SIPO1-AD: A Phase II Clinical Trial for the Assessment of Safety, Tolerability, and Efficacy of Siponimod in Patients with Mild Alzheimer's Disease Dementia

Protocol Number: SIPO

National Clinical Trial (NCT) Identified Number: 06639282

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IND Sponsor: Barrow Neurological Institute, Phoenix, AZ 85013

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STATEMENT OF COMPLIANCE

The trial will be conducted in accordance with International Conference on Harmonization Good Clinical Practice (ICH GCP) and the following:

United States (US) Code of Federal Regulations (CFR) applicable to clinical studies (45 CFR Part 46, 21 CFR Part 50, 21 CFR Part 56, 21 CFR Part 312, and/or 21 CFR Part 812)

National Institutes of Health (NIH)-funded investigators and clinical trial site staff who are responsible for the conduct, management, or oversight of NIH-funded clinical trials have completed Human Subjects Protection and ICH GCP Training.

The protocol, informed consent form(s), recruitment materials, and all participant materials will be submitted to the Institutional Review Board (IRB) for review and approval. Approval of both the protocol and the consent form must be obtained before any participant is enrolled. Any amendment to the protocol will require review and approval by the IRB before the changes are implemented to the study. In addition, all changes to the consent form will be IRB-approved; a determination will be made regarding whether a new consent needs to be obtained from participants who provided consent, using a previously approved consent form.]

1. PROTOCOL SUMMARY

SYNOPSIS

Title:	SIPO1-AD: A Phase II Clinical Trial for the Assessment of Safety, Tolerability, and Efficacy of Siponimod in Patients with Mild Alzheimer's Disease
Study Description:	Alzheimer's disease (AD) is a neurodegenerative disorder with several complex neuropathologies suspected to develop sequentially but that overlap over time as symptoms progress to dementia. Thus, to be effective, future intervention strategies will likely require combination therapies or pleiotropic agents to tackle several AD molecular pathogenic pathways simultaneously. For more than a decade, our group has been exploring the repurposing of immunomodulators for AD. Recent discussions with collaborators who specialize in multiple sclerosis suggest that sphingosine-1-phosphate receptor (S1PR) modulators are strong candidates for repurposing in AD. Indeed, S1PR modulators are blood brain barrier (BBB) penetrant and display pleiotropic actions, including immunomodulation and neuroprotective properties. S1P is a versatile endogenous molecule that regulates several signaling pathways by binding to five G-protein-coupled receptors, which are expressed in high levels in cardiac, vascular, immune, and brain cells. This widespread localization of S1PR was the historical basis for Novartis Pharmaceuticals, Inc, to develop oral formulations of S1PR modulators for multiple sclerosis (MS), which proved successful and resulted in two marketed oral compounds (i.e., fingolimod and Siponimod). In the present project, we intend to collaborate with Novartis to use the most recently FDA-approved S1PR modulator, Siponimod. Based on MS and animal experimentation literature, we hypothesize that Siponimod could alter the rate of neurodegeneration as measured by lowering the rate of brain atrophy in mild AD dementia subjects. In this Phase II, proof-of-concept, translational clinical study, mild AD dementia subjects will be randomized 2:1 and will receive gradual titration regimen of Siponimod starting at 0.25mg/day and increasing up to final dosage of 1 mg/day (N=70) or placebo (N=35) for 12 months. This will be followed by a 6-month washout period. Siponimod has demonstrated positive immunomodulatory and neuroprotective actions in MS patients, and because its toxicity profile is favorable for use in older individuals, Siponimod has a strong potential to alter markers of mild AD dementia pathology and disease trajectory.
Objectives:	The primary objectives are to evaluate the safety and tolerability of the drug in subjects with mild Alzheimer's disease dementia, as assessed through regular clinical evaluations throughout the dosing period. Critically, eventual treatment-emergent toxicities will drive our decision process to pursue or stop dosing. The secondary objective is to determine drug effect on relative annual brain atrophy rates in the two groups by comparing pre- and post-exposure volumetric MRI data. Tertiary objectives will be evaluated through the utilization of cognitive assessments throughout study, The study design will support two exploratory objectives using blood, CSF, and saliva biospecimen collection from subjects with mild AD dementia. Blood and CSF samples will be analyzed for AD biomarkers (amyloid beta, tau, and p-tau)

