IMAGING TAU IN ALZHEIMER'S DISEASE AND NORMAL AGING

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Study Product: ^{18}*F-THK-5351*

Protocol Number: *AAAQ7868*

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List of Abbreviations

PET	Positron emission tomography
MRI	Magnetic resonance imaging
AD	Alzheimer's disease
MCI	Mild cognitive impairment
AD	Alzheimer's disease
SUVR	Standardized uptake value ratio
^{18}F-THK-5351	((S)-1-(fluoro-^{18}F)-3-((2-(6-(methylamino)pyridin-3-yl)quinolin-6-yl)oxy)propan-2-ol).
mCi	millicurie
MBq	Megabecquerel
μSv	microSievert
μGy	microGray

Study Summary

Title	Imaging tau in Alzheimer's disease and normal aging
Short Title	Imaging tau in Alzheimer's disease and normal aging
Protocol Number	AAAQ7868
Phase	II
Methodology	This is a single center PET study.

Screening will include history and physical examination, brain MRI, and neuropsychological examination. Subjects who meet criteria for Alzheimer's disease (AD) or mild cognitive impairment (MCI), or who are cognitively normal will then have one brain PET scan with ^{18}F-THK-5351. Vital signs will be checked prior to injection of ^{18}F-THK-5351, then at the completion of the PET scan.

Subjects will have the option of having a lumbar puncture to measure CSF concentrations of markers of inflammation and neurodegeneration. |
Study Duration	5 years
Study Center(s)	Taub Institute, CUMC
Objectives	The primary objective is to determine the relationship between tau, normal aging, and Alzheimer's disease.
Number of Subjects	Up to 60 of these subjects will have ^{18}F-THK-5351. PET. Subjects will be 50 years and older. Thirty subjects will have normal cognition and the other thirty will have cognitive impairment.

Diagnosis and Main Inclusion Criteria	Inclusion criteria: 1. Age 50 and older 2. Meet criteria for either a) amnestic mild cognitive impairment (single or mixed domain) or mild Alzheimer's disease, or b) have no cognitive impairment, based on history, exam, neuropsychological testing, and consensus diagnosis. MCI and mild AD patients must have Clinical Dementia Rating scale score of 0.5 or 1. Unimpaired subjects must have Clinical Dementia Rating scale score of 0. 3. Subjects unable to provide informed consent must have a surrogate decision maker 4. Written and oral fluency in English or Spanish 5. Able to participate in all scheduled evaluations and to complete all required tests and procedures. 6. In the opinion of the investigator, the subject must be considered likely to comply with the study protocol and to have a high probability of completing the study. Exclusion criteria: 1. Past or present history of certain brain disorders other than MCI or AD. 2. Certain significant medical conditions, which make study procedures of the current study unsafe. Such serious medical conditions include uncontrolled epilepsy and multiple serious injuries. 3. Contraindication to MRI scanning 4. Conditions precluding entry into the scanners (e.g. morbid obesity, claustrophobia, etc.). 5. History of kidney disease or presence of impaired kidney function based on laboratory tests at screening visit. Benign or resolved kidney conditions such as uncomplicated kidney stones or treated infection would be allowed if kidney function is normal at screening visit. 6. History of liver disease or presence of impaired liver function based on laboratory tests at screening visit. Benign or resolved liver conditions such as uncomplicated cysts or resolved viral infections would be allowed if liver function is normal at screening visit. 7. Participation in the last year in a clinical trial for a disease modifying drug for AD. 8. Inability to have a catheter in subject's vein for the injection of radioligand. 9. Inability to have blood drawn from subject's veins.
Study Product, Dose, Route, Regimen	^{18}F-THK-5351, up to 5 mCi (185 MBq), IV, total of one injection.

Duration of administration	A single dose of radioligand will be injected over 1 minute for each PET scan.
Reference therapy	N/A
Statistical Methodology	*Comparing PET imaging among groups* The primary outcome measure will be regional standardized uptake value ratio (SUVR) values for ^{18}F-THK-5351 using cerebellum as reference region. Multiple brain regions will be measured, with particular attention to medial temporal cortex structures such as hippocampus and entorhinal cortex. Analysis will be performed using a one-way ANOVA with cognitive status (AD vs. MCI vs. normal) as independent variable. Effect of age and education on ^{18}F-THK-5351 will be determined in post-hoc analysis. *Sample size determination* Results from a small published study showed greater ^{18}F-THK-5351 binding in hippocampus in AD patients (n = 3) than controls (n = 3), with effect size of 1.95. This would allow us to detect differences in ^{18}F-THK-5351 between AD patients and controls in hippocampus with n = 10 subjects per group. However, we don't know the effect size when MCI patients are compared to AD patients and when MCI patients are compared to controls. We wish to scan up to 60 subjects total (30 impaired and 30 unimpaired) to ensure power to see effect on ^{18}F-THK-5351 binding between groups. We will also perform linear regression to look for correlations between change in ^{18}F-THK-5351 binding and cognitive scores, CSF concentration of biomarkers of inflammation and neurodegeneration, and atrophy measured with MRI.

