the first-in-human radiotracer validation study TAK-418-0002 (CCI [redacted]).
- Percent enzyme occupancy calculated from quantitative estimates of binding (= 100 × [baseline – postdose]/baseline).
- Plasma PK parameters including, if feasible, but not limited to:
  - Maximum observed concentration.
  - Area under the concentration-time curve from time 0 to time t.

- Area under the concentration-time curve from time 0 to time infinity.

The secondary endpoints will be assessed through evaluation of the following parameters:

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

Statistical Considerations:

Enzyme Occupancy from PET:

Descriptive summaries for PET radiotracer binding (eg, K_i from irreversible 2-tissue compartment model) and target occupancy will be provided. Enzyme occupancy of brain LSD1 by TAK-418 in brain regions including the cerebellum will be calculated from the percent reduction of binding by comparing the baseline and postdose imaging values. The relationship between enzyme occupancy and drug exposure will be modeled using a maximum drug-induced effect model, or similar, and presented as an enzyme occupancy versus TAK-418 drug exposure curve. The relationship between enzyme occupancy and administered dose will also be assessed.

Safety:

Descriptive statistics will be provided for safety, demographics, and subject disposition data by dose level. Descriptive statistics for continuous data will include means, medians, SDs, and ranges, while categorical data may be summarized using frequency counts and percentages.

PKs:

TAK-418 concentrations will be tabulated using descriptive statistics during the imaging period for each dose of TAK-418. Appropriate PK endpoints will be used to model the relationship between plasma exposure and enzyme occupancy.

Sample Size Justification:

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.

1.1 Protocol Amendment 2 Summary of Changes

This section describes the changes in reference to the Protocol incorporating Amendment 2.

There are 2 main purposes of this amendment. One is to clarify in the Schedule of Study Procedures that there will be 2 confinement periods, one for the baseline period and one for the treatment period. Second, statements were removed that would necessitate including data for subjects who do not receive study drug in the eCRFs or in tables, listings and figures. Other minor changes in procedures are proposed. Minor grammatical and editorial changes were made for clarification purposes only.

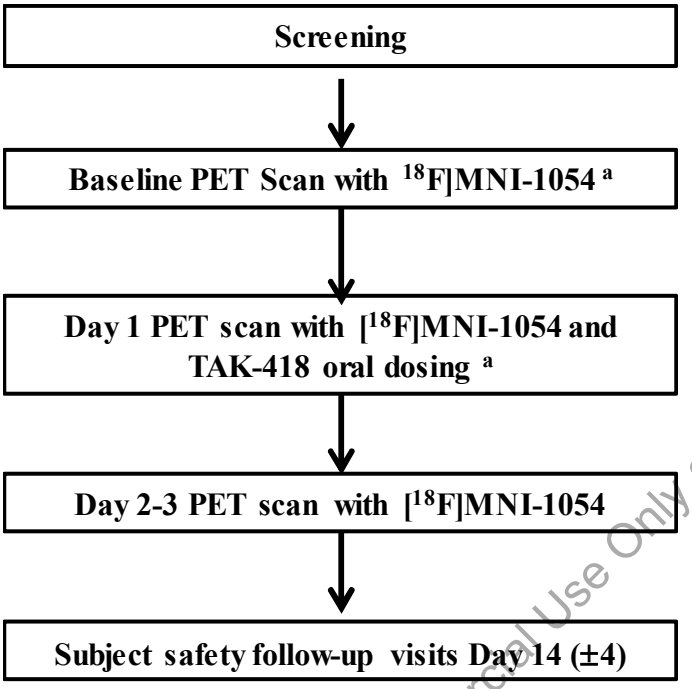
For descriptions and locations of text changes listed below, as well as rationales for these changes, see Appendix E.

Changes in Amendment 2:

1. Clarified that there will be 2 separate confinement periods.
2. Allen's test was added to Day 2-3 procedures.
3. Corrected terminology referring to the 2 versions of the Columbia-Suicide Severity Rating Scale (C-SSRS).
4. Modified text to remove any requirement to complete eCRFs or produce statistical tables, listings or figures for subjects who do not receive study drug (TAK-418 or [18F]MNI-1054).
5. Updated serious adverse event (SAE) reporting guidance.

2.0 STUDY SCHEMATIC

Figure 2.a Adaptive Design Evaluation - Evaluation of First 2 Subjects to Optimize Timing of PET Scan With [^{18}F]MNI-1054 and TAK-418 Oral Dosing Before Enrolling Additional Subjects (≈14)



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

3.0 SCHEDULE OF STUDY PROCEDURES

Study Day:	Screening	Baseline (BL) Period [a]		Treatment Period [a]			ET	Follow-up
	Days -45 to BL Check-in	Check-in Day -15 to Day -3	PET Scan Days -14 to -2	Check-in Day -1	Day 1	Day 2-3		Day 14 (±4)
Administrative Procedures								
Informed consent	X							
Inclusion/exclusion criteria	X	X	X	X	X			
Medical history/demographics	X							
Prior and concomitant medication review	X	X	X	X	X	X	X	X
Clinical Procedures/Assessments								
Full physical examination	X	X	X	X	X	X	X	X
Neurological examination	X	X	X	X	X	X	X	X
Height	X							
Weight	X	X	X	X	X			
Body mass index	X							
Semirecumbent vital signs (heart rate, systolic and diastolic blood pressure)	X	X [b]	X [b]	X [b]	X [b]	X	X	X
Vital signs (respiratory rate, oral temperature)	X	X	X	X	X	X	X	X
Standard 12-lead ECG	X	X [b]	X [b]	X [b]	X [b]	X	X	X
Allen's test (source only)	X		X		X	X		
Brain MRI	X							
C-SSRS [c]	X	X	X	X	X	X	X	X
PTE assessment [d]	X	X						
AE assessment [e]			X	X	X	X	X	X
Insertion of arterial catheter [f]			X		X	X		
Insertion of venous catheters			X		X	X		
Study drug (TAK-418) administration [g]					X (t = 0)			
Laboratory Procedures/Assessments								
Hematology [h]	X	X	X	X	X	X	X	X
Chemistry [h]	X	X	X	X	X	X	X	X
Urinalysis [h]	X	X	X	X	X	X	X	X
Hepatitis screen	X							
HIV screen	X							
PT and PTT	X							

Study Day:	Screening	Baseline (BL) Period [a]		Treatment Period [a]			ET	Follow-up
	Days -45 to BL Check-in	Check-in Day -15 to Day -3	PET Scan Days -14 to -2	Check-in Day -1	Day 1	Day 2-3		Day 14 (±4)
Serum hCG/ [i]	X	X		X			X	X
Urine dip stick pregnancy test [i]			X		X	X		
Urine drug screen	X	X		X			X	X
Alcohol screen	X	X		X				
Cotinine screen	X	X		X				
PK Evaluations								
Plasma sample for TAK-418 PK					X [j]	X [j]	X	
PD-Related Evaluations								
[^{18}F]MNI-1054 PET radiotracer injection [l]			X		X	X		
PET brain imaging [l, m]			X		X	X		
Arterial blood sampling for [^{18}F]MNI-1054 radio equivalents [f]			X		X	X		
Other								
Standardized meals		X	X	X	X	X		

AE: adverse event; BL, baseline; C-SSRS: Columbia-Suicide Severity Rating Scale; ECG: electrocardiogram; ET: early termination; FSH: follicle-stimulating hormone; hCG: human chorionic gonadotropin; LH: luteinizing hormone-releasing hormone; MRI: magnetic resonance imaging; PD: pharmacodynamic; PET: positron emission tomography; PK: pharmacokinetic; PT: prothrombin time; PTE: pretreatment event; PTT: partial prothrombin time.

[a] There will be 2 confinement periods. The baseline confinement period is from the day before the baseline PET scan through completion of all baseline assessments. The treatment confinement period is from Day -1 until completion of Day 2 or 3 assessments.

[b] Will be obtained as described in Sections 9.2.5 (Vital Signs) and 9.2.7 (12-Lead ECG).

[c] C-SSRS: The current C-SSRS Screening/Baseline will be administered at screening. The current C-SSRS Since Last Visit assessment will be administered on all other days.

[d] PTEs will be collected from signing of informed consent up until first administration of [^{18}F]MNI-1054 radiotracer.

[e] AEs will be collected from the time of first administration of [^{18}F]MNI-1054 radiotracer up until the Day 14 (±4) follow-up visit.

[f] The arterial catheter may be inserted as described in Section 9.2.10.

[g] Subjects will fast for at least 8 hours before TAK-418 dosing on Day 1 and will continue to fast for an additional 4 hours after dosing. The single oral dose of TAK-418 will be administered with approximately 240 mL of water (see Section 7.4.1 for additional dietary information).

[h] For details of hematology, chemistry and urinalysis assessments, refer to Sections 9.3.1, 9.3.2 and 9.3.3, respectively. Prior to imaging on Day 1. subjects will have chemistry, hematology, and urinalysis performed if they have not been confined since prior safety labs. For other visits that include PET scans, labs are to be performed within approximately 1 hour after each PET scan.

[i] For women of childbearing potential (see Appendix D for definition). To be performed before any administration of study drug or PET scan. Results from urine dip stick pregnancy tests may be used if results from serum hCG pregnancy tests are not available in time for study procedures or study drug administration.

[j] Will occur pre-dose, approximately 0.5, 1, and 3 hours after TAK-418 administration, immediately before and immediately after the Day 1 PET scan. Subjects will fast for at least 8 hours before the pre-dose PK sample is collected and will continue to fast for an additional 4 hours after TAK-418 dosing (Section 7.4.1). On Day 2 (or Day 3), PK samples will be collected within 30 minutes before and within 30 minutes after the follow-up Day 2 (or Day 3) PET scan (Section 9.4.1).

[k] On Day 1, subjects will fast for at least 8 hours before the pre-dose PK sample is collected and will continue to fast for an additional 4 hours after TAK-418 dosing (Section 7.4.1). On Day 2 (or Day 3), PK samples will be collected within 30 minutes before and within 30 minutes after the follow-up Day 2 (or Day 3) PET scan (Section 9.4.1). Fasting is not required on non-dosing days (Day 2-3 and end of treatment).
[l] 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.

4.0 INTRODUCTION

4.1 Background

4.1.1 TAK-418

Lysine-specific demethylase 1 (LSD1), a member of the lysine-specific demethylase (KDM) family, is involved in a wide variety of cellular processes and pathologies, including signal transduction, transcriptional regulation, viral pathogenesis, cell proliferation, cancer and metastasis, cell development, cell differentiation, and chromatin remodeling [1]. As such, LSD1 has emerged as a potential therapeutic target for development disorders involving chromatin remodeling defects, such as autism spectrum disorder (ASD) and other neurodevelopmental disorders [2].

One of the modifications that has been extensively studied in patients with ASD is methylation of lysine in position 4 of type 3 histone (H3K4) [3], and LSD1 is an important regulator of H3K4 methylation. LSD1 uses a flavin adenine dinucleotide (FAD) cofactor to oxidize carbon-nitrogen bonds with subsequent production of a demethylated lysine residue and formaldehyde by-product. The first reported specific substrate of LSD1 was histone H3K4me1/2, and subsequently additional specific substrates have been identified [4].