	as well as CSF inflammatory markers. Saliva samples will be assessed for amyloid-beta biomarkers (primarily Aβ40 and Aβ42), which may serve as dynamic surrogate markers of therapeutic drug efficacy.
Endpoints:	Primary Endpoint: Safety to be evaluated through reported adverse events (AEs), clinical laboratory tests (hematology, and serum chemistry,), vital signs, physical and neurological examinations, and electrocardiograms (EKGs). Secondary Endpoints: Change in rates of neurodegeneration as measured with volumetric MRI. Tertiary Endpoint: Cognitive changes throughout duration of the study to be evaluated through cognitive assessments, including, ADCOMS (with ADAS-Cog13, MMSE, CDR-SOB as source measures) Exploratory: ADCS-ADL + CSF/blood/saliva biomarkers
Study Population:	105 male and female outpatients, 50-85 years of age, diagnosed with early/mild Alzheimer's disease dementia primarily residing in Phoenix, AZ and greater surrounding areas. Because of the local demographics, the study population will mostly consist of Caucasians, although all efforts will be made to enroll other ethnicities.
Inclusion and Exclusion Criteria	<u>Inclusion Criteria</u> To be eligible for this study, subjects must meet the following inclusion criteria: 1. Male or female at least 50 years of age, but less than 85 (84 at time of screening) 2. Females must be of non-childbearing potential or have negative pregnancy test at time of screening. Women of non-childbearing potential are defined as women who are either permanently sterilized (hysterectomy, bilateral oophorectomy or bilateral salpingectomy) or who are postmenopausal. Women will be considered postmenopausal if they have been amenorrheic for >12 months prior to the planned date of enrollment. 3. Must have a diagnosis of mild Alzheimer's Dementia determined by medical record review. 4. Vision and hearing must be sufficient to comply with study procedures. 5. Be able to take oral medications. 6. Must be able to attend all study visits indicated in the schedule of visits. 7. Must have a collateral informant/study partner who has significant direct contact with the patient at least 10 hours per week and who is willing to accompany the patient to specified clinic visits, supervise administration of all study medication, and be available for telephone visits/interviews. 8. Documented Mini Mental State Exam (MMSE) score between 20-26 at Screening Visit Day 1. 9. CT or MRI scan of the brain within 12 months of Screening Visit Day 1 showing no evidence of significant focal lesions or other pathology which could contribute to dementia. If neither a CT or a MRI scan is available from

the past 12 months, a scan fulfilling the requirements must be obtained before randomization.

10. Hachinski ischemic score must be ≤ 4.
11. Geriatric depression scale must be ≤ 10.
12. Prior to dosing all randomized study subjects must show proof they have received immunization to varicella (VZV IgG).
13. Each patient must be assessed for capacity to consent by the principal or sub-investigators in order to provide informed consent. If the patient is deemed unable to provide informed consent, they must have a legally authorized representative (LAR), and the LAR must review and sign the informed consent form. If the patient does not have a LAR, the patient must appear able to provide informed consent and must review and sign the informed consent form. If the patient is deemed unable to provide informed consent and does not have a LAR, they cannot participate in the study. In addition, the patient's study partner/informant (as defined in the study inclusion criteria) must sign the informed consent form. If the LAR and the patient's study partner/informant are the same individual, he/she should sign under both.
14. No active suicidality identified on Columbia-Suicide Severity Rating Scale (C-SSRS).
15. Patients with stable prostate cancer may be included at the discretion of the Medical Monitor.
16. Patients who are on monoclonal antibody medication for the treatment of Alzheimer's (ex. lecanemab, donanemab) for > 6 months or have discontinued monoclonal antibody medication for the treatment of Alzheimer's for > 6 months may be included if all other criteria have been met.

Exclusion Criteria

Subjects will be excluded if they have any of the conditions listed below:

1. Taking one of the following medications: Medications for treatment of cancer or other drugs that weaken the immune system (ex. Natalizumab and Rituximab), Amiodarone, Bishydroxycoumarin, Chloramphenicol, Cimetidine, Fluconazole, Fluvastatin, Miconazole, Phenylbutazone, Sulphinpyrazone, Sulphadiazine, Sulphamethizole, Sulfamethoxazole, Sulphaphenazole, Trimethoprim, and Zafirlukast.
2. Current active infection in participants including, but not limited to, herpes zoster, herpes infection, bronchitis, sinusitis, upper respiratory infection and fungal skin infection. Siponimod may increase the risk in participants with active infections.
3. If participant received mRNA COVID-19 vaccination, must have received last dose at least 3 months prior to first dose of study drug/placebo.
4. Current evidence or history within the last 3 years of a neurological or psychiatric illness that could contribute to