1 Introduction

This document is a protocol for a human research study. This study is to be conducted according to US and international standards of Good Clinical Practice (FDA Title 21 part 312 and International Conference on Harmonization guidelines), applicable government regulations and Institutional research policies and procedures.

1.1 Background

1.1.1. Background on Alzheimer's disease

While amyloid plaques are a pathological hallmark of Alzheimer's disease (AD), whether β-amyloid is the causal agent of neurodegeneration in AD remains unclear. Evidence supporting the hypothesis that amyloid is a primary contributor to AD pathogenesis (the amyloid-first hypothesis) include the finding that genetic mutations responsible for inherited forms of AD cause increased amyloid production. Studies using positron emission tomography (PET) to measure amyloid plaque burden in vivo have demonstrated amyloid positivity before obvious atrophy or cognitive decline in healthy older controls. While tau positive neurofibrillary tangles represent the second hallmark of AD, tau mutations are seen in patients with variants of frontotemporal dementia, not AD. The propagation of tau pathology is thought to follow amyloidosis, suggesting amyloid in some way induces tau mediated degeneration. However, the link between amyloid and tau not clear as there are spatial and temporal distinctions. For example, bulk of amyloid burden in medial parietal cortex and frontal cortex early on, while tau burden begins in medial temporal cortex.

While β-amyloid has been shown to have direct neurotoxic effects in vitro, these effects are seen at concentrations of β-amyloid much higher than found in human brain[1].Therefore, amyloid may not confer significant direct toxicity. In addition, autopsy findings of pre-tangle pathology occurring in midlife prior to amyloidosis[2] suggest that tauopathy may begin prior and independent to significant amyloid plaque deposition. Since amyloid burden correlates poorly with cognition[3], tau may be a more important mediator of neurodegeneration in AD.

4.1.1. Background on tau pathology in neurodegenerative diseases.

Deposition of hyperphosphorylated tau protein is observed in several neurodegenerative diseases including Alzheimer's disease, progressive supranuclear palsy, corticobasal degeneration, chronic traumatic encephalopathy, and frontotemporal lobar degeneration [4]. Tau is a microtubular protein and its native function is to provide structural support to neurons. Paired helical filaments composed of dysfunctional tau protein are found in several neurodegenerative diseases. In Alzheimer's disease, the clinical progression of dementia has been shown to correlate with the amount and topographical spread of tau throughout the brain. Therefore, detecting and quantifying tau aggregate load in brain would have diagnostic and prognostic potential in clinical management of several neurological diseases. As disease modifying drugs that target tau are being developed, there is a critical need for a reliable method of detecting tau aggregates to confirm pathology in patients entering clinical trials.

4.1.2. Background on PET imaging of tau

Positron emission tomography (PET) imaging allows in vivo measurement of protein density after administration of radiolabeled ligands that bind to the target protein. ^{18}F-FDDNP has demonstrated increased binding in patients with tauopathies such as Alzheimer's disease [5-

8] and progressive supranuclear palsy [9]. Reports using this radioligand found increased binding in former NFL players in medial temporal and brainstem structures known to be affected in chronic traumatic encephalopathy [10, 11]. However, FDDNP binds to both fibrillar amyloid and tau, so the PET signal is nonselective and the greater density of amyloid plaque than tau-positive tangles is expected to confound measurement of tau in patients with co-pathology. The more recently developed radioligand ^{18}F-T807 (now ^{18}F-AV-1451) has been shown to have greater affinity for tau aggregates than amyloid plaques. While only a small number of studies using ^{18}F-AV-1451 have been published to date [12, 13], several clinical trials are currently employing this radioligand.

[11]C-PBB3 has high affinity and high selectivity for tau [14]. Human studies to date have included healthy controls, patients with Alzheimer's disease, and one patient with corticobasal syndrome. However, ^{11}C-PBB3 has limitations in that radiometabolites enter the brain [15] and the radioligand undergoes photo-isomerization in fluorescent light [16].

Other alternative tau radioligands include ^{18}F-THK-5117, which has greater binding in AD patients than controls in expected distribution of tangle pathology [17]. However, this compound has a relatively high amount of white matter binding that could confound accurate quantification of tau aggregation in vivo. Nevertheless, ^{18}F-THK-5117 binding increases in patients with AD, but not healthy controls over time, and this increase correlates with worsening of clinical symptoms[18].