CCI

[redacted]

CCI

[redacted]

CCI

[redacted]

CCI

[redacted]

CCI

[redacted]

Please refer to the current TAK-418 Investigator's Brochure for additional background information.

4.1.2 [^{18}F]MNI-1054

CCI

[redacted]

CCI

The TAK-418-0002 study results indicate that [^{18}F]MNI-1054 is a suitable PET radiotracer for measuring LSD1 occupancy in human studies.

4.2 Rationale for the Proposed Study

This study will yield a mathematical relationship between the plasma levels of TAK-418 and the degree of enzyme occupancy in the brain (target engagement). This will enable doses to be chosen for subsequent studies with an understanding of the corresponding occupancy of the TAK-418

binding site in the brain. This relationship is not expected to be different in any of the potential patient populations under consideration for TAK-418; thus, a healthy subject population is deemed appropriate.

CCI [redacted] An objective of this study is thus to achieve both a predicted and measured enzyme occupancy, at the first postdose scan time point, that will exceed 90% and correspond to near-maximal inhibition within the range of doses to be tested. The second postdose PET scan will provide information on the LSD1 enzyme turnover rate in humans.

4.3 Benefit-Risk Profile

CCI [redacted]

CCI

[redacted]

CCI

[redacted]

5.0 STUDY OBJECTIVES AND ENDPOINTS

5.1 Hypothesis

At 1 or more doses tested, TAK-418 will produce ≥90% occupancy of LSD1 enzyme in at least 2 subjects per dose level, in human brain region(s) of high receptor binding (eg, cerebellum).

5.2 Study Objectives

5.2.1 Study Primary Objective

The primary objective of the study is to determine the relationship between the occupancy of LSD1 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.

5.2.2 Study Secondary Objectives

The secondary objectives 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 Endpoints

5.3.1 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). The modeling approach will be informed by the data from the ongoing FIH radiotracer validation study TAK-418-0002 ([redacted].
- 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_∞.

5.3.2 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.

6.0 STUDY DESIGN AND DESCRIPTION

6.1 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 is 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 in 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 2 confinement periods as described in the Schedule of Study Procedures (Section 3.0): 1 for the baseline PET scan and 1 for the treatment period PET scans. Plasma samples will be taken at prescribed intervals to assess the peripheral 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.

6.2 Dose Selection

The starting dose in this study will be 1.5 mg (see Section 6.3.2). 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.

6.3 Rationale for Study Design, Dose, and Endpoints

6.3.1 Rationale of Study Design

The study design is typical for phase 1 receptor- or enzyme-occupancy studies and considered appropriate for the objectives of this study. The adaptive nature of the design ensures that doses are selected over sufficient range that the exposure-occupancy relationship is determined with

sufficient accuracy to inform dose selection, and that the number of subjects exposed to the investigational compound and ionizing radiation is minimized.

6.3.3 Rationale for Endpoints

6.3.3.1 Efficacy Endpoints

This is a phase 1 biomarker study designed to quantify target engagement after single doses in healthy subjects; no efficacy endpoints are included.

6.3.3.2 Safety Endpoints

Safety assessments are included to ensure adequate monitoring of the safety and tolerability of TAK-418 and of [^{18}F]MNI-1054 in this study.

6.3.3.3 PK Endpoints

The PK analyses will provide measures of TAK-418 exposure in plasma so that their relationship to enzyme occupancy in the brain can be determined.

6.3.4 Critical Procedures Based on Study Objectives: Timing of Procedures

For this study, the administration of [^{18}F]MNI-1054 and TAK-418, collection of PET imaging data and PK sampling are the critical procedures.

- At any time point following the administration of [^{18}F]MNI-1054 and TAK-418, collection of PET imaging data and PK sampling should be performed as close to scheduled time as possible.
- All other procedures should be performed as close as possible (either before or after) the scheduled times.
- ECG and vital signs measurements should be performed before the nominal time of the administration of [^{18}F]MNI-1054 and TAK-418, collection of PET imaging data, and PK sampling if scheduled together.
- The order of priority can be changed during the study with joint agreement of the investigator and the sponsor.
- Any nonscheduled procedures required for urgent evaluation of safety concerns take precedence over all routine scheduled procedures.

6.4 Study Design/Dosing/Procedures Modifications Permitted Within Protocol Parameters

This is a phase 1 assessment of [^{18}F]MNI-1054 and TAK-418 in adult men and women, and the PK, PD, and safety profiles of the compounds are still being elucidated. This protocol is written with some flexibility to accommodate the inherent dynamic nature of phase 1 clinical studies. Subjects will receive a single oral dose of TAK-418 administered with approximately 240 mL of water on Day 1 (Sections 7.4.1 and 9.2.8). Modifications to the doses, dosing regimen, and/or clinical or laboratory procedures, as outlined below, may be required to achieve the scientific goals of the study objectives and/or to ensure appropriate safety monitoring of the study subjects.

As such, the following alterations from the currently outlined dose and/or dosing regimen may be permitted on the basis of newly available data, but the maximum daily dose/exposure may not exceed that currently outlined in Section 6.3.2:

- A subject will receive a single dose of TAK-418. If the critical procedures cannot be performed (eg, in the case of synthesis failure of [^{18}F]MNI-1054 or PET scanner malfunction), the same subject will not be used for a repeat dose and a backup subject will receive that dose. However, a subject already dosed may, at the discretion of the investigators, be scanned later the same day (first postdose scan) or within the window of Days 2 to 3 (second postdose scan) if the technical issues are resolved in time (eg, resynthesis of [^{18}F]MNI-1054).
- The number of days in the study may be increased on the basis of emerging information.
- Instructions to take study drug with or without food or drink may be modified on the basis of newly available data.
- The PK/PD sampling scheme may be modified during the study on the basis of newly available PK or PD data (eg, to obtain data closer to the time of first occurrence of C_{max}). If indicated, these collected samples may also be assayed in an exploratory manner for metabolites and/or additional PD markers.

- The timing of planned procedures for assessment of safety procedures (eg, vital signs, ECGs, safety laboratory tests) may be modified during the study on the basis of newly available safety, tolerability, PK, or PD data (eg, to obtain data closer to the time of first occurrence of C_{max}). These changes will not increase the number of study procedures for a given subject during her participation in the entire study.
- Additional laboratory safety tests may be added to blood samples previously drawn to obtain additional safety information.
- It is understood that the current study may employ some or none of the alterations previously described. Any alteration made to this protocol to meet the study objectives must be detailed by the sponsor in a letter to the study file and forwarded to the investigator for retention. The letter may be forwarded to the institutional review board (IRB)/independent ethics committee (IEC) at the discretion of the investigator.

6.5 Study Beginning and End/Completion

6.5.1 Definition of Beginning of the Study

The overall study begins when the first subject signs the study informed consent form.

6.5.2 Definition of End of the Study

The overall study ends when the last subject completes the last planned or follow-up visit/interaction associated with a planned visit, discontinues from the study, or is lost to follow-up (ie, the investigator is unable to contact the subject).

6.5.3 Definition of Study Discontinuation

Study discontinuation because of nonsafety reasons, such as the following:

- A finding (eg, PK, PD, efficacy, biologic targets) from another nonclinical or clinical study using the study treatment(s) results in the study being stopped for a nonsafety-related reason.
- Data from comparator(s), drug(s) of the same class, or methodology(ies) used in this study become available and results in the study being stopped for a nonsafety-related reason.
- The study is stopped because of nonscientific and nonsafety reasons, such as slow enrollment.

Study discontinuation because of safety reasons:

- Early study termination because of unanticipated concerns of safety to the study subjects arising from clinical or nonclinical studies with the study treatment(s), comparator(s), drug(s) of the same class, or methodology(ies) used in this study.

6.5.4 Criteria for Premature Termination or Suspension of the Study

6.5.4.1 Criteria for Premature Termination or Suspension of Study Sites

A study site may be terminated prematurely or suspended if the site (including the investigator) is found in significant violation of Good Clinical Practice (GCP), protocol, or contractual agreement, or is unable to ensure adequate performance of the study, or as otherwise permitted by the contractual agreement.

6.5.4.2 Procedures for Premature Termination or Suspension of the Study or the Participation of Study Sites

In the event that the sponsor, an IRB/IEC, or regulatory authority elects to terminate or suspend the study or the participation of an investigational site, a study-specific procedure for early termination or suspension will be provided by the sponsor; the procedure will be followed by applicable investigational sites during the course of termination or study suspension.

7.0 SELECTION AND DISCONTINUATION/WITHDRAWAL OF SUBJECTS

7.1 Inclusion Criteria

Subject eligibility is determined according to the following criteria before entry into the study:

1. The subject must understand the study procedures and agree to participate by providing written informed consent.
2. The subject must be willing and able to comply with all study procedures and restrictions.
3. The subject is male or female and aged 18 to 65 years, inclusive, at the screening visit.
4. The subject must have a body mass index (BMI) ≥18.5 and ≤30.0 kg/m^2 at the screening visit.
5. The subject must be a current nonsmoker at screening as demonstrated by negative cotinine test.
6. The subject must be judged to be in good health by the investigator, on the basis of clinical evaluations including laboratory safety tests, medical history, physical examination, ECG, and vital sign measurements performed at the screening visit and before the first dose of study drug or first invasive procedure.

   The subject has adequate circulation to both hands for safe placement of arterial lines (as determined by Allen's test [source documentation only]).
7. A female subject is EITHER of nonchildbearing potential* OR, if of childbearing potential*, is using at least 1 of the following highly effective methods of contraception with low user dependency* from screening, throughout the entire duration of the study, and for 31 days after study drug administration and follow necessary precautions listed in Appendix D.

   a. Intrauterine device (IUD).

   b. Intrauterine hormone-releasing system (IUS).

   In addition, female subjects must also agree not to donate ova during this period.

   *Definitions and acceptable methods of contraception are provided in Appendix D.
8. Male subjects who are non-sterilized* and sexually active with a partner of childbearing potential* must use adequate contraception from screening, throughout the duration of the study, and for 91 days after study drug administration and follow necessary precautions listed in Appendix D.

   In addition, male subjects must agree not to donate sperm during this period.

   *Definitions and acceptable methods of contraception are provided in Appendix D.