	dementia, including (but not limited to) epilepsy, focal brain lesion, Parkinson's disease, seizure disorder, or head injury with loss of consciousness. 5. Meets DSM IV criteria for any major psychiatric disorder including psychosis, major depression and bipolar disorder. 6. Known history of or self-reported active alcohol and/ or substance abuse within the past three years. 7. Isolated living circumstances which would prohibit a study partner from providing sufficient and credible information about the participant. 8. Poorly controlled hypertension 9. Known Atrioventricular heart block, known heart block type I-III. 10. History of myocardial infarction or signs or symptoms of unstable coronary artery disease within the last year (including revascularization procedure/angioplasty). 11. Severe pulmonary disease (including chronic obstructive pulmonary disease) requiring more than 2 hospitalizations within the past year. 12. Untreated obstructive sleep apnea. 13. Any thyroid disease (unless euthyroid on treatment for at least 6 months prior to screening). 14. Active neoplastic disease (except for skin tumors other than melanoma) within five years. 15. Absolute lymphocytopenia of <1,000/mm3, or a history of lymphocytopenia within the past two years. 16. Absolute neutropenia of <1,000/mm3, or a history of neutropenia within the past two years. 17. History of/ or current thromboembolism (including deep venous thrombosis). 18. Any clinically significant hepatic or renal disease (including presence of Hepatitis B or C surface antigen or an elevated transaminase levels of greater than 2x the upper limit of normal (ULN) or creatinine greater than 1.5 x upper limit of normal (ULN)). 19. Clinically significant hematologic or coagulation disorder including any unexplained anemia, or a platelet count less than 100,000/µL at screening. 20. Use of any investigational drug within 30 days or within five half-lives of the investigational agent, whichever is longer. 21. Unwilling or unable to undergo CT or MRI imaging. 22. In the opinion of the investigator, participation would not be in the best interest of the subject. 23. Subjects with CYP2C9*3/*3 genotyping *Waiver/exemption requests for Inclusion/Exclusion Criteria may be submitted to the medical monitor for review and approval on an individual basis.
Phase:	IIb

Description of Sites/Facilities Enrolling Participants:	This is a single site study located at Barrow Neurological Institute in Phoenix, Arizona.
Description of Study Intervention:	Siponimod 1 mg/day vs. placebo taken daily orally (ratio 2:1 drug: placebo). The drug escalation scale will be used starting from 0.25mg/daily ending with final dosage of 1mg/day.
Study Duration:	The trial will last 4-5 years.
Participant Duration:	Participants completing the study will be involved for up to 20 months in duration, from screening to end of study. Treatment period will be 52 weeks and 6-month washout/observation

1.1 STUDY PROCEDURES

Visit 1, Day -56 to 0 Screening Visit (Up to 8 weeks)
During the screening visit, the subject (or their legally authorized representative as appropriate) and study partner / informant (if not the same as their LAR) will sign the informed consent document prior to undergoing any study procedures.
The following tests and evaluations are used to determine subject eligibility:

- Informed consent
- Inclusion and exclusion criteria
- *CYP2C9* genotyping (buccal swab)
- Medical History & Verify Diagnosis
- Concomitant medication reconciliation
- Serum pregnancy test if applicable
- Neurological and physical examination
- Vital signs, height and weight
- Labs (CBC, CMP, Hep B, Hep C, HIV, TSH)
- Coagulation panel (PT, PTT, INR) (optional, only for patients who opt into LP/CSF substudy)
- Cognitive assessments (MMSE, Hachinski, GDS)
- Columbia Suicide Severity Rating Scale (C-SSRS)
- MRI (MRI or CT within the past 12 months of Screening Visit Day 1)
- EKG (electrocardiogram)

Patients who do not meet the eligibility criteria will not be randomized and will be considered screen failures. Screen failures may be re-screened once per PI discretion for eligibility..

> *If no MRI or CT scan has been completed within the past 12 months: Participants who consent will be scheduled for volumetric MRI (vMRI) to be completed prior to the Baseline Visit.*

In addition, participants will be asked to complete an ophthalmic evaluation to obtain a baseline evaluation prior to the start of treatment.