^{18}F-THK-5351 is a recently developed radioligand with lower nonspecific binding than ^{18}F-THK-5117. Ex vivo studies show that binding of ^{3}H-THK-5351 colocalizes to tau immunostaining in AD brain tissue, with lower white matter binding than seen with the earlier tau radioligand THK-5117[19]. Therefore, this recently developed radioligand has promise for reliable quantification of tau pathology in Alzheimer's disease and other tauopathies.

1.2 Investigational Agent

^{18}F-THK-5351 is a PET radioligand that binds to paired helical filament tau aggregates. This radioligand has only been used for research purposes. ^{18}F-THK-5351 will be administered in tracer doses (≤10 μg) at activity of up to 5 mCi (185 MBq) per injection.

2 Study Objectives

2.1. The primary objective is to determine the extent and spatial distribution of tau at different stages along the cognitive spectrum of aging (cognitively normal older adults, adults with mild cognitive impairment, and adults with Alzheimer's disease dementia).

2.2. Secondary objectives are to determine how ^{18}F-THK-5351 binding in brain relates to cognitive performance, amyloid binding, atrophy on MRI, and CSF concentrations of markers of inflammation and neurodegeneration.

3 Study Design

3.1 General Design

3.1.1. Subject recruitment and screening procedures

Men and women age 50 and older will be recruited from the CUMC Washington Heights-Inwood Community Aging Project (known locally as WHICAP, R01 AG037212, PI Richard Mayeux), the CUMC Alzheimer's Disease Research Center (ADRC), and Dr. Davangere Devanand's Questionable Dementia 2 (QD2) cohort, and CUMC clinics. Subjects may also be co-enrolled in Columbia University Medical Center protocols AAAO1151 (PI William Kreisl), AAAO9758 (PI Adam Brickman), AAAI2752 (PI Yaakov Stern), AAAQ8249 (PI Yaakov Stern), and AAAQ8096 (PI Yaakov Stern). Recruitment may come from additional research cohorts, as this protocol will encourage data sharing to reduce the total number of subjects that require ^{18}F-THK-5351 imaging for project completion. Only subjects from established research cohorts that previously consented to be contacted about future research studies will be approached about this study. Informed consent will be obtained prior to study enrollment.

For subjects with cognitive impairment, prior to obtaining informed consent, a licensed physician who is not listed as personnel on this protocol will determine whether the subjects has capacity to provide their own consent. If the subject lacks consent, a legally authorized representative will be required to provide consent for the subject.

Up to 60 subjects will undergo screening including history, physical examination, routine laboratory studies, neuropsychological testing, and brain MRI. Subjects will be identified as either cognitively normal or cognitively impaired in consensus conference. Subjects must have Clinical Dementia Rating scale score of 0.5 or 1 and meet clinical criteria for either amnestic MCI (single or multiple-domain)[20] or Alzheimer's disease[21] to be included in the cognitively impaired category.

Target sample size is 30 elders with impairment and 30 elders without impairment (60 subjects total).

3.1.2 Neuropsychological data analysis

Selected neuropsychological tests scores will be combined into four composite scores (memory, language, executive/speed, and visuospatial). Memory testing will include Selective Reminding Test and Benton Visual Retention Test (BVRT), Recognition Memory Multiple Choice version. Z-scores for each cognitive measure will be calculated and averaged to create a composite z-score for each domain. These factor domain scores will be subsequently averaged to produce a composite cognitive z-score to indicate mean cognition. Higher z-score indicates better cognitive performance.

3.1.3. Imaging procedures

One brain MRI will be performed on each subject using a 3 T Philips scanner. Sequences performed will include 3D T1 (MPRAGE, 180 slice 1 mm resolution, 256 x 256 voxel count) for volumetric analysis and clinical sequences to exclude subjects with significant intracranial pathology unrelated to AD, such as malignant brain tumor of subdural hematoma. PET scans will take place on a Biograph mCT PET scanner (Siemens Healthcare) at the CUMC Kreitchman PET Center. Subjects will have one PET scan with ^{18}F-THK-5351 (injected activity up to 5 mCi = 185 MBq). PET imaging will be performed without arterial sampling. Vital signs (blood pressure, heart rate, respiratory rate, and temperature) will be checked prior to injection of ^{18}F-THK-5351, then at the completion of the PET scan. ^{18}F-THK-5351 will be synthesized by the CUMC PET Department Radiochemistry Laboratory. Subjects will not be informed of ^{18}F-THK-5351 PET scan results, as this scan is used only in research and have not yet been validated for clinical use. Subjects will be informed if a clinically important abnormality is detected on MRI or PET imaging (e.g., brain tumor).

3.1.4 Image processing

FreeSurfer (http://surfer.nmr.mgh.harvard.edu/), the MRI software package comprising a suite of automated tools for segmentation, reconstruction, and derivation of regional volumes and surface-based rendering, will be used for derivation of regions-of-interest (ROIs). Eleven ROIs will be extracted from the structural T1 image: entorhinal cortex, hippocampus, inferior temporal cortex, combined superior and middle temporal cortex, superior parietal lobule, inferior parietal lobule, precuneus, occipital cortex, prefrontal cortex, striatum, and thalamus.