7.2 Exclusion Criteria

Any subject who meets any of the following criteria will not qualify for entry into the study:

1. The subject has participated in another investigational study within 4 weeks (or based on local regulations) before the screening visit. The 4-week window will be derived from the date of the last study procedure and/or AE related to the study procedure in the previous study to the screening visit of the current study.
2. The subject is an employee of the sponsor or study site or immediate family member (eg, spouse, parent, child, sibling) of the sponsor or study site.
3. The subject has a history of cancer (malignancy).
4. The subject has a history of significant multiple and/or severe allergies (eg, food, drug, latex allergy) or has had an anaphylactic reaction or significant intolerance to prescription or nonprescription drugs or food.
5. Has a known hypersensitivity to any component of the formulation of TAK-418 or related compounds, including [^{18}F]MNI-1054.
6. The subject has a positive alcohol or drug screen.
7. The subject has a positive pregnancy test.
8. The subject is a lactating/nursing female.
9. The subject has a positive test result for hepatitis B surface antigen (HBsAg), hepatitis C virus (HCV) antibody, or human immunodeficiency antibody/antigen, at the screening visit. Note: Subjects with positive hepatitis B virus (HBV) or HCV serology may be enrolled if quantitative polymerase chain reaction for HBV or HCV RNA is negative.
10. The subject had major surgery or donated or lost 1 unit of blood (approximately 500 mL) within 4 weeks before the screening visit.
11. The subject is unable to refrain from or anticipates using any concomitant medications other than those allowed per Section 9.1.3 beginning approximately 7 days before administration of the first dose of study drug, throughout the study, until the final follow-up visit.
12. The subject has a history of alcohol consumption exceeding 2 standard drinks per day on average (1 glass is approximately equivalent to the following: beer [354 mL/12 oz], wine [118 mL/4 oz], or distilled spirits [29.5 mL/1 oz] per day).
13. The subject consumes excessive amounts, defined as greater than 6 servings (1 serving is approximately equivalent to 120 mg of caffeine) of coffee, tea, cola, energy drinks, or other caffeinated beverages per day.
14. The subject has a substance abuse disorder.
15. The subject cannot tolerate venipuncture or has poor venous access that would cause difficulty in collecting blood samples.

16. The subject has contraindications to undergoing MRI examination including but not limited to implants, such as implanted cardiac pacemakers or defibrillators, insulin pumps, cochlear implants, metallic ocular foreign body, implanted neural stimulators, central nervous system aneurysm clips, and other medical implants that have not been certified for MRI, or history of claustrophobia in MRI.
17. The subject has prior participation in other research protocols or clinical care in the last year in addition to the radiation exposure expected from participation in this clinical study, such that radiation exposure exceeds the ED of 50 mSv, which would be above the acceptable annual limit established by the United States Federal Guidelines.
18. The subject has clinically significant abnormal findings on brain MRI scan that in the opinion of the investigator may interfere with the interpretation of the PET imaging.
19. The subject has experienced an acute illness within 10 days before the screening visit.
20. The subject has a risk of suicide according to the investigator's clinical judgment per the C-SSRS at screening or has made a suicide attempt in the 12 months before screening.
21. The subject has luteinizing hormone, FSH, or estradiol levels that are clinically abnormal.
22. The subject has an ECG at the screening visit, baseline or pre-dose on Day 1 that reveals a QT interval with Fridericia correction method (QTcF) >450 milliseconds for males and 470 milliseconds for females.
23. The subject has a clinically significant resting heart rate outside of the range of 50 to 100 beats per minute, confirmed on repeat testing within a maximum of 30 minutes, at the screening visit, baseline or pre-dose on Day 1.
24. The subject has existing skin rashes that can be diagnosed as dermatitis.
25. Subject is in the opinion of the investigator, unsuitable in any other way to participate in this study.

7.3 Excluded/Allowed Concomitant Medications, Supplements, Dietary Products

7.3.1 Concomitant Medications

Throughout the study, contraception (IUD or IUS; Appendix D) and the occasional use of acetaminophen (approximately <1 g/day) are the only concomitant medications allowed.

The use of concomitant medications (see Section 9.1.3) from the day of the baseline PET scan (occurring between Day -14 to Day -1 per the Schedule of Study Procedures in Section 3.0) until the final follow-up visit (Day 14 ±4) is not permitted. Subjects must be instructed not to take any medications without first consulting with the investigator. Any concomitant medication use must first be discussed with the sponsor, unless the investigator or designee considers immediate administration is necessitated.

7.3.2 Fruit Juice

Subjects will refrain from consuming grapefruit juice, grapefruits, and products containing grapefruit beginning approximately 2 weeks before administration of the first dose of [^{18}F]MNI-1054 and TAK-418, throughout the study and until 1 week after administration of TAK-418.

Subjects also will refrain from consuming all juices 24 hours before and after administration of each dose of [^{18}F]MNI-1054 and TAK-418 on PK sampling days. Consumption of all fruits other than grapefruit is allowed on all other days of the study.

7.3.3 Alcohol

Subjects will refrain from consuming alcohol 7 days before the screening visit and from 7 days before TAK-418 dosing on Day 1 until the last PK blood sample has been collected. During study participation, when permitted, alcohol consumption is limited to no more than approximately 2 alcoholic beverages or equivalent (1 alcoholic beverage is approximately equivalent to: beer [354 mL/12 oz], wine [118 mL/4 oz], or distilled spirits [29.5 mL/1 oz]) per day.

7.3.4 Caffeine

Subjects will refrain from consuming caffeinated beverages 72 hours before each PET scan and for 7 days after administration of TAK-418, and from 24 hours before the first and until the last PK blood sample has been collected. During study participation when permitted, caffeinated beverages or xanthine-containing products will be limited to amounts of no more than 6 units per day (1 unit = 120 mg of caffeine).

7.3.5 Smoking

Smoking is not permitted during the study.

7.4 Diet, Fluids, and Activity

7.4.1 Diet and Fluids

Subjects will fast for at least 8 hours before the pre-dose PK sample and will continue to fast for an additional 4 hours after dosing. Subjects will receive a single oral dose of TAK-418 administered with approximately 240 mL of water on Day 1. Subjects may consume water ad libitum except for 1 hour before and 1 hour after study drug administration.

During the confinement periods, subjects will be given a standard menu that includes 3 meals and an evening snack. The meals served on the day of TAK-418 dosing may differ from the standard menu because of fasting requirements but should be similar in nutritional content for each subject in the study. Subjects will fast from all food and drink except water between meals and snacks. The caloric content and composition of meals will be similar for all subjects.

7.4.2 Activity

Subjects will avoid unaccustomed strenuous physical activity (eg, weight lifting, running, bicycling) from the screening visit until administration of the initial dose of study drug, throughout the study (including washout intervals between treatment periods), and for 7 days after administration of TAK-418,

7.5 Criteria for Discontinuation or Withdrawal of a Subject

The primary reason for discontinuation or withdrawal of the subject from the study or study drug should be recorded in the electronic case report form (eCRF) using the following categories.

1. Pretreatment event (PTE) or AE. The subject has experienced a PTE or AE that requires early termination because continued participation imposes an unacceptable risk to the subject's health or the subject is unwilling to continue because of the PTE or AE.
   - Liver function test (LFT) Abnormalities

     Administration of TAK-418 and [^{18}F]MNI-1054 should be discontinued immediately with appropriate clinical follow-up (including repeat laboratory tests, until a subject's laboratory profile has returned to normal/baseline status, see Section 10.2.8.4), if the following circumstances occur at any time during study drug treatment:
     - Alanine aminotransferase (ALT) or aspartate aminotransferase (AST) >8 times the upper limit of normal (ULN), or
     - ALT or AST >5 times the ULN and persists for more than 2 weeks, or
     - ALT or AST >3 times the ULN in conjunction with elevated total bilirubin >2 times the ULN or international normalized ratio >1.5, or
     - ALT or AST >3 times the ULN with appearance of fatigue, nausea, vomiting, right upper quadrant pain or tenderness, fever, rash, and/or eosinophilia (>5%).
2. Significant protocol deviation. The discovery after the first dose of [^{18}F]MNI-1054 and/or TAK-418 that the subject failed to meet protocol entry criteria or did not adhere to protocol requirements, and continued participation poses an unacceptable risk to the subject's health.
3. Lost to follow-up. The subject did not return to the clinic and attempts to contact the subject were unsuccessful. Attempts to contact the subject must be documented in the subject's source documents.
4. Voluntary withdrawal. The subject (or subject's legally acceptable representative) wishes to withdraw from the study. The reason for withdrawal, if provided, should be recorded in the eCRF.

   Note: All attempts should be made to determine the underlying reason for the withdrawal and, where possible, the primary underlying reason should be recorded (ie, withdrawal due to an AE should not be recorded in the "voluntary withdrawal" category).
5. Study termination. The sponsor or IRB terminates the study.

6. Pregnancy. The subject is found to be pregnant.

   Note: If the subject is found to be pregnant, the subject must be withdrawn immediately.

CCI

8. Other.

   Note: The specific reasons should be recorded in the "specify" field of the eCRF.

7.6 Procedures for Discontinuation or Withdrawal of a Subject

The investigator may discontinue a subject's study participation at any time during the study when the subject meets the study termination criteria described in Section 7.5. In addition, a subject may discontinue his or her participation without giving a reason at any time during the study. Should a subject's participation be discontinued, the primary criterion for termination must be recorded by the investigator. In addition, efforts should be made to perform all procedures scheduled for the early termination visit.

7.7 Subject Replacement

If a subject discontinues from the study, a replacement subject may be enrolled, if deemed appropriate by the investigator and sponsor. The study site should contact the sponsor for the replacement of subject's treatment assignment and allocation number.

8.0 CLINICAL STUDY MATERIAL MANAGEMENT

8.1 Clinical Study Drug

Details regarding the dosage form description and strengths, or composition for the extemporaneous preparation, of the active drug can be found in the pharmacy manual or in the referenced compounding manual when applicable. Study drug will be packaged to support enrollment and replacement of subjects as required.

8.1.1 Clinical Study Drug Labeling

A clinical label will be affixed to study drug containers in accordance with local regulatory requirements.

8.1.2 Clinical Study Drug Inventory and Storage

Study drug must be stored in a secure, limited-access location under the storage conditions specified on the label and must remain in the original container until dispensed. The temperature excursion information can be found in the pharmacy manual or in the referenced compounding manual when applicable. Receipt and dispensing of study drug must be recorded by authorized personnel at the study site.

8.1.3 Clinical Study Drug Blinding

This is an open-label study.

8.1.4 Accountability and Destruction of Sponsor-Supplied Drugs

The investigator is responsible for keeping accurate records of the study drug (TAK-418) and [^{18}F]MNI-1054 precursor (CCI [redacted]) received from the sponsor or designee, the amount dispensed to and returned by the subjects, and the amount remaining at the end of the study. For all study sites, the local country sponsor personnel or designee will provide appropriate documentation that must be completed for study drug accountability, return, and destruction.

8.2 Ancillary Supplies

All ancillary supplies will be provided by either the study site or the sponsor or designee, depending upon availability. The list of ancillary supplies and source information can be found in the pharmacy manual or in the referenced compounding manual when applicable. If provided by the sponsor, unused ancillary supplies will be accounted for and disposed of as directed by the sponsor or designee.

9.0 STUDY PROCEDURES

The following sections describe the study procedures to be performed and data to be collected as indicated in the Schedule of Study Procedures (Section 3.0). For each procedure, subjects are to be assessed by the same investigator or site personnel whenever possible. Please note that it may become necessary to perform the following procedures at unscheduled time periods, per the discretion of the investigator.

9.1 Administrative Procedures

9.1.1 Informed Consent

Informed consent must be obtained before the subject enters into the study and before any protocol-directed procedures are performed. The requirements of informed consent are described in Appendix B.

9.1.1.1 Assignment of Screening Numbers

All consented subjects will be given a unique screening number that will be used to identify the subject for all procedures that occur. Each subject will be assigned only 1 screening number. Screening numbers must not be reused for different subjects. Any subject who is screened multiple times will be assigned a new screening number for each screening event.