Optional Lumbar Punctures for CSF Sub-Study
For lumbar punctures, the principal investigator (or sub-investigator) will perform the procedure. A separate section will be added to the consent form, which the subject (or their legally authorized representative) will sign to authorize the lumbar puncture and collection of cerebrospinal fluid (CSF).
To ensure subject safety, PT, INR, and PTT levels will be checked at baseline prior to any lumbar puncture procedure and subsequently prior to each follow-up visit where a lumbar puncture is being performed.
A follow-up telephone check will be conducted the day after each lumbar puncture within 24-48 hours. If there is persistent spinal headache, a blood patch may be administered at the discretion of the investigator.

Optional Saliva Sub-Study
Participants will have the option to provide a saliva sample via passive drooling at Baseline, Week 24, Week 52, Week 56 and Week 78. We will measure Aβ40 and Aβ42 in all saliva samples, as done previously[37].

Visit 2 at Day 1 of dosing, Baseline Visit, 0.25mg/day

Upon confirmation that all eligibility criteria have been met, baseline evaluations will be performed with subjects. These evaluations will include an assessment of concomitant therapy/medications, measurement of vital signs and body weight, collection of blood samples, vMRI (if not performed at screening) and cognitive assessments.

The following procedures and tests will be performed at baseline:
Do not dose patient on Baseline visit if the patient opted for LP. Refer to Visit 2.5 instead

- Inclusion/exclusion check
- Concomitant medication reconciliation
- Labs (CBC w/ diff, CMP, TSH)
- Labs (PT, PTT, INR) to be completed and reviewed pre-lumbar puncture for all lumbar punctures
- Optical Coherence Tomography (OCT) Eye Exam (must be done before dose)
- Serum pregnancy test (if applicable)
- EKG (EKG to be completed at pre- and 6-hours post-dose ± 10 minutes)
- Vital signs including weight
- Vital signs, post-dose, every hour for 6 hours ±10 minutes
- Neuro and physical exam
- Cognitive assessments for data collection only (MMSE, ADAS-cog 13, CDR, ADCS-ADL)
- Volumetric MRI (+ 4 weeks window) if vMRI was not completed during Screening
- Blood biomarkers
- Randomization*
- Dispense study drug, patient dose to be completed in-clinic*
- Drug diary provided to patient*
- Drug accountability including instructions on how to take treatment drug*
- Adverse event Assessment
  The following are to be completed if participants opted into one, or both, of the optional studies:
    - *Lumbar puncture followed by phone check in 24hrs-48 hrs post-lumbar puncture*
    - *Saliva collection pre- treatment dose*

Visit 2.5 at Day 1 of dosing, Baseline Visit, 0.25mg/day (Only for patients who undergo LP at Baseline)

- Vistal signs including weight (pre-dose), hourly vital signs ±10 minutes (post-dose observation for 6 hours
- EKG (pre-dose and post-dose at 6 hours ±10 minutes
- Randomization
- Dispense study drug, patient dose to be completed in-clinic
- Drug diary provided to patient
- Drug accountability including instructions on how to take treatment drug
- Adverse event assessment

Every effort will be made to complete baseline visits in the morning to allow for ample time to monitor participants after the first visit. Subjects will be administered first treatment dose in clinic and will be monitored for the following 6 hours. Every hour subjects will undergo BP/HR monitoring and AE symptom check. Investigator will be notified of BP/HR each hour with HRs rate less than 55 BPM being indicative of immediate intervention by investigator.
At the 6-hour mark after first dose participant will undergo an EKG for safety monitoring. Investigators will review EKG results for bradyarrhythmia and prolonged QT interval. Review of EKG will be completed prior to participant leaving clinic for participant safety.

Prior to leaving, patients will be given a sufficient supply of study medication (or placebo) to last until the next in-clinic visit at Visit 3 along with instructions for their use. Patients should be instructed to take their medication every morning thereafter at or around the same time.

If a titration dose is missed, or if 4 or more consecutive daily doses are missed during treatment, restart at last dose taken.

Telephone visits at Day 7,21,35,49,63,84,140,196,252, and 308 (±7 days)

Telephone visits will be used between dose increases to monitor any safety events with participants. In addition, telephone visits will serve as safety check-in.