^{18}F-THK-5351 PET images will be analyzed as follows: Freesurfer based ROIs will be applied to coregistered PET images. Correction for partial volume effects using a region-based voxel-wise method[22] will be applied. Regional time-activity curves using 40-60 min of scan data will be extracted from the PET scans, including cerebellum which will be used as a reference region. Standardized uptake value ratio (SUVR) values will be calculated by dividing SUV values for each target region by that of the cerebellum

3.1.5. Cerebrospinal fluid sampling and analysis

Subjects will have the option of having one lumbar puncture (LP) for CSF collection. Subjects may refuse the LP and still participate in the other study procedures. CSF analysis will be performed to determine CSF concentrations of total tau, phospho-tau, β-amyloid, and markers of inflammation such as IL-1β, TNF-α, glial fibrillary acidic protein, S100B, and YLK-40.

3.1.6. Procedures performed in other protocols

If any of the above procedures have been performed within one year of the ^{18}F-THK-5351 PET scan, then the procedures will not need to be repeated under this protocol. Rather the previously obtained results will be used.

3.2 Primary Study Endpoints

Because the drugs used in this study are radioligands given at tracer doses, there are no clinical endpoints of the study.

The primary outcome measures are:

1. Amount of ^{18}F-THK-5351 binding.

Secondary outcome measures are:
1. CSF concentration of inflammatory markers
2. CSF concentration of β-amyloid , tau and phospho-tau

3.3 Secondary Study Endpoints
N/A

3.4 Primary Safety Endpoints
N/A

4 Subject Selection and Withdrawal

4.1 Inclusion Criteria
1. Age 50 and older
2. Meet criteria for either a) amnestic mild cognitive impairment (single or mixed domain) or mild Alzheimer's disease, or b) have no cognitive impairment based on history, exam, neuropsychological testing, and consensus diagnosis. MCI and mild AD patients must have Clinical Dementia Rating scale score of 0.5 or 1. Unimpaired subjects must have Clinical Dementia Rating scale score of 0.
3. Subjects unable to provide informed consent must have a surrogate decision maker
4. Written and oral fluency in English or Spanish
5. Able to participate in all scheduled evaluations and to complete all required tests and procedures.
6. In the opinion of the investigator, the subject must be considered likely to comply with the study protocol and to have a high probability of completing the study.

4.2 Exclusion Criteria
1. Past or present history of certain brain disorders other than MCI or AD.

2. Certain significant medical conditions, which make study procedures of the current study unsafe. Such serious medical conditions include uncontrolled epilepsy and multiple serious injuries.
3. Contraindication to MRI scanning
4. Conditions precluding entry into the scanners (e.g. morbid obesity, claustrophobia, etc.).
5. History of kidney disease or presence of impaired kidney function based on laboratory tests at screening visit. Benign or resolved kidney conditions such as uncomplicated kidney stones or treated infection would be allowed if kidney function is normal at screening visit.
6. History of liver disease or presence of impaired liver function based on laboratory tests at screening visit. Benign or resolved liver conditions such as uncomplicated cysts or resolved viral infections would be allowed if liver function is normal at screening visit.
7. Participation in the last year in a clinical trial for a disease modifying drug for AD.
8. Inability to have a catheter in subject's vein for the injection of radioligand.
9. Inability to have blood drawn from subject's veins.

Impaired kidney function is defined here as estimated glomerular filtration rate < 60 mL/min/1.73m^2 at screening. Impaired liver function is defined here as aspartate aminotransferase, alanine aminotransferase, or alkaline phosphatase > 2 times upper limit of normal at screening visit.

4.3 Subject Recruitment and Screening

Up to 60 elderly men and women will be recruited from CUMC Washington Heights-Inwood Community Aging Project (known locally as WHICAP, R01 AG037212, PI Richard Mayeux), the CUMC Alzheimer's Disease Research Center (ADRC), and Dr. Davangere Devanand's Questionable Dementia 2 (QD2) cohort, and CUMC clinics. Recruitment may come from additional research cohorts, as this protocol will encourage data sharing to reduce the total number of subjects that require ^{18}F-THK-5351 imaging for project completion. Only subjects from established research cohorts that previously consented to be contacted about future research studies will be approached about this study. Informed consent will be obtained by a qualified investigator prior to study enrollment. Subjects will undergo screening including history, physical examination, routine laboratory studies, neuropsychological testing, and brain MRI to identify cohorts who are either cognitively impairment, meeting criteria for MCI or AD, (target n = 30), or cognitively normal (n = 30). Subjects co-enrolled from other protocols may have any of the above screening procedures done through the earlier study, as long as screening laboratories, MRI, and neuropsychological testing are done within 12 months of ^{18}F-THK-5351 imaging.