All eligible subjects will receive a unique number. The unique number identifies the subject for all procedures occurring after meeting admission criteria. Once a unique number is assigned to a subject, it can never be reassigned to another subject. A single subject cannot be assigned more than 1 unique number.

9.1.1.2 Study Drug Assignment

On Day 1, subjects will be assigned to receive a number in ascending numerical order at the clinical site. Each subject will be dispensed and TAK-418, labeled with his/her unique number, throughout the study.

Subjects will be assigned to receive a 4-digit number. For dose level X, the sequence number will be X001 to X006. For example, the sequence numbers will be 1001 to 1006 if there are 6 subjects for dose level 1, and 2001 to 2006 if there are 6 subjects for dose level 2, etc. This 4-digit number will be used by the clinical site to facilitate the prelabeling of PK samples, as applicable, and will be the only subject identifier used on all PK sample collections. It should also be contained on the PK transport vials shipped to the bioanalytical laboratory and will be used by the laboratory to report the subject data results. This 4-digit number should only be used for the purposes described in this section. It does not replace the 3-digit subject number that is assigned at the time the informed consent is obtained and that is used for all other procedures to identify the subjects throughout the study.

9.1.2 Inclusion and Exclusion Criteria

Each subject will be assessed, according to the eligibility criteria provided in Section 7.0.

9.1.3 Medical History, Demographics, and Prior and Concomitant Medications

At screening, qualified site personnel will collect the subject's significant medical history (past and concurrent medical conditions), per the clinical site's standard of care and appropriate clinical judgment, and subject demographics. Throughout the study, contraception (IUD or IUS; Appendix D) and the occasional use of acetaminophen (approximately <1 g/day; Section 7.3.1) are the only concomitant medications allowed.

9.2 Clinical Procedures and Assessments

9.2.1 Full Physical Examination

A complete physical examination will include the examination of general appearance, skin, neck (including thyroid), eyes, ears, nose, throat, lungs, heart, abdomen, back, extremities, and musculoskeletal system. Rectal, breast, and genital examinations are not required as part of this examination.

9.2.2 Neurologic Examination

A neurologic examination will be performed and collected in the eCRF. This will include testing mental status, gait, cerebellar function, cranial nerves, motor function (including strength and reflexes), and sensation.

9.2.3 Height and Weight

Body weight and height will be obtained with the subject's shoes off and any jacket or coat removed. Height will only be obtained only at the screening visit.

9.2.4 BMI

BMI equals a subject's weight in kilograms divided by height in meters squared (BMI = kg/m^2). BMI will be rounded to the nearest whole number according to the standard convention of 0.1 to 0.4, round down, and 0.5 to 0.9, round up.

9.2.5 Vital Signs

Body temperature will be measured with an oral (temperature taken at floor of the mouth) thermometer.

Subjects should rest in a semirecumbent position for at least 5 minutes before vital signs are measured. Vital signs will include pulse rate, respiratory rate, and systolic and diastolic blood pressure. The same method (eg, same size cuff, manual or automated) must be used for all measurements for each individual subject and should be the same for all subjects.

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.

9.2.6 Assessment of Suicidal Ideation and Behavior

The current C-SSRS Screening/Baseline will be administered at screening. The current C-SSRS Since Last Visit assessment will be administered at all other timepoints. The C-SSRS was developed by researchers at Columbia University as a tool to help systematically assess suicidal ideation and behavior in subjects during participation in a clinical trial of centrally acting drugs.

The C-SSRS is composed of 3 questions addressing suicidal behavior and 5 questions addressing suicidal ideation, with subquestions assessing the severity. The tool is administered via interview with the subject.

9.2.7 12-Lead ECG

Special care must be taken for proper lead placement by qualified personnel. Skin should be clean and dry before lead placement. Subjects may need to be shaved to ensure proper lead placement. Female subjects may need to remove their bra.

Subjects should be resting in a semirecumbent position for at least 5 minutes before each ECG measurement.

QTcF intervals will be calculated in this study.

A standard 12-lead ECG will be obtained 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 pre-dose ECG will be obtained within approximately 1 hour before study drug dosing. This measurement will be used as the baseline assessment. The principal investigator should arrange to have a study cardiologist available as needed to review ECG abnormalities.

If a subject demonstrates an increase in QTcF interval ≥40 milliseconds compared with a pre-dose baseline measurement, the ECG will be repeated within 5 minutes. The average value of the QTcF interval from the 2 ECGs will represent the value at that time point. If the average QTcF interval increase from baseline for any postdose time point is ≥40 milliseconds, the subject will continue to be monitored by repeat 12-lead ECGs every 60 minutes for at least 4 hours or until the QTcF interval is within 40 milliseconds of the baseline value. If prolongation of the QTcF interval ≥40 milliseconds persists, a consultation with a study cardiologist may be appropriate and the sponsor should be notified.

If the QTcF interval is ≥500 milliseconds, the sponsor should be notified and the ECGs should be reviewed by a cardiologist. The subject should be monitored by telemetry (until the QTcF interval is <500 milliseconds) or should be considered for transfer to a location where closer monitoring is available.

If the subject has unstable hemodynamics, or has any clinically significant dysrhythmias noted by telemetry, the subject should be immediately transferred to an acute care setting for definitive therapy.

If repeat ECGs are required, the clinical site will decide whether to leave the electrodes in place or mark the position of the electrodes for subsequent ECGs. To mark the position of the electrodes, 12-lead electrode sites will be marked on the skin of each subject with an ECG skin marker pen to ensure reproducible electrode placement.

The following ECG parameters will be recorded: heart rate, PR interval, QRS interval, QT interval, QTcF interval, and the interpretation of the ECG profile by the principal investigator.

9.2.8 Study Drug Administration

On Day 1, a single oral dose of study drug (TAK-418) will be administered with approximately 240 mL of water as described in Sections 6.4 and 7.4.1. As described in Section 9.2.10, [^{18}F]MNI-1054 will be administered on Day -1, Day 1, and either Day 2 or 3.

9.2.9 MRI

As part of the screening visit, eligible subjects will undergo a 3-dimensional high-resolution T1 MRI scan without gadolinium contrast on a 1.5T or 3T MRI scanner.

9.2.10 [^{18}F]MNI-1054 PET Imaging

Venous catheters will be placed in the forearm. For the baseline PET scan (once between Days -14 and -1), there will be 1 venous catheter for [^{18}F]MNI-1054 administration. For the postdose PET scans (on Days 1 and 2-3), 2 venous catheters will be placed, 1 for [^{18}F]MNI-1054 administration and a second for blood PK collections.

An arterial catheter may be inserted, and arterial sampling done, for all 3 PET scans. Optionally (eg, if quantification methods are developed that do not require arterial sampling), some or all the PET scans may be performed without arterial sampling.

Each subject will receive an intravenous injection not exceeding 10 mCi of [^{18}F]MNI-1054 over 3 minutes using an infusion pump. Serial PET imaging will commence immediately following injection. Serial dynamic imaging (6 × 30-second, 4 × 1-minute, 4 × 2-minute PET frames, with the remainder comprising 5-minute PET frames) will be obtained over a total duration of up to 180 minutes.

If performed, arterial sampling (up to 5 mL blood per sample during the PET scan) will be obtained throughout the course of the imaging for radioactive metabolite analysis to determine the percent parent fraction of [^{18}F]MNI-1054 in plasma. In addition, 3 samples (approximately 5 mL per sample) will be drawn before [^{18}F]MNI-1054 injection for estimation of the free fraction (not bound to protein) of parent [^{18}F]MNI-1054 in plasma. Blood samples will be analyzed by high-performance liquid chromatography at the study site. The arterial catheter may be inserted, and arterial sampling done, for all 3 PET scans. Optionally (eg, if quantification methods are developed that do not require arterial sampling), some or all the PET scans may be performed without arterial sampling.

If the critical procedures cannot be performed (eg, in the case of synthesis failure of [^{18}F]MNI-1054 or PET scanner malfunction), the same subject will not be used for a repeat dose

and backup subject will receive that dose. However, a subject already dosed may, at the discretion of the investigators, be scanned later the same day (first postdose scan) or within the Days 2 to 3 window (second postdose scan) if the technical issues are resolved in time (eg, resynthesis of [^{18}F]MNI-1054).

9.2.11 AE Monitoring

PTE monitoring begins after signing of the informed consent form until [^{18}F]MNI-1054 dose and AE monitoring begins after that. Changes in subject health status from the baseline assessment until study drug administration should be captured in the subject's medical history. A complete description of AE collections and procedures is provided in Section 10.0.

9.3 Laboratory Procedures and Assessments

Laboratory samples will be collected in accordance with acceptable laboratory procedures.

Fasting is not required for safety labs (Sections 9.3.1, 9.3.2, and 9.3.3), but fasting status must be documented. Fasting is only required for the collection of Day 1 PK samples (Section 9.4).

It is anticipated that the total blood volume drawn, including PK samples (Section 9.4), will be approximately 318 mL for each subject.

9.3.1 Hematology

The hematology assessment will include the following tests:

Erythrocytes (red blood cells)	Hemoglobin
Hematocrit	Platelets
Leukocytes (white blood cells) with absolute differential	

9.3.2 Chemistry

The chemistry assessment will include the following tests:

Albumin	Alkaline phosphatase
ALT	AST
Blood urea nitrogen	Calcium
Carbon dioxide	Chloride
Creatinine	Glucose
γ-glutamyl transferase	Sodium
Potassium	Bilirubin (total); if above the ULN total bilirubin will be fractionated
Protein (total)	PT and PTT [a]
Serum FSH/LH/estradiol [b]	

ALT: alanine aminotransferase; AST: aspartate aminotransferase; FSH: follicle-stimulating hormone; LH: luteinizing hormone; PT: prothrombin time; PTT: partial thromboplastin time; ULN: upper limit of normal.

[a] Test will be performed only at the screening visit.
[b] On Day 1, this test will be performed within 24 hours of TAK-418 dose.

If subjects experience ALT or AST >3 times the ULN, follow-up laboratory tests (at a minimum, serum alkaline phosphatase, ALT, AST, total bilirubin, γ-glutamyl transferase, and international normalized ratio) should be performed 48 to 72 hours after the abnormality was noted.

If ALT or AST remains elevated >3 times the ULN on these 2 consecutive occasions, the investigator must contact the medical monitor for consideration of additional testing, close monitoring, possible discontinuation of study drug, and discussion of the relevant subject details and possible alternative etiologies. The abnormality should be recorded as an AE.

Please refer to Section 7.5 for subject discontinuation criteria regarding abnormal LFT results and Section 10.2.8.4 for guidance on reporting abnormal LFT results.

9.3.3 Urinalysis

The urinalysis assessment will include the following tests:

Protein	Glucose
Blood	Nitrite

Urine microscopy will be performed if urinalysis is abnormal. Microscopy consists of red blood cells/high-power field, white blood cells/high-power field, and casts.