The following procedures will be performed at all scheduled telephone visits as shown in the schedule of events:

- Medication reconciliation
- AE assessment

Visit 3 at Day 14 (±7 days) 0.50mg/day

The following tests and procedures will be completed in clinic:

- Concomitant medication reconciliation
- Labs (CBC w/ diff, CMP, TSH)
- EKG (pre-dose, post-dose at 6-hours ±10 minutes)
- Vital signs including weight (pre-dose)
- Vital signs (post-dose every hour ±10 minutes for 6 hours)
- Neuro and physical exam
- Blood biomarkers
- Dispense study drug and drug diary
- Drug accountability
- Adverse event assessment

Visit 4 at Day 28 (± 7 days) 0.75mg/day

The following tests and procedures will be completed in clinic:

- Concomitant medication reconciliation
- Labs (CBC w/ diff, CMP, TSH)
- EKG (pre-dose, post-dose at 6-hours ±10 minutes)
- Vital signs including weight (pre-dose)
- Vital signs (post-dose every hour ±10 minutes for 6 hours)
- Neuro and physical exam
- Blood biomarkers
- Dispense study drug and drug diary in clinic
- Drug accountability
- Adverse event assessment

Visit 5 at Day 42 (± 7 days) 1 mg/day

The following tests and procedures will be completed in clinic:

- Concomitant medication reconciliation
- Labs (CBC w/ diff, CMP, TSH)
- EKG (pre-dose, post-dose at 6-hours ±10 minutes)
- Vital signs including weight (pre-dose)

- Vital signs (post-dose every hour ±10 minutes for 6 hours)
- Neuro and physical exam
- Blood biomarkers
- Dispense study drug and drug diary in clinic
- Drug accountability
- Adverse event assessment

Visit 6 at Day 56 (± 7 days) 1 mg/day
The following tests and procedures will be completed in clinic:

- Concomitant medication reconciliation
- Labs (CBC w/ diff, CMP, TSH)
- EKG
- Vital signs including weight
- Neuro and physical exam
- Dispense study drug and drug diary in clinic
- Drug accountability
- Adverse event assessment
- Blood biomarkers
- OCT Eye Exam

Visit 7 at Day 70 (± 7 days) 1mg/day
The following tests and procedures will be completed in clinic:

- Concomitant medication reconciliation
- Labs (CBC w/ diff, CMP, TSH)
- EKG
- Vital signs including weight
- Neuro and physical exam
- Blood biomarkers
- Dispense study drug and drug diary
- Drug accountability
- Adverse event assessment

Visit 8 at Day 112 (± 7 days) 1mg/day
The following tests and procedures will be completed in clinic:

- Concomitant medication reconciliation
- Labs (CBC w/ diff, CMP, TSH)
- EKG
- Vital signs including weight
- Neuro and physical exam
- Blood biomarkers
- Dispense study drug and drug diary
- Drug accountability
- Adverse event assessment

Visit 9 at Day 168 (± 7 days) 1mg/day
The following tests and procedures will be completed in clinic:

- Concomitant medication reconciliation
- Labs (CBC w/ diff, CMP, TSH)
- EKG
- OCT Eye exam
- Vital signs including weight

- Neuro and physical exam
- Blood biomarkers
- Cognitive assessments (MMSE, ADAS-Cog 13, CDR-SOB, ADCS-ADL)
- Dispense study drug and drug diary
- Drug accountability
- Volumetric MRI (+4 week window)
- Adverse event assessment

The following are to be completed if participants opted into one, or both, of the optional studies:

- *Saliva collection*

Visit 10 at Day 224 (± 7 days) 1mg/day

The following tests and procedures will be completed in clinic:

- Concomitant medication reconciliation
- Labs (CBC w/ diff, CMP, TSH)
- EKG
- Vital signs including weight
- Neuro and physical exam
- Blood biomarkers
- Dispense study drug and drug diary
- Drug accountability
- Adverse event assessment

Visit 11 at Day 280 (± 7 days) 1mg/day

The following tests and procedures will be completed in clinic:

- Concomitant medication reconciliation
- Labs (CBC w/ diff, CMP, TSH)
- EKG
- Vital signs including weight
- Neuro and physical exam
- Blood biomarkers
- Cognitive Assessments (MMSE, ADAS-Cog 13, CDR-SOB)
- Dispense study drug
- Drug accountability
- Adverse event assessment

The following are to be completed if participants opted into one, or both, of the optional studies:
Saliva collection

Visit 12 at Day 336 (± 7 days)

The following tests and procedures will be completed in clinic:

- Concomitant medication reconciliation
- Labs (CBC w/ diff, CMP, TSH)
- EKG
- Vital signs including weight
- Neuro and physical exam
- Blood biomarkers
- Dispense study drug and drug diary
- Drug accountability
- Adverse event assessment