During screening, participants will be administered a neuropsychological test battery and a medical interview. Individuals with significant cognitive or functional impairment are referred for evaluation by a board-certified physician. All participants' data will be reviewed in a consensus conference where a working diagnosis is established. The diagnostic criteria for dementia will be based on DSM-IV criteria. The diagnosis of MCI or AD will be based on updated criteria[20, 21]. The neuropsychological test battery will include the following tests: the Mini-Mental State Examination, Selective Reminding Test, Benton Visual Retention Test (BVRT), Recognition Memory Multiple Choice version, Rosen Drawing Test, 5 selected items,

BVRT Matching, multiple-choice version, Boston Naming Test, 15 selected items, The Controlled Word Association Test and Category naming: Animals, Food and Clothing, Complex Ideational Material Subtest of the Boston Diagnostic Aphasia Evaluation (BDAE): first 6 items, The Color Trails Test part 1, WAIS-R Similarities, and Color Trails part 2. At minimum, subjects will receive testing in Selective Reminding Test, Trail Making Test, and Verbal Fluency (CFL, and Animals). Raw scores and demographically corrected T-scores (age, years of education, sex, and ethnicity) will be determined. Subjects will also undergo University of Pennsylvania Smell Identification Test (UPSIT), a non-invasive test that measures odor identification abilities.

4.4 Early Withdrawal of Subjects

4.4.1 When and How to Withdraw Subjects

Subjects will be withdrawn if they develop serious medical illness during the study, defined as an event determined to be grade 3 or higher, i.e., severe or life-threatening. The exception is a single event of syncope if felt by the PI to be vasovagal in etiology and related to catheter placement or venipuncture. Vasovagal syncope in response to blood drawing and IV placement is common and does not warrant study withdrawal if the subject's loss of consciousness is brief (i.e., less than 5 minutes) and does not require further medical attention.

4.4.2 Data Collection and Follow-up for Withdrawn Subjects

Data collected prior to withdrawal will be analyzed if possible. No data collection will take place after withdrawal in subjects who drop out of the study.

5 Study Drug

5.1 Description

^{18}F-THK-5351 is a PET radioligand that binds to tau aggregates. ^{18}F-THK-5351 will be administered at tracer doses and is not expected to have a pharmacological effect.

5.2 Treatment Regimen

N/A

5.3 Method for Assigning Subjects to Treatment Groups

N/A

5.4 Preparation and Administration of Study Drug

^{18}F-THK-5351 will be synthesized and administered by the CUMC PET Department. ^{18}F-THK-5351 injection will be administered at a target imaging dose of up to 5 mCi (185 MBq).

Calculation of activity to be injected will be performed by a qualified radiopharmacist in the CUMC PET department.

5.5 Subject Compliance Monitoring

N/A

5.6 Prior and Concomitant Therapy

Subjects will not be included in the study if they have participated in the last year in a clinical trial for a disease modifying drug for AD.

5.7 Packaging

N/A

5.8 Blinding of Study Drug

N/A

5.9 Receiving, Storage, Dispensing and Return

5.9.1 Receipt of Drug Supplies

Upon receipt of the of the study treatment supplies, the CUMC PET Center staff will perform an inventory and a drug receipt log will be filled out and signed by the person accepting the shipment. Any damaged or unusable study drug in a given shipment (active drug or comparator) will be documented in the study files.

5.9.2 Storage

N/A

5.9.3 Dispensing of Study Drug

^{18}F-THK-5351 will be administered the same day of synthesis by the CUMC PET Department.

5.9.4 Return or Destruction of Study Drug

Any unused radiopharmaceutical will be on site and documented in the study files.

6 Study Procedures

This study will involve up to 5 outpatient visits. See schedule below.

At the first screening visit, subjects will sign informed consent, undergo history and physical and neurological examination, neuropsychological testing, and have blood drawn routine safety laboratories. Additional screening visits may be required to complete all procedures. Additional visits will be required for brain MRI. Flexibility is allowed in screening procedures such that procedures may be performed in any order and may be performed on the same day if schedule allows for subject convenience. Subjects who already had screening procedures performed under a previous research protocol do not need to have them repeated unless performed more than 12

months prior to the ^{18}F-THK-5351 PET scan. A total of 3 screening visits are anticipated. However, additional visits may be necessary if certain tests must be scheduled on different days due to scheduling.

Up to two study visits will be required. One visit will be for the ^{18}F-THK-5351 PET scan and the second for the optional lumbar puncture. Subjects may decline the lumbar puncture and still participate in other parts of the study. If schedule allows, the ^{18}F-THK-5351 PET and LP could be performed on the same day.

^{18}F-THK-5351 PET scans and LP must be completed within 12 months of the brain MRI.