9.3.4 Diagnostic Screening

9.3.4.1 Serum

The serum diagnostic screening assessment will include the following tests:

HIV	Hepatitis screen (HBsAg, HCV antibody)

HBsAg: hepatitis B surface antigen; HCV: hepatitis C virus.

9.3.4.2 Alcohol Screen

A urine alcohol test will be performed as indicated in the Schedule of Study Procedures (Section 3.0) and at the discretion of the investigator.

9.3.4.3 Urine

The urine drug screening assessment will include the following tests:

Amphetamines	3,4-methylenedioxy-methamphetamine
Barbiturates	Methadone/metabolite
Benzodiazepines	Opiates
Buprenorphine/metabolite	Oxycodone/oxymorphone
Tetrahydro cannabinoids	Phencyclidine
Cocaine/metabolites	Cotinine

9.3.4.4 Serum Human Chorionic Gonadotropin/Urine Dipstick

A urine dipstick or serum human chorionic gonadotropin (hCG) pregnancy test will be administered to women of childbearing potential (see Appendix D for definition) before any administration of study drug or PET scan, as indicated in the Schedule of Study Procedures (Section 3.0).

9.4 PK Samples

9.4.1 PK Evaluations

Samples for PK analysis will be collected at the time points stipulated in the schedule of procedures (Section 3.0). PK samples will be collected at pre-dose, 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.

On Day 1 only, subjects must be fasting overnight for at least 8 hours before collection of the pre-dose PK sample and will continue to fast for an additional 4 hours after TAK-418 dosing, for a total of approximately 12 hours. PK samples will be collected pre-dose, approximately 0.5, 1, and 3 hours after TAK-418 administration, and immediately before and after the Day 1 PET scan.

On Day 2 (or Day 3), PK samples will be collected within 30 minutes before and within 30 minutes after the PET scan. Fasting is not required on non-dosing days (Day 23 and at end of treatment).

Please refer to the laboratory manual for information on the collection, processing, and shipment of samples to the central laboratory.

The decision as to which collected samples will be assayed for evaluation of PK will be determined by the sponsor. If indicated, these samples may also be assayed and/or pooled to measure metabolites in an exploratory manner.

Primary specimen collection parameters are provided in Table 9.a.

Table 9.a Primary Specimen Collections

Specimen Name	Primary Specimen	Description of Intended Use	Sample Collection
Plasma sample for TAK-418 PK	Plasma	Pharmacokinetic measurements	Mandatory

9.4.2 PK Measurements

The PK parameters of 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 involving sampling times.

The following PK parameters will be calculated from plasma concentrations of TAK-418 when feasible, unless otherwise specified:

- C_{max}.
- AUC_t.
- AUC_∞.

Additional PK parameters may be calculated as appropriate. A detailed PK analysis plan will be prepared before PK parameter computation.

9.4.2.1 Plasma for PK Measurements

Blood samples for PK analysis of TAK-418 will be collected into chilled blood collection tubes (vacutainer) containing the anticoagulant K_2EDTA.

The actual time of sample collection will be recorded on the source document and eCRF. Sampling time points may be adjusted based on the preliminary emerging concentration data collected from prior subject(s), but the total number of samples collected per subject should not exceed the planned number.

9.5 C-SSRS

The C-SSRS will be completed at every visit, including screening, as indicated in the Schedule of Study Procedures (Section 3.0).

9.6 Confinement

There will be 2 confinement periods. Subjects will be confined overnight before the baseline PET scan and again before the Day 1 PET scan and will leave after completion of study-related procedures for Day 2-3. At the discretion of the investigator, subjects may be requested to remain at the clinical site longer.

10.0 ADVERSE EVENTS

10.1 Definitions and Elements of AEs

An AE is defined as any untoward medical occurrence in a clinical investigation subject who has signed informed consent to participate in a study; it does not necessarily have to have a causal relationship with the treatment.

An AE can therefore be any unfavorable and unintended sign (eg, a clinically significant abnormal laboratory finding), symptom, or disease temporally associated with the use of a drug, whether or not it is considered related to the drug.

An untoward finding generally may:

- Indicate a new diagnosis or unexpected worsening of a pre-existing condition. (Intermittent events for pre-existing conditions or underlying disease should not be considered AEs.)
- Necessitate therapeutic intervention.
- Require an invasive diagnostic procedure.
- Require discontinuation or a change in dose of study medication or a concomitant medication.
- Be considered unfavorable by the investigator for any reason.

Diagnoses versus signs and symptoms:

- Each event should be recorded to represent a single diagnosis. Accompanying signs (including abnormal laboratory values or ECG findings) or symptoms should NOT be recorded as additional AEs. If a diagnosis is unknown, sign(s) or symptom(s) should be recorded appropriately as an AE(s).

Laboratory values and ECG findings:

- Changes in laboratory values or ECG parameters may be considered AEs if they are judged to be clinically significant (ie, if some action or intervention is required or if the investigator judges the change to be beyond the range of normal physiologic fluctuation). A laboratory re-test and/or continued monitoring of an abnormal value are not considered an intervention. In addition, repeated or additional noninvasive testing for verification, evaluation or monitoring of an abnormality is not considered an intervention.
- If abnormal laboratory values or ECG findings are the result of pathology for which there is an overall diagnosis (eg, increased creatinine in renal failure), the diagnosis only should be reported appropriately as an AE.

Pre-existing conditions:

- A pre-existing condition (present at the time of signing of informed consent) is considered a concurrent medical history condition and should NOT be recorded as an AE. A baseline evaluation (eg, laboratory test, ECG, x-ray) should NOT be recorded as an AE unless related to a study procedure. However, if the subject experiences a worsening or complication of such a

concurrent medical history condition, the worsening or complication should be recorded appropriately as an AE (worsening or complication occurs after informed consent is signed). Investigators should ensure that the event term recorded captures the change in the condition (eg, "worsening of…").

- If a subject has a pre-existing episodic condition (eg, asthma, epilepsy), any occurrence of an episode should only be captured as an AE if the episodes become more frequent, serious, or severe in nature, that is, investigators should ensure that the AE term recorded captures the change from Baseline in the condition (eg "worsening of…").
- If a subject has a degenerative concurrent condition (eg, cataracts, rheumatoid arthritis), worsening of the condition should only be captured as an AE if occurring to a greater extent to that which would be expected. Again, investigators should ensure that the AE term recorded captures the change in the condition (eg, "worsening of…").

Worsening of AEs:

- If the subject experiences a worsening or complication of an AE after the first administration of study medication or after any change in study medication, the worsening or complication should be recorded as a new AE. Investigators should ensure that the AE term recorded captures the change in the condition (eg, "worsening of…").

Changes in severity of AEs:

- If the subject experiences a change in the severity of an AE that is not associated with a change in study medication, the event should be captured once with the maximum severity recorded.

Preplanned surgeries or procedures:

- Preplanned procedures (surgeries or therapies) that were scheduled before signing of informed consent are not considered AEs. However, if a preplanned procedure is performed early (eg, as an emergency) due to a worsening of the pre-existing condition, the worsening of the condition should be captured appropriately as an AE. Complications resulting from any planned surgery should be reported as AEs.

Elective surgeries or procedures:

- Elective procedures performed where there is no change in the subject's medical condition should not be recorded as AEs but should be documented in the subject's source documents. Complications resulting from an elective surgery should be reported as AEs.

Overdose:

- An overdose is defined as a known deliberate or accidental administration of investigational drug, to or by a study subject, at a dose above that which is assigned to that individual subject according to the study protocol. It is up to the investigator or the reporting physician to decide whether a dose is to be considered an overdose, in consultation with the sponsor.

- All cases of overdose (with or without associated AEs) will be documented in the eCRF to capture this important safety information consistently in the database. AEs associated with an overdose will be documented on AE CRF(s) according to Section 10.0.
- SAEs of overdose should be reported according to the procedure outlined in Section 10.2.8.
- In the event of drug overdose, the subject should be treated symptomatically.

10.1.1 SAEs

An SAE is defined as any untoward medical occurrence that at any dose:

1. Results in DEATH.
2. Is LIFE THREATENING.
   - The term "life threatening" refers to an event in which the subject was at risk of death at the time of the event; it does not refer to an event that hypothetically might have caused death if it were more severe.
3. Requires inpatient HOSPITALIZATION or prolongation of existing hospitalization.
4. Results in persistent or significant DISABILITY/INCAPACITY.
5. Is a CONGENITAL ANOMALY/BIRTH DEFECT.
6. Is an IMPORTANT MEDICAL EVENT that satisfies any of the following:
   - May require intervention to prevent items 1 through 5 above.
   - May expose the subject to danger, even though the event is not immediately life threatening or fatal or does not result in hospitalization.
   - Includes any event or synonym described in the Takeda Medically Significant AE List (Table 10.a).

CCI

[redacted]

AEs that fulfill 1 or more of the serious criteria above are to be considered SAEs and should be reported and followed up in the same manner (see Sections 10.1 and 10.1.1).

10.1.2 Special Interest AEs

CCI

[redacted]

10.2 AE Procedures

10.2.1 Assigning Severity/Intensity of AEs

The different categories of severity/intensity are:

Mild: An AE that is usually transient and may require only minimal treatment or therapeutic intervention. The event does not generally interfere with usual activities of daily living.

Moderate: An AE that is usually alleviated with additional specific therapeutic intervention. The event interferes with usual activities of daily living, causing discomfort but poses no significant or permanent risk of harm to the research participant.

Severe: An AE that interrupts usual activities of daily living, or significantly affects clinical status, or may require intensive therapeutic intervention.

10.2.2 Assigning Causality of AEs

The relationship of each AE to study medication(s) will be assessed using the following categories:

Related: An AE that follows a reasonable temporal sequence from administration of a drug (including the course after withdrawal of the drug), or for which a causal relationship is at least a reasonable possibility, ie, the relationship cannot be ruled out, although factors other than the drug, such as underlying diseases, complications, concomitant drugs and concurrent treatments, may also be responsible.

Not Related: An AE that does not follow a reasonable temporal sequence from administration of a drug and/or that can reasonably be explained by other factors, such as underlying diseases, complications, concomitant medications and concurrent treatments.

10.2.3 Start Date

The start date of the AE is the date that the first signs/symptoms were noted by the subject and/or investigator.

10.2.4 End Date

The end date of the AE is the date at which the subject recovered, the event resolved but with sequelae, or the subject died.

10.2.5 Pattern of AE (Frequency)

Episodic AEs (eg, headache) or those that occur repeatedly over a period of consecutive days are intermittent. All other events are continuous.

10.2.6 Action Taken With Study Treatment

Drug withdrawn or study procedures stopped: a study medication or [^{18}F]MNI-1054 PET scans stopped due to the particular AE.

- Dose not changed: the particular AE did not require stopping a study medication or study procedures.
- Unknown: only to be used if it has not been possible to determine what action has been taken.
- Not applicable: a study medication or study procedure was stopped for a reason other than the particular AE (eg, the study has been terminated, the subject died, dosing with study medication had not yet started, or dosing with study medication was already stopped before the onset of the AE).
- Dose reduced: the dose was reduced due to the particular AE.
- Dose increased: the dose was increased due to the particular AE.

- Drug interrupted: the dose was interrupted due to the particular AE.