Visit 13 at Day 364 End of Study Drug Treatment (± 7 days)
The following tests and procedures will be completed in clinic:

- Concomitant medication reconciliation
- Drug accountability and drug diary
- Coagulation panel labs (PT, PTT, INR) to be completed pre-optional lumbar puncture (LP).
- Labs (CBC w/ diff, CMP, TSH)
- EKG
- Vital signs including weight
- Neuro and physical exam
- Blood biomarkers
- OCT Eye Exam
- Volumetric MRI (+4 weeks)
- Cognitive Assessments (MMSE, ADAS-Cog13, CDR-SOB, ADCS-ADL)
- Drug accountability and collect drug diary
- Adverse event assessment

The following are to be completed if participants opted into one, or both, of the optional studies:

- *Lumbar puncture (to measure CSF biomarkers) followed by phone check in 24hrs post-lumbar puncture*
- *Saliva collection*

Visit 14 at Day 392 (± 7 days)
The following tests and procedures will be completed in clinic:

- Concomitant medication reconciliation
- Labs (CBC w/ diff, CMP, TSH)
- EKG
- Vital signs including weight
- Neuro and physical exam
- Blood biomarkers
- Adverse event assessment

The following are to be completed if participants opted for the Saliva sub-study:

- *Saliva collection*

End of Study at Day 546 (± 7 days)
The following tests and procedures will be completed in clinic:

- Concomitant medication reconciliation
- Labs (CBC w/ diff, CMP, TSH)
- EKG
- Vital signs including weight
- Neuro and physical exam
- Volumetric MRI (±4 weeks, patients can come before or after visit for end of study)
- OCT Eye exam
- Blood biomarkers
- Cognitive assessments (MMSE, ADAS-Cog 13, CDR-SOB, ADCS-ADL)
- Adverse event assessment

The following are to be completed if participants opted Saliva sub-study:

- *Saliva collection*

Unscheduled Visits
Unscheduled visits will be at the investigator's discretion. All unscheduled visits will include the following tests and procedures:

- EKG
- Vital Signs and Weight
- Neurological and Physical Exam
- Labs (CBC with differential, CMP, TSH)
- Blood Biomarkers
- Medication reconciliation
- Adverse Event Assessment
- OCT Eye exam (in the case of blurred vision or other ocular issues)

1.2 SCHEDULE OF EVENTS (SOE)

The Schedule of Events is summarized in tabular form above and detailed below in table 1. All subjects will be expected to complete all evaluations in study.

Table 1: Study Schedule of Events

		0.25 mg/day		0.50 mg/day		0.75 mg/day		1 mg/day						
Study Visit	**Screening Visit**	**Baseline Visit**	**Phone Visit**	**Visit 3**	**Phone Visit**	**Visit 4**	**Phone Visit**	**Visit 5**	**Phone Visit**	**Visit 6**	**Phone Visit**	**Visit 7**	**Phone Visit**	**Visit 8**
Study Day (+/- 7 days)	**-56 to 0**	**1**	**7**	**14**	**21**	**28**	**35**	**42**	**49**	**56**	**63**	**70**	**84**	**112**
Study Week		0	1	2	3	4	5	6	7	8	9	10	12	16
Informed Consent	X													
Eligibility Criteria	X													
CYP2C9 genotyping	X													
Medical History	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Concomitant Medications	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Labs (CBC, CMP, TSH)	X	X		X		X		X		X		X		X
Coagulation panel (PT, PTT, INR)	X[e]	X[e]												
Serum Pregnancy Test	X[i]	X[i]												
EKG	X	X[h]		X[h]		X[h]		X[h]		X		X		X
Vital signs, Height/Weight	X[k]	X[mk]		X[mk]		X[mk]		X[mk]		X[k]		X[k]		X[k]
Neuro and Physical Exam	X	X		X		X		X		X		X		X
Cognitive Assessments	X[a]	X[b,c]												
Eye exam (OCT)		X[n]								X				
MRI (preferred) or CT scan[g]	X[g]													
Randomization		X												
Volumetric MRI		X[o]												
Blood biomarkers		X		X		X		X		X		X		X
(Optional) Saliva		X[r]												
(Optional) Lumbar Puncture[d]		X[d,e,r,s]												
Drug Accountability		X		X		X		X		X		X		X
Adverse Event Assessment		X	X	X	X	X	X	X	X	X	X	X	X	X
Dispense Study Drug and Drug Diary		X[s]		X		X		X		X		X	X	X