Study Schedule

Study Procedures	Screen [a]	Study [b]	
	≤12 months	Visit 1	Visit 2
Informed consent	X		
Medical/psychiatric history	X		
Inclusion/exclusion criteria	X	X	X
Neuropsychological testing	X		
MRI	X		
^{18}F-THK-5351 PET [c]		X	
Lumbar puncture [c, d]			X
Laboratory determinations [e]	X	X	
Neurological examination	X		
Vital signs	X		
Height and weight	X	X	
Adverse events [f]	X	X	X

a. Screening procedures may be performed in any order and may be performed on the same day if schedule allows for subject convenience. Subjects who already had screening procedures performed under a different protocol do not need to have them repeated unless performed more than 12 months prior to inclusion in this study. Up to 3 screening visits are anticipated. However, additional visits may be necessary if certain tests must be scheduled on different days due to scheduling.
b. Study procedures must be completed within 12 months of MRI.
c. ^{18}F-THK-5351 PET and LP may be performed on same day.
d. Lumbar puncture is optional.
e. Laboratory determinations here include complete blood count, basic metabolic panel, liver functions tests, thyroid stimulating hormone, and urinalysis.
f. Adverse event reporting to be performed at Visit 1 after the PET scan is completed. Subject will be called by telephone 24 hours after the PET scan for follow up adverse event reporting.

7 Statistical Plan

7.1 Sample Size Determination

Because ^{18}F-THK-5351 is a recently developed radioligand, we do not have adequate preliminary data for a formal power calculation. However, results from a small published study showed greater ^{18}F-THK-5351 binding in hippocampus in AD patients (n = 3) than controls (n = 3), with effect size of 1.95[19]. This would allow us to detect differences in ^{18}F-THK-5351 between AD patients and controls in hippocampus with n = 10 subjects per group. However, we don't know the effect size when MCI patients are compared to AD patients and when MCI

patients are compared to controls. We wish to scan up to 60 subjects total (30 impaired and 30 unimpaired) to ensure power to see effect on ^{18}F-THK-5351 binding between groups.

7.2 Statistical Methods

The primary outcome measure will be regional standardized uptake value ratio (SUVR) values for ^{18}F-THK-5351 using cerebellum as reference region. Multiple brain regions will be measured, with particular attention to medial temporal cortex structures such as hippocampus and entorhinal cortex. Analysis will be performed using a one-way ANOVA with cognitive status (AD vs. MCI vs. normal) as independent variable. Effect of age and education will be determined in post-hoc analysis.

Regression models will be fit for ^{18}F-THK-5351 binding as dependent variable and cognitive scores, voxel count on MRI, and CSF concentrations of markers of inflammation and neurodegeneration as independent variables.

7.3 Subject Population(s) for Analysis

Subjects will be elders age 50 and older with normal cognition, mild cognitive impairment, or mild AD.

8 Safety and Adverse Events

8.1 Definitions

Adverse Event

An *adverse event* (AE) is any symptom, sign, illness or experience that develops or worsens in severity during the course of the study. Intercurrent illnesses or injuries should be regarded as adverse events. Abnormal results of diagnostic procedures are considered to be adverse events if the abnormality:

- results in study withdrawal
- is associated with a serious adverse event
- is associated with clinical signs or symptoms
- leads to additional treatment or to further diagnostic tests
- is considered by the investigator to be of clinical significance

Serious Adverse Event

Adverse events are classified as serious or non-serious. A *serious adverse event* is any AE that is:

- fatal
- life-threatening
- requires or prolongs hospital stay
- results in persistent or significant disability or incapacity
- a congenital anomaly or birth defect
- an important medical event

Important medical events are those that may not be immediately life threatening, but are clearly of major clinical significance. They may jeopardize the subject, and may require intervention to prevent one of the other serious outcomes noted above. For example, drug overdose or abuse, a seizure that did not result in in-patient hospitalization, or intensive treatment of bronchospasm in an emergency department would typically be considered serious.

All adverse events that do not meet any of the criteria for serious should be regarded as *non-serious adverse events*.

Adverse Event Reporting Period

The study period during which adverse events must be reported is normally defined as the period from the initiation of any study procedures to the end of the study treatment follow-up. For this study, the study drug follow-up is defined as 24 hours following administration of study drug. Subjects will receive a telephone call 24 hours after the PET scan for adverse event reporting.

Preexisting Condition

A preexisting condition is one that is present at the start of the study. A preexisting condition will be recorded as an adverse event if the frequency, intensity, or the character of the condition worsens during the study period.

General Physical Examination Findings

At screening, any clinically significant abnormality will be recorded as a preexisting condition. At the end of the study, any new clinically significant findings/abnormalities that meet the definition of an adverse event will also be recorded and documented as an adverse event.