10.2.7 Outcome

- Recovered/resolved: subject returned to first assessment status with respect to the AE.
- Recovering/resolving: the intensity is lowered by 1 or more stages: the diagnosis or signs/symptoms have almost disappeared; the abnormal laboratory value improved but has not returned to the normal range or to the baseline value; the subject died from a cause other than the particular AE with the condition remaining "recovering/resolving."
- Not recovered/not resolved: there is no change in the diagnosis, signs or symptoms; the intensity of the diagnosis, signs/symptoms or laboratory value on the last day of the observed study period has become worse than when it started; is an irreversible congenital anomaly; the subject died from another cause with the particular AE state remaining "Not recovered/not resolved."
- Recovered/resolved with sequelae: the subject recovered from an acute AE but was left with permanent/significant impairment (eg, recovered from a cardiovascular accident but with some persisting paresis).
- Fatal: an AE that is considered as the cause of death.
- Unknown: the course of the AE cannot be followed up due to hospital change or residence change at the end of the subject's participation in the study.

10.2.8 Collection and Reporting of AEs, SAEs, Special Interest AEs, and Abnormal LFTs

10.2.8.1 Collection Period

PTE monitoring begins after signing of the informed consent form until [^{18}F]MNI-1054 dose; subsequently, collection of AEs (ie, AEs, SAEs, special interest AEs, and abnormal LFTs) begins. Routine collection of AEs will continue through the Day 14 (±4) safety follow-up safety visit. For subjects who discontinue before the administration of study medication, AEs will be followed until the subject discontinues study participation.

10.2.8.2 Reporting AEs

At each study visit, the investigator will assess whether any subjective AEs have occurred. A neutral question, such as "How have you been feeling since your last visit?" may be asked. Subjects may report AEs occurring at any other time during the study. Subjects experiencing an SAE before the first exposure to investigational product must be monitored until the symptoms subside and any clinically relevant changes in laboratory values have returned to baseline, or until there is a satisfactory explanation for the change. Nonserious AEs that begin before the first exposure to investigational product, related or unrelated to the study procedure, need not be followed up for the purposes of the protocol.

All subjects experiencing AEs, whether considered associated with the use of the study medication or not, must be monitored until the symptoms subside and any clinically relevant changes in laboratory values have returned to baseline or until there is a satisfactory explanation for the changes observed. All AEs will be documented in the AE page of the eCRF, whether or not the investigator concludes that the event is related to the drug treatment. The following information will be documented for each event:

- Event term.
- Start and end date and time.
- Pattern of AE (frequency).
- Severity/intensity.
- Causality (investigator's opinion of the causal relationship between the event and administration of study drug[s]).
- Action taken with study drug.
- Outcome of event.
- Seriousness.

10.2.8.3 Reporting SAEs

When an SAE occurs through the AE collection period, it should be reported according to the procedure outlined below:

A Takeda SAE form must be completed, in English, and signed by the investigator immediately or within 24 hours of first onset or notification of the event. The information should be completed as fully as possible but contain, at a minimum:

- A short description of the event and the reason why the event is categorized as serious.
- Subject identification number.
- Investigator's name.
- Name of the study medication(s).
- Causality assessment.

The SAE form should be transmitted within 24 hours to the attention of the contact listed in Section 14.1.1.

The paper SAE forms should be submitted via fax (preferred method). In case of fax, site personnel need to confirm successful transmission of all pages and include an email address on the fax cover sheet so that an acknowledgment of receipt (AOR) can be returned via email within 1 business day.

Email submission of SAE forms with a PDF attachment should only be used in the case where fax is not possible within 24 hours of receiving the event. In case of email, site personnel need to confirm successful transmission by awaiting an AOR via email within 1 business day.

If SAE forms are submitted via fax or email, a confirmation of receipt will be sent to the sites indicating that the SAE has been received. It is the site's responsibility to ensure an AOR has been obtained when email or fax are used. If AOR is not received within 1 business day, the site investigator should escalate it immediately to their clinical research associate.

Any SAE spontaneously reported to the investigator following the AE collection period should be reported to the sponsor if considered related to study participation.

Reporting of SAEs that begin before first administration of investigational product will follow the same procedure for SAEs occurring on treatment.

SAE Follow-up

If information is not available at the time of the first report becomes available at a later date, the investigator should complete a follow-up SAE form or provide other written documentation and fax it immediately within 24 hours of receipt. Copies of any relevant data from the hospital notes (eg, ECGs, laboratory tests, discharge summary, postmortem results) should be sent to the addressee, if requested.

All SAEs should be followed up until resolution or permanent outcome of the event. The timelines and procedure for follow-up reports are the same as those for the initial report.

10.2.8.4 Reporting of Abnormal LFTs

If a subject is noted to have ALT or AST elevated >3 times the ULN on 2 consecutive occasions, the abnormality should be recorded as an AE. In addition, an LFT increase eCRF must be completed to provide additional information on relevant recent history, risk factors, clinical signs and symptoms, and results of any additional diagnostic tests performed.

If a subject is noted to have ALT or AST >3 times the ULN and total bilirubin >2 times the ULN for which an alternative etiology has not been identified, the event should be recorded as an SAE and reported per Section 10.2.8.3. The investigator must contact the medical monitor for discussion of the relevant subject details and possible alternative etiologies, such as acute viral hepatitis A or B or other acute liver disease. Follow-up laboratory tests as described in Section 9.3 must also be performed. In addition, the relevant eCRF(s) must be completed and transmitted with the Takeda SAE form (per Section 10.2.9).

10.2.9 Safety Reporting to Investigators, IRBs or IECs, and Regulatory Authorities

The sponsor will be responsible for reporting all suspected unexpected serious adverse reactions (SUSARs) and any other applicable SAEs to regulatory authorities, investigators, and IRBs or IECs, as applicable, in accordance with national regulations in the countries where the study is conducted. Relative to the first awareness of the event by/or further provision to the sponsor or sponsor's designee, SUSARs will be submitted within 7 days for fatal and life-threatening events

and 15 days for other serious events, unless otherwise required by national regulations. The sponsor will also prepare an expedited report for other safety issues where these might materially alter the current benefit-risk assessment of an investigational medicinal product or that would be sufficient to consider changes in the investigational medicinal products administration or in the overall conduct of the study. The investigational site also will forward a copy of all expedited reports to his or her IRB or IEC in accordance with national regulations.

11.0 STATISTICAL METHODS

11.1 Statistical and Analytical Plans

A statistical analysis plan (SAP) will be prepared and finalized before database lock. This document will provide further details regarding the definition of analysis variables and analysis methodology to address all study objectives.

A targeted data review will be conducted before database lock. This review will assess the accuracy and completeness of the study database, subject evaluability, and appropriateness of the planned statistical methods.

11.1.1 Analysis Sets

11.1.1.1 Safety Set

The safety analysis set will consist of all subjects who are enrolled and receive an investigational drug ([^{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.

11.1.1.2 PK Set

The PK set will consist of all subjects who receive study drug (TAK-418) and have at least 1 measurable plasma concentration of TAK-418.

11.1.1.3 PET Set

The PET target occupancy set will consist of all subjects who receive study drug (TAK-418) and have a technically adequate baseline PET scan and at least 1 technically adequate post–TAK-418 dose PET scan.

If any subjects are found to have 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.

11.1.2 Analysis of Demography and Other Baseline Characteristics

Descriptive statistics (N, mean, SD, median, minimum, and maximum) will be generated for continuous demographic variables and baseline characteristics variables (eg, age, height, weight, and BMI) for TAK-418 dose level, and overall. The number and percentage of subjects in each class of the categorical demographic variables and baseline characteristics variables (eg, sex, ethnicity, race, smoking status) will also be tabulated. Individual subject demographic and baseline characteristics data will be listed.

11.1.3 PK Analysis

The concentrations of TAK-418 in plasma will be summarized by dose level over each scheduled sampling time point (each of the 2 post–TAK-418 dose PET scans) using descriptive statistics. Individual plasma concentration data versus time will be presented in a data listing.

PK parameters of TAK-418 will be summarized using descriptive statistics.

A more detailed analysis will be presented in the SAP.

11.1.4 PET Analysis

11.1.4.1 PET Enzyme Occupancy and Brain Kinetics for TAK-418

Dynamic uptake and washout of [^{18}F]MNI-1054 will be evaluated. The MRI and PET images will be coaligned for anatomy-based definition of ROIs for analysis of regional [^{18}F]MNI-1054 binding. Analysis will focus on the decay-corrected time activity data in brain regions of high uptake (eg, cerebellum). Imaging data will be analyzed with modeling approaches appropriate for irreversible binding that will be informed by the ongoing FIH validation study TAK-418-0002 (I[CCI]) (eg, irreversible 2-tissue compartment model). The primary imaging outcome measure for [^{18}F]MNI-1054 is expected to be a parameter from such a model.

LSD1 enzyme occupancy in ROI after a single dose of TAK-418 will be obtained from the model binding parameter (eg, inhibitory constant [K_i]) for the first postdose scan as follows:

Occupancy (1st postdose) = (K_i [baseline] - K_i [1st postdose]) / Ki (baseline)

The second postdose scan will be used to assess the rate of enzyme turnover in humans. The data will be analyzed like the first postdose scan to calculate an apparent occupancy value:

Apparent occupancy (2nd postdose) = (K_i [baseline] - K_i [2nd postdose]) / K_i (baseline).

Descriptive summaries for modeled binding parameters and target occupancy will be provided.

11.1.4.2 Relationship of PET Enzyme Occupancy to Plasma TAK-418 Levels

Modeling will be performed to assess the relationship between target occupancy and TAK-418 plasma exposure. The relationship between LSD1 enzyme occupancy and TAK-418 exposure measures will be graphically explored. If appropriate, linear or nonlinear models may be used to quantify this information. The TAK-418 plasma exposure and dose required for LSD1 occupancy of at least 90% will be established.

11.1.5 Safety Analysis

AEs will be presented in listings, and TEAEs will be summarized.

Individual results of laboratory tests (hematology, serum chemistry, and urinalysis), vital signs and ECG parameters will be listed. Baseline, postdose, and change from baseline to postdose laboratory data, vital signs, and ECG parameters will be summarized. All safety summaries will be performed for [^{18}F]MNI-1054 and each TAK-418 dose level.

Physical examination findings, visual examinations, and suicidal assessments will be presented in data listings.

11.2 Interim Analysis and Criteria for Early Termination

No interim analysis is planned.

11.3 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.

12.0 QUALITY CONTROL AND QUALITY ASSURANCE

12.1 Study-Site Monitoring Visits

Monitoring visits to the study site will be made periodically during the study to ensure that all aspects of the protocol are followed. Source documents will be reviewed for verification of data recorded on the eCRFs. Source documents are defined as original documents, data, and records. The investigator and study site guarantee access to source documents by the sponsor or its designee (contract research organization) and by the IRB or IEC.

All aspects of the study and its documentation will be subject to review by the sponsor or the sponsor's designee (as long as blinding is not jeopardized), including but not limited to the investigator's binder, study drug, subject medical records, informed consent documentation, and review of eCRFs and associated source documents. It is important that the investigator and other study personnel are available during the monitoring visits and that sufficient time is devoted to the process.