a. Screening MMSE, Hachinski, Geriatric Depression Scale (GDS), and CSSR-S
b. ADCOMS (MMSE, ADAS-Cog 13, CDR-SOB)
c. ADCS-ADL
d. All lumbar punctures will be followed by phone call 24-48hrs after lumbar puncture to ensure participant safety
e. Drawn only if opted in for LP. Results required before LP is completed
schedule
f. Dispense of visit drug and first treatment to be completed in clinic
g. If not done within 12 months. Study willing to accept historical CT if done in past 12 months
h. EKG 6 hour post-dose observation. ±10 minutes
i. If participant is of child-bearing potential
k. Height will only be recorded at screening
m. Post-dose vitals every hour ±10 minutes
n. Eye exam (OCT)must be pre-dose
o. vMRI can be done within window of 4 weeks
p. End of study vMRI can be done within window of ±4 weeks
r. Optional substudy procedure if patient opts in
s. If patients opt in for LP on Baseline visit do not dose until 24 to 48 hrs after LP. Follow Visit 2.5
t. Collect drug diary only on this day

	1 mg/day										
Study Visit	**Phone Visit**	**Visit 9**	**Phone Visit**	**Visit 10**	**Phone Visit**	**Visit 11**	**Phone Visit**	**Visit 12**	**End of Study Drug – Visit 13**	**Visit 14**	**End of Study Visit**
Study Day	**140**	**168**	**196**	**224**	**252**	**280**	**308**	**336**	**364**	**392**	**546**
Study Week	20	24	28	32	36	40	44	48	52	56	78
Medical History	X	X	X	X	X	X	X	X	X	X	X
Concomitant Medications	X	X	X	X	X	X	X	X	X	X	X
Labs (CBC, CMP, TSH)		X		X		X		X	X	X	X
Coagulation panel (PT, PTT, INR)									X[e]		
EKG		X		X		X		X	X	X	X
Vital Signs, Height/Weight		X[k]		X[k]		X[k]		X[k]	X[k]	X[k]	X[k]
Neuro and Physical Exam		X		X		X		X	X	X	X
Cognitive Assessments		X[b,c]				X[b,c]			X[b,c]		X[b,c]
Eye exam (OCT)		X							X		X
MRI (preferred) or CT scan											
Randomization											
Volumetric MRI		X[o]							X[o]		X[o]
Blood Biomarkers		X		X		X		X	X	X	X
(Optional) Saliva		X[r]						X[r]	X[r]	X[r]	X[r]
(Optional) Lumbar Puncture[d]									X[d,e,r]		
Drug Accountability		X		X		X		X	X		
Adverse Event Assessment	X	X	X	X	X	X	X	X	X	X	X
Dispense Study Drug and Drug Diary		X		X		X		X	X[t]		

a. Screening MMSE, Hachinski, Geriatric Depression Scale (GDS), and CSSR-S
b. ADCOMS (MMSE, ADAS-Cog 13, CDR-SOB)
c. ADCS-ADL
d. All lumbar punctures will be followed by phone call 24-48hrs after lumbar puncture to ensure participant safety
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i. If participant is of child-bearing potential
k. Height will only be recorded at screening
m. Post-dose vitals every hour ±10 minutes
n. Eye exam (OCT) must be pre-dose
o. vMRI can be done within window of 4 weeks
p. End of study vMRI can be done within window of ±4 weeks
r. Optional substudy procedure if patient opts in
s. If patients opt in for LP on Baseline visit do not dose until 24 to 48 hrs after LP. Follow Visit 2.5
t. Collect drug diary only on this day