Post-study Adverse Event

All unresolved adverse events will be followed by the investigator until the events are resolved, the subject is lost to follow-up, or the adverse event is otherwise explained. At the last scheduled visit, the investigator will instruct each subject to report any subsequent event(s) that the subject, or the subject's personal physician, believes might reasonably be related to participation in this study. The investigator will notify IRB and FDA of any death or adverse event occurring at any time after a subject has discontinued or terminated study participation that may reasonably be related to this study.

Abnormal Laboratory Values

A clinical laboratory abnormality will be documented as an adverse event if <u>any one of the following</u> conditions is met:

- The laboratory abnormality is not otherwise refuted by a repeat test to confirm the abnormality
- The abnormality suggests a disease and/or organ toxicity
- The abnormality is of a degree that requires active management; e.g. change of dose, discontinuation of the drug, more frequent follow-up assessments, further diagnostic investigation, etc.

Hospitalization, Prolonged Hospitalization or Surgery

Any adverse event that results in hospitalization or prolonged hospitalization will be documented and reported as a serious adverse event unless specifically instructed otherwise in this protocol. Any condition responsible for surgery should be documented as an adverse event if the condition meets the criteria for and adverse event.

Neither the condition, hospitalization, prolonged hospitalization, nor surgery are reported as an adverse event in the following circumstances:

- Hospitalization or prolonged hospitalization for diagnostic or elective surgical procedures for a preexisting condition. Surgery should ***not*** be reported as an outcome of an adverse event if the purpose of the surgery was elective or diagnostic and the outcome was uneventful.
- Hospitalization or prolonged hospitalization required to allow efficacy measurement for the study.
- Hospitalization or prolonged hospitalization for therapy of the target disease of the study, unless it is a worsening or increase in frequency of hospital admissions as judged by the clinical investigator.

8.2 Recording of Adverse Events

At each contact with the subject, the investigator will seek information on adverse events (AEs) by specific questioning and, as appropriate, by examination. Information on all adverse events will be recorded immediately in the source document. All clearly related signs, symptoms, and abnormal diagnostic procedures results will be recorded in the source document.

All adverse events occurring during the study period will be recorded. The clinical course of each event will be followed until resolution, stabilization, or until it has been determined that the study treatment or participation is not the cause. Serious adverse events that are still ongoing at the end of the study period will be followed up to determine the final outcome. Any serious adverse event that occurs after the study period and is considered to be possibly related to the study treatment or study participation will be recorded and reported immediately.

As per CUMC IRB's Policy, Unanticipated Problems will be defined as follows:

Unanticipated Problem (UP) is any incident, experience or outcome involving risk to subjects or others in any human subjects research that meets all of the following criteria:

• Unexpected (in terms of nature, severity or frequency) given (a) the research procedures that are described in the IRB-approval protocol and informed consent document and (b) the characteristics of the subject population being studied;
• Related or possibly related to participation in such research (i.e., there is a reasonable possibility that the incident, experience or outcome may have been caused by the procedures involved in such research); and
• Suggests that the research places subjects or others at a greater risk of harm (including physical, psychological, economic or social harm) than was previously known or recognized (HHS IRB Guidance, Section I).

The following AEs will be considered Unexpected Problems:

• A single occurrence of a serious, unexpected event that is uncommon and strongly associated with drug exposure
• A single occurrence, or a small number of occurrences, of a serious, unexpected event that is not commonly associated with drug exposure
• Multiple occurrences of an AE that, based on an aggregate analysis, are determined not to be isolated occurrences and involve risk to human subjects
• An AE that is described or addressed in the investigator's brochure, protocol or informed consent documents (a Described AE), but occurs at a specificity or severity that is inconsistent with prior observations
• A serious Described AE, but for which the rate of occurrence represents a clinically significant increase in the expected rate of occurrence
• Any other AE or safety finding that would cause the sponsor to modify the investigator's brochure, study protocol or informed consent documents or would prompt other action by the IRB to ensure the protection of human subjects (2009 FDA Guidance, Section III (A)).

8.3 Reporting of Serious Adverse Events

8.3.1 IRB Notification by Investigator

At the time of the Occurrence of an Unanticipated Problem:

Each UP will be reported to the IRB, whether or not (a) it is serious or non-serious or (b) it occurs at a site at which the PI is conducting the research.

The UP will be reported promptly, but not later than one week following the occurrence of the UP or the PI's acquiring knowledge of the UP.

The PI will make the determination as to whether an incident, experience or outcome constitutes a UP.

Each Unanticipated Problem will be reported to the IRB using the Unanticipated Problem Report module in Rascal.

The investigator must conclude in the Unanticipated Problem Report whether the protocol and/or consent form(s) should be modified as the result of the UP. If the protocol and/or consent document(s) requires a revision, a modification must be submitted in Rascal.