12.2 Protocol Deviations

The investigator should not deviate from the protocol, except where necessary to eliminate an immediate hazard to study subjects. Should other unexpected circumstances arise that will require deviation from protocol-specified procedures, the investigator should consult with the sponsor or designee (and IRB or IEC, as required) to determine the appropriate course of action. There will be no exemptions (a prospectively approved deviation) from the inclusion or exclusion criteria.

Significant deviations include, but are not limited to, those that involve fraud or misconduct, increase the health risk to the subject, or confound interpretation of primary study assessment.

12.3 Quality Assurance Audits and Regulatory Agency Inspections

The study site also may be subject to quality assurance audits by the sponsor or designees. In this circumstance, the sponsor-designated auditor will contact the site in advance to arrange an auditing visit. The auditor may ask to visit the facilities where laboratory samples are collected, where the medication is stored and prepared, and any other facility used during the study. In addition, there is the possibility that this study may be inspected by regulatory agencies, including those of foreign governments (eg, the Food and Drug Administration [FDA], the United Kingdom Medicines and Healthcare products Regulatory Agency, the Pharmaceuticals and Medical Devices Agency of Japan). If the study site is contacted for an inspection by a regulatory body, the sponsor should be notified immediately. The investigator guarantees access for quality assurance auditors to all study documents as described in Section 12.1.

13.0 ETHICAL ASPECTS OF THE STUDY

This study will be conducted with the highest respect for the individual participants (ie, subjects) according to the protocol, the ethical principles that have their origin in the Declaration of Helsinki, and the International Council for Harmonisation (ICH) Harmonised Tripartite Guideline for GCP. Each investigator will conduct the study according to applicable local or regional regulatory requirements and align his or her conduct in accordance with the "Responsibilities of the Investigator" that are listed in Appendix A. The principles of Helsinki are addressed through the protocol and through appendices containing requirements for informed consent and investigator responsibilities.

13.1 IRB and/or IEC Approval

IRBs and IECs must be constituted according to the applicable state and federal/local requirements of each participating region. The sponsor or designee will require documentation noting all names and titles of members who make up the respective IRB or IEC. If any member of the IRB or IEC has direct participation in this study, written notification regarding his or her abstinence from voting must also be obtained. Those Americas sites unwilling to provide names and titles of all members due to privacy and conflict of interest concerns should instead provide a Federal Wide Assurance Number or comparable number assigned by the Department of Health and Human Services.

The sponsor or designee will supply relevant documents for submission to the respective IRB or IEC for the protocol's review and approval. This protocol, the investigator's brochure, a copy of the informed consent form, and, if applicable, subject recruitment materials and/or advertisements and other documents required by all applicable laws and regulations, must be submitted to a central or local IRB or IEC for approval. The IRB's or IEC's written approval of the protocol and subject informed consent must be obtained and submitted to the sponsor or designee before commencement of the study (ie, before shipment of the sponsor-supplied drug or study specific screening activity). The IRB or IEC approval must refer to the study by exact protocol title, number, and version date; identify versions of other documents (eg, informed consent form) reviewed; and state the approval date. The sponsor will ship drug/notify site once the sponsor has confirmed the adequacy of site regulatory documentation and, when applicable, the sponsor has received permission from competent authority to begin the study. Until the site receives drug/notification no protocol activities, including screening, may occur.

Sites must adhere to all requirements stipulated by their respective IRB or IEC. This may include notification to the IRB or IEC regarding protocol amendments, updates to the informed consent form, recruitment materials intended for viewing by subjects, local safety reporting requirements, reports and updates regarding the ongoing review of the study at intervals specified by the respective IRB or IEC, and submission of the investigator's final status report to IRB or IEC. All IRB and IEC approvals and relevant documentation for these items must be provided to the sponsor or its designee.

Subject incentives should not exert undue influence for participation. Payments to subjects must be approved by the IRB or IEC and sponsor.

13.2 Subject Information, Informed Consent, and Subject Authorization

Written consent documents will embody the elements of informed consent as described in the Declaration of Helsinki and the ICH Guidelines for GCP and will be in accordance with all applicable laws and regulations. The informed consent form, subject authorization form (if applicable), and subject information sheet (if applicable) describe the planned and permitted uses, transfers, and disclosures of the subject's personal and personal health information for purposes of conducting the study. The informed consent form and the subject information sheet (if applicable) further explain the nature of the study, its objectives, and potential risks and benefits, as well as the date informed consent is given. The informed consent form will detail the requirements of the participant and the fact that he or she is free to withdraw at any time without giving a reason and without prejudice to his or her further medical care.

The investigator is responsible for the preparation, content, and IRB or IEC approval of the informed consent form and, if applicable, the subject authorization form. The informed consent form, subject authorization form (if applicable), and subject information sheet (if applicable) must be approved by both the IRB or IEC and the sponsor before use.

The informed consent form, subject authorization form (if applicable), and subject information sheet (if applicable) must be written in a language fully comprehensible to the prospective subject. It is the responsibility of the investigator to explain the detailed elements of the informed consent form, subject authorization form (if applicable), and subject information sheet (if applicable) to the subject. Information should be given in both oral and written form whenever possible and in the manner deemed appropriate by the IRB or IEC. In the event the subject is not capable of rendering adequate written informed consent, then the subject's legally acceptable representative may provide such consent for the subject in accordance with applicable laws and regulations.

The subject, or the subject's legally acceptable representative, must be given ample opportunity to: (1) inquire about details of the study, and (2) decide whether or not to participate in the study. If the subject, or the subject's legally acceptable representative, determines he or she will participate in the study, then the informed consent form and subject authorization form (if applicable) must be signed and dated by the subject, or the subject's legally acceptable representative, at the time of consent and before the subject entering into the study. The subject or the subject's legally acceptable representative should be instructed to sign using their legal names, not nicknames, using blue or black ballpoint ink. The investigator must also sign and date the informed consent form and subject authorization (if applicable) at the time of consent and before subject entering into the study; however, the sponsor may allow a designee of the investigator to sign to the extent permitted by applicable law.

Once signed, the original informed consent form, subject authorization form (if applicable), and subject information sheet (if applicable) will be stored in the investigator's site file. The investigator must document the date the subject signs the informed consent in the subject's medical record. Copies of the signed informed consent form, the signed subject authorization form (if applicable), and subject information sheet (if applicable) shall be given to the subject.

All revised informed consent forms must be reviewed and signed by relevant subjects or the relevant subject's legally acceptable representative in the same manner as the original informed consent. The date the revised consent was obtained should be recorded in the subject's medical record, and the subject should receive a copy of the revised informed consent form.

Subjects who consented and provided a pharmacogenomic sample for DNA and RNA analysis can withdraw their consent and request disposal of a stored sample at any time before analysis. Notify sponsor of consent withdrawal.

13.3 Subject Confidentiality

The sponsor and designees affirm and uphold the principle of the subject's right to protection against invasion of privacy. Throughout this study, a subject's source data will only be linked to the sponsor's clinical study database or documentation via a unique identification number. As permitted by all applicable laws and regulations, limited subject attributes, such as sex, age, or date of birth, and subject initials may be used to verify the subject and accuracy of the subject's unique identification number.

To comply with ICH Guidelines for GCP and to verify compliance with this protocol, the sponsor requires the investigator to permit its monitor or designee's monitor, representatives from any regulatory authority (eg, FDA, Medicines and Healthcare products Regulatory Agency, Pharmaceuticals and Medical Devices Agency), the sponsor's designated auditors, and the appropriate IRBs and IECs to review the subject's original medical records (source data or documents), including, but not limited to, laboratory test result reports, ECG reports, admission and discharge summaries for hospital admissions occurring during a subject's study participation, and autopsy reports. Access to a subject's original medical records requires the specific authorization of the subject as part of the informed consent process (see Section 13.2).

Copies of any subject source documents that are provided to the sponsor must have certain personally identifiable information removed (ie, subject name, address, and other identifier fields not collected on the subject's eCRF).

13.4 Publication, Disclosure, and Clinical Trial Registration Policy

13.4.1 Publication and Disclosure

The investigator is obliged to provide the sponsor with complete test results and all data derived by the investigator from the study. During and after the study, only the sponsor may make study information available to other study investigators or to regulatory agencies, except as required by law or regulation. Except as otherwise allowable in the clinical study site agreement, any public disclosure (including publicly accessible websites) related to the protocol or study results, other than study recruitment materials and/or advertisements, is the sole responsibility of the sponsor.

The sponsor may publish any data and information from the study (including data and information generated by the investigator) without the consent of the investigator. Manuscript authorship for any peer-reviewed publication will appropriately reflect contributions to the production and review of the document. All publications and presentations must be prepared in accordance with

this section and the clinical study site agreement. In the event of any discrepancy between the protocol and the clinical study site agreement, the clinical study site agreement will prevail.

13.4.2 Clinical Trial Registration

In order to ensure that information on clinical trials reaches the public in a timely manner and to comply with applicable laws, regulations and guidance, Takeda will, at a minimum register all interventional clinical trials it sponsors anywhere in the world on ClinicalTrials.gov and/or other publicly accessible websites before start of study, as defined in Takeda policy/standard. Takeda contact information, along with investigator's city, state (for Americas investigators), country, and recruiting status will be registered and available for public viewing.

For some registries, Takeda will assist callers in locating study sites closest to their homes by providing the investigator name, address, and phone number to the callers requesting trial information. Once subjects receive investigator contact information, they may call the site requesting enrollment into the trial. The investigative sites are encouraged to handle the trial inquiries according to their established subject screening process. If the caller asks additional questions beyond the topic of trial enrollment, they should be referred to the sponsor.

Any investigator who objects to the sponsor providing this information to callers must provide the sponsor with a written notice requesting that their information not be listed on the registry site.

13.4.3 Clinical Trial Results Disclosure

Takeda will post the results of clinical trials on ClinicalTrials.gov or other publicly accessible websites, as required by Takeda policy/standard, applicable laws and/or regulations.

13.5 Insurance and Compensation for Injury

Each subject in the study must be insured in accordance with the regulations applicable to the site where the subject is participating. If a local underwriter is required, then the sponsor or sponsor's designee will obtain clinical study insurance against the risk of injury to study subjects. Refer to the study site agreement regarding the sponsor's policy on subject compensation and treatment for injury. If the investigator has questions regarding this policy, he or she should contact the sponsor or sponsor's designee.

14.0 ADMINISTRATIVE AND REFERENCE INFORMATION

14.1 Administrative Information

14.1.1 Study Contact Information

Contact Type / Role	Contact
Serious adverse event and pregnancy reporting	PPD

14.1.2 INVESTIGATOR AGREEMENT

I confirm that I have read and that I understand this protocol, the investigator's brochure, package insert, and any other product information provided by the sponsor. I agree to conduct this study in accordance with the requirements of this protocol and also to protect the rights, safety, privacy, and well-being of study subjects in accordance with the following:

- The ethical principles that have their origin in the Declaration of Helsinki.
- ICH, E6 GCP: Consolidated Guideline.
- All applicable laws and regulations, including, without limitation, data privacy laws and regulations.
- Regulatory requirements for reporting SAEs defined in Section 10.2.9 of this protocol.
- Terms outlined in the study site agreement.
- Responsibilities of the investigator (Appendix A).

I further authorize that my personal information may be processed and transferred in accordance with the uses contemplated in Appendix C of this protocol.

_____

Signature of Investigator Date

_____

Investigator Name (print or type)

_____

Investigator's Title

_____

Location of Facility (City, State/Province)

_____

Location of Facility (Country)

14.1.3 List of Abbreviations

%ID	Percent injected dose
AE	adverse event
ALT	alanine aminotransferase
AOR	acknowledgment of receipt
ASD	autism spectrum disorder
AST	aspartate aminotransferase
AUC_{24}	area under the concentration-time curve from time 0 to 24 hours
AUC_{∞}	area under the concentration-time curve from time 0 to infinity
AUC_t	area under the concentration-time curve from time 0 to time t
BMI	body mass index
CFR	Code of Federal Regulations
C_{max}	maximum observed concentration
CSF	cerebrospinal fluid
C-SSRS	Columbia-Suicide Severity Rating Scale
ECG	electrocardiogram
eCRF	electronic case report form
ED	effective dose
FAD	flavin adenine dinucleotide
FDA	Food and Drug Administration
F-FAD	formylated flavin adenine dinucleotide
FIH	first-in-human
FSH	follicle-stimulating hormone
GCP	Good Clinical Practice
H3K4	lysine in position 4 of type 3 histone
HBsAg	hepatitis B surface antigen
HBV	hepatitis B virus
hCG	human chorionic gonadotropin
HCV	hepatitis C virus
ICH	International Council for Harmonisation
ICRP	International Commission on Radiological Protection
IEC	independent ethics committee
IRB	institutional review board
IUD	intrauterine device
IUS	intrauterine hormone-releasing system
K_i	inhibitory constant
LFT	liver function test
LSD1	lysine-specific demethylase 1A, also known as KDM1A
MRD	multiple-rising dose
MRI	magnetic resonance imaging

NOAEL	no-observed-adverse-event-level
PD	pharmacodynamic(s)
PET	positron emission tomography
PK	pharmacokinetic(s)
PTE	pretreatment event
QD	once daily
QTcF	QT interval with Fridericia correction method
rem	Roentgen equivalent man
ROI	regions of interest
SAE	serious adverse event
SAP	statistical analysis plan
SRD	single-rising dose
SUSAR	suspected unexpected serious adverse reactions
TAC	time-activity curve
TEAE	treatment-emergent adverse event
ULN	upper limit of normal

15.0 DATA HANDLING AND RECORDKEEPING

The full details of procedures for data handling will be documented in the data management plan. AEs, medical history, and concurrent conditions will be coded using the Medical Dictionary for Regulatory Activities. Drugs will be coded using the World Health Organization Drug Dictionary.

15.1 eCRFs

Completed eCRFs are required for each subject who receives any investigational drug ([18F]MNI-1054 or TAK-418) as part of this study.

The sponsor or its designee will supply investigative sites with access to eCRFs. The sponsor will make arrangements to train appropriate site staff in the use of the eCRF. These forms are used to transmit the information collected in the performance of this study to the sponsor and regulatory authorities. eCRFs must be completed in English. Data are transcribed directly onto eCRFs.

After completion of the entry process, computer logic checks will be run to identify items, such as inconsistent dates, missing data, and questionable values. Queries may be issued by Takeda personnel (or designees) and will be answered by the site.

Corrections are recorded in an audit trail that captures the old information, the new information, identification of the person making the correction, the date the correction was made, and the reason for change. Reasons for significant corrections should additionally be included.

The principal investigator must review the eCRFs for completeness and accuracy and must sign and date the appropriate eCRFs as indicated. Furthermore, the investigator must retain full responsibility for the accuracy and authenticity of all data entered on the eCRFs.

After the lock of the clinical study database, any change of, modification of, or addition to the data on the eCRFs should be made by the investigator with use of change and modification records of the eCRFs. The principal investigator must review the data change for completeness and accuracy and must sign and date.

eCRFs will be reviewed for completeness and acceptability at the study site during periodic visits by study monitors. The sponsor or its designee will be permitted to review the subject's medical and hospital records pertinent to the study to ensure accuracy of the eCRFs. The completed eCRFs are the sole property of the sponsor and should not be made available in any form to third parties, except for authorized representatives of appropriate governmental health or regulatory authorities, without written permission of the sponsor.

15.2 Record Retention

The investigator agrees to keep the records stipulated in Section 15.1 and those documents that include (but are not limited to) the study-specific documents, the identification log of all participating subjects, medical records, temporary media such as thermal sensitive paper, source worksheets, all original signed and dated informed consent forms, subject authorization forms regarding the use of personal health information (if separate from the informed consent forms), electronic copy of eCRFs, including the audit trail, and detailed records of drug disposition to

enable evaluations or audits from regulatory authorities, the sponsor or its designees. Any source documentation printed on degradable thermal sensitive paper should be photocopied by the site and filed with the original in the subject's chart to ensure long-term legibility. Furthermore, ICH E6 Section 4.9.5 requires the investigator to retain essential documents specified in ICH E6 (Section 8) until at least 2 years after the last approval of a marketing application for a specified drug indication being investigated or, if an application is not approved, until at least 2 years after the investigation is discontinued and regulatory authorities are notified. In addition, ICH E6 Section 4.9.5 states that the study records should be retained until an amount of time specified by applicable regulatory requirements or for a time specified in the clinical study site agreement between the investigator and sponsor.

Refer to the clinical study site agreement for the sponsor's requirements on record retention. The investigator and the head of the institution should contact and receive written approval from the sponsor before disposing of any such documents.

16.0 REFERENCES

1. Burg JM, Link JE, Morgan BS, Heller FJ, Hargrove AE, McCafferty DG. KDM1 class flavin-dependent protein lysine demethylases. Biopolymers 2015;104(4):213-46.
2. Sanders SJ, He X, Willsey AJ, Ercan-Sencicek AG, Samocha KE, Cicek AE, et al. Insights into Autism Spectrum Disorder Genomic Architecture and Biology from 71 Risk Loci. Neuron 2015;87(6):1215-33.
3. Shen E, Shulha H, Weng Z, Akbarian S. Regulation of histone H3K4 methylation in brain development and disease. Philos Trans R Soc Lond B Biol Sci 2014;369(1652).
4. Rudolph T, Beuch S, Reuter G. Lysine-specific histone demethylase LSD1 and the dynamic control of chromatin. Biol Chem 2013;394(8):1019-28.

17.0 APPENDICES

CCI

[redacted]

CCI

[redacted]

Appendix B Elements of the Subject Informed Consent

CCI

[redacted]

CCI

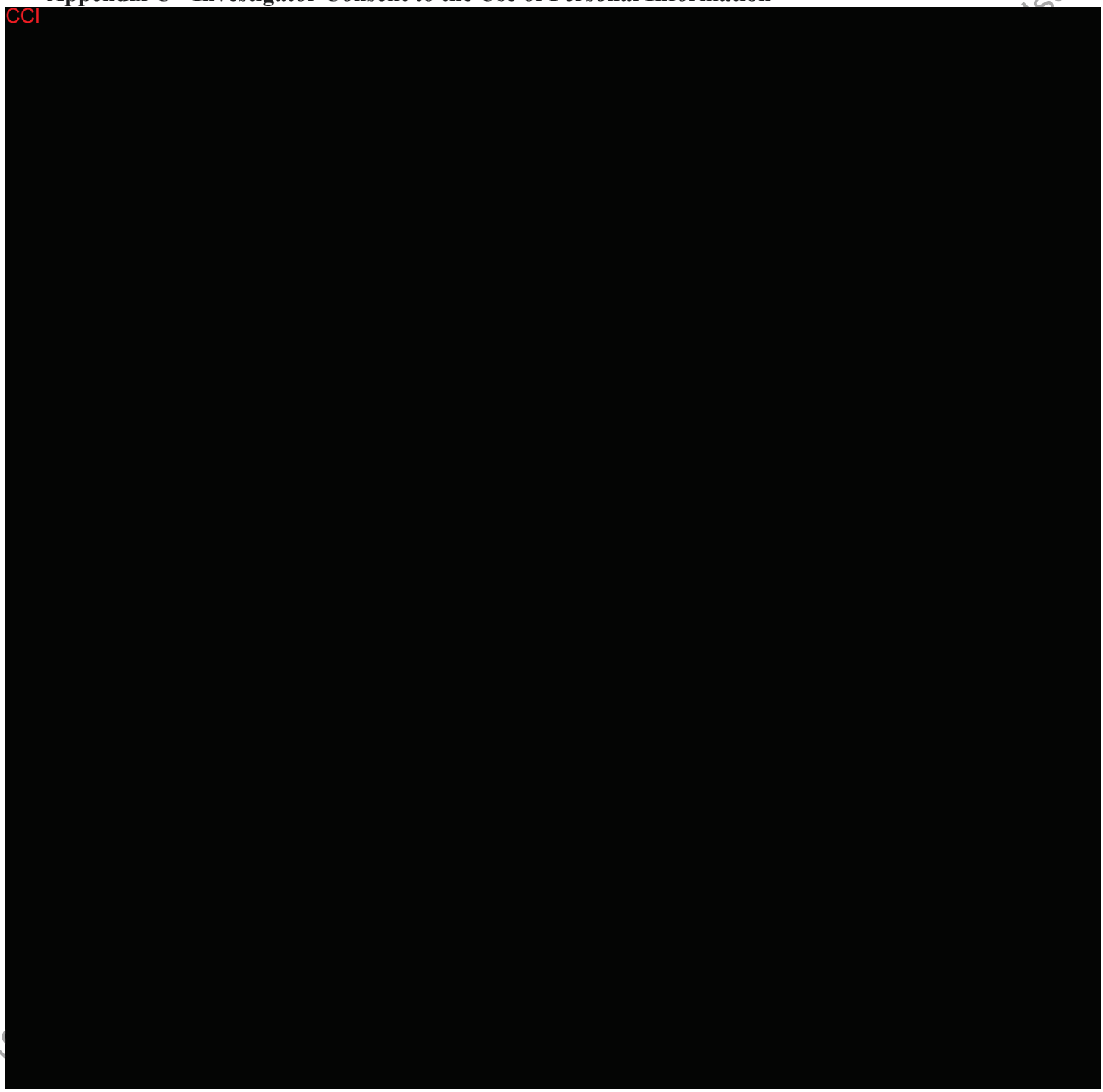
[redacted]

CCI

[redacted]

Appendix C Investigator Consent to the Use of Personal Information

CCI



Appendix D Pregnancy and Contraception

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Appendix E Detailed Description of Amendments to Text

CCI

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[redacted]

ELECTRONIC SIGNATURES

Signed by	Meaning of Signature	Server Date (dd-MMM-yyyy HH:mm 'UTC')
PPD	Clinical Pharmacology Approval	05-Mar-2020 14:05 UTC
	Biostatistics Approval	05-Mar-2020 14:26 UTC
	Clinical Approval	05-Mar-2020 14:38 UTC