2. INTRODUCTION

2.1 STUDY RATIONALE

Alzheimer's disease (AD) is the major form of dementia worldwide. AD is a neurodegenerative disorder (NDD) with clinical trajectory unique to each patient and displaying several complex neuropathologies suspected to develop sequentially but which overlap at the time dementia is diagnosed[22]. These pathologies include extracellular amyloid beta deposits forming senile plaques, intracellular neurofibrillary tangles composed of hyperphosphorylated tau protein, synaptic degeneration, neuronal death, and central inflammation, which altogether produce an unalterable cognitive decline over time[23]. Most therapeutic interventions targeting Alzheimer's disease until now have focused on monotherapies to alter a single neuropathology, including BACE1 inhibitors, gamma-secretase inhibitors and modulators, and active and passive immunization against amyloid beta. Unfortunately, very few of the therapeutic approaches tested to date have been able to significantly alter disease trajectory[44]. This emphasizes the urgent need for novel therapeutic interventions to reduce several AD neuropathologies simultaneously. In fact, the complexity of AD likely requires combination therapies or pleiotropic agents[44]. While the role of inflammation in AD remains enigmatic, it develops concomitantly with other neuropathologies[13.] Although previous attempts to lower neuroinflammation in humans have been inconclusive regarding disease trajectory and slowing of cognitive decline (including the now FDA-approved aducanumab and lecanemab), strategies targeting neuroinflammation in animal models are capable of mitigating AD-like pathologies.[4,30]

Inflammation is pervasive to many neurological disorders, yet no clinical trial has demonstrated the efficacy of anti-inflammatory agents for AD. Interestingly, chronic peripheral low-grade inflammation is associated with aging and increases the risk for disease and mortality, including AD. Accumulating evidence indicates that nuclear factor-kappa B, tumor necrosis factor alpha (TNFα), interleukins (e.g. IL-1beta, IL-2, and IL-6), and chemokines (e.g. IL-8) are found elevated both in the blood and central nervous system (CNS) of AD patients. This suggests that proper and efficient modulation of neuroinflammation could be a viable intervention to alter disease trajectory in AD. In the present application, we intend to repurpose the FDA-approved drug Siponimod that is used to treat multiple sclerosis (MS) in early AD patients.

Throughout the protocol:

- The term 'study intervention' and 'study drug' refers to Siponimod and placebo in the double-blind treatment period.
- The term 'sponsor' refers to the entities listed in the Contact Information page(s)
- The term 'participant' refers to the common term 'subject'

2.2 BACKGROUND

Recently, the modulation of sphingolipids has been proposed as a possible avenue to treat AD, since they regulate synaptic plasticity, amyloidogenesis, and tau physiology[6]. A very potent signaling molecule involved in numerous cellular processes is sphingosine-1-phosphate (S1P). S1P is an extracellular phosphorylated form of the membrane lipid sphingosine present in the circulation and CSF that is bound to albumin and lipoproteins, such as ApoE[6]. S1P regulates cellular pathways by binding to five G-protein-coupled receptors, termed S1PR1-5, which are present in high levels in cardiac, vascular, immune, and brain cells[5]. Interestingly, S1PR are inhibited by organic compounds known as trifluoromethylbenzenes, which are derived from myriocin, a metabolite of the fungus Isaria sinclairii. Binding of the agonists/modulators to S1PR induces the internalization and intracellular sequestration of the receptors, thus inhibits the corresponding signaling pathways[5]. The pleiotropic actions of S1PR modulators have been a strong motivation for Novartis Pharmaceuticals to investigate them in the past 30 years to treat multiple sclerosis (MS). After successful testing in animal models and in clinical trials, the first S1PR modulator, fingolimod, obtained FDA approval for MS in 2010[5]. More recently, Novartis has developed a less cardio-toxic analog with higher specificity for SP1R1 and 5 named Siponimod, which was FDA-approved, also for MS, in 2019.

S1PR modulators are brain penetrant[4], immunomodulatory, and neuroprotective in MS[5].

Immuno-modulation is mostly achieved by preventing egression of lymphocytes from lymphoid organs, and the modulation of cytokine expression, including microglia[5]. For T lymphocytes, it was shown that S1PR modulators prevent the infiltration of CD8+ memory cells in the central nervous system (CNS). This property could be key for AD since, very recently, the group of Wyss-Coray showed that CD8+ T cells likely play a critical role in the adaptive immune response in the CNS of AD patients[17,42]. On the other hand, the molecular mechanisms for neuroprotection are currently poorly understood. Nonetheless, both fingolimod and Siponimod lower the rate of brain atrophy in remitting relapsing MS (RRMS)[7]. Very interestingly, Siponimod was found to reduce brain atrophy at later stages of MS called secondary progressive MS (SPMS), making it the first drug with a demonstrated neuroprotective effect in SPMS[12.] More precisely, at month 12 the percentage of total brain volume change from baseline adjusted mean were −0.28% and −0.46% ($p<0.0001$) in Siponimod vs placebo-treated SPMS subjects, respectively[25].

Beyond MS, fingolimod was tested successfully