At the Time of Continuing Review of a Protocol:

At the time of continuing review of a protocol, the PI will submit a summary of all UPs that occurred during the review period and since the beginning of the study. The summary for each UP should include:

• The number of subjects who experienced the UP;

• The investigator's determination as to whether or not the UP was serious;

• The investigator's determination as to the UP's relationship to the study procedures (e.g., definitely related, probably related or possibly related).

8.3.2 FDA Notification by Investigator

The principal investigator shall notify the FDA by telephone or by facsimile transmission of any unexpected fatal or life-threatening experience associated with the use of the drug as soon as possible but no later than 7 calendar days from the investigator's original receipt of the information.

If a previous adverse event that was not initially deemed reportable is later found to fit the criteria for reporting, the principal investigator will submit the adverse event in a written report to the FDA as soon as possible, but no later than 15 calendar days from the time the determination is made.

8.4 Unblinding Procedures

N/A

8.5 Stopping Rules

N/A

8.6 Medical Monitoring

We will not have a specific Data and Safety Monitoring Board. Adverse events will be documented in the patient's chart. Notification of UPs to the IRB and SUSARs to the FDA will take place as described in Section 8.3.

9 Data Handling and Record Keeping

9.1 Confidentiality

Information about study subjects will be kept confidential and managed according to the requirements of the Health Insurance Portability and Accountability Act (HIPAA). Those regulations require a signed subject authorization informing the subject of the following:

- What protected health information (PHI) will be collected from subjects in this study

- Who will have access to that information and why

- Who will use or disclose that information

- The rights of a research subject to revoke their authorization for use of their PHI.

In the event that a subject revokes authorization to collect or use PHI, the investigator, by regulation, retains the ability to use all information collected prior to the revocation of subject authorization. For subjects that have revoked authorization to collect or use PHI, attempts should be made to obtain permission to collect at least vital status (i.e. that the subject is alive) at the end of their scheduled study period.

9.2 Source Documents

Source documents will be used to record data for this study. Source documents will be kept in secure location. For the PET visit, a checklist will be used to ensure all study procedures are completed.

9.3 Case Report Forms

Case report forms (CRFs) will be used as the primary research data collecting tool. CRFs may also be source documents in some instances. CRFs will be kept in a secure location.

10 Study Monitoring, Auditing, and Inspecting

10.1 Study Monitoring Plan

This study will be monitored by the Principal Investigator. Dr. Kreisl will submit annual progress reports to the FDA within 60 days of the anniversary of the date that the IND became active (the date clinical studies were permitted to begin) in accordance with 21 CFR 312.33.

11 Ethical Considerations

This study is to be conducted according to US and international standards of Good Clinical Practice (FDA Title 21 part 312 and International Conference on Harmonization guidelines), applicable government regulations and Institutional research policies and procedures.

This protocol and any amendments will be submitted to a properly constituted independent Institutional Review Board (IRB), in agreement with local legal prescriptions, for formal approval of the study conduct. The decision of the IRB concerning the conduct of the study will be made in writing to the investigator and a copy of this decision will be provided to the sponsor before commencement of this study. The investigator should provide a list of EC/IRB members and their affiliate to the sponsor.

All subjects for this study will be provided a consent form describing this study and providing sufficient information for subjects to make an informed decision about their participation in this study. See attached copy of the Subject Informed Consent Form. This consent form will be submitted with the protocol for review and approval by the IRB for the study. The formal consent of a subject, using the IRB-approved consent form, must be obtained before that subject is submitted to any study procedure. This consent form must be signed by the subject or legally acceptable surrogate, and the investigator-designated research professional obtaining the consent.

12 Study Finances

12.1 Funding Source

This study is funded by a Rose and Boris Katz Assistant Professorship endowed to Dr. Kreisl. Additional funding is being sought through an NIA K23 (PI = Dr. Kreisl).

12.2 Conflict of Interest

Any investigator who has a conflict of interest with this study (patent ownership, royalties, or financial gain greater than the minimum allowable by their institution, etc.) must have the conflict reviewed by a properly constituted Conflict of Interest Committee with a Committee-sanctioned conflict management plan that has been reviewed and approved by the study sponsor prior to participation in this study. All Columbia University Medical Center investigators will follow the University conflict of interest policy.

12.3 Subject Stipends or Payments

Subjects will be compensated commensurate to similar studies at CUMC. We wish to reduce the burden of travel and inconvenience of study procedures on patients and caregivers. Subjects will be given $100 for the screening visit, $100 for the MRI, $100 for the PET scan, and $150 for the lumbar puncture.

13 Publication Plan

Neither the complete nor any part of the results of the study carried out under this protocol, nor any of the information provided by the sponsor for the purposes of performing the study, will be published or passed on to any third party without the consent of the study sponsor. Any investigator involved with this study is obligated to provide the sponsor with complete test results and all data derived from the study.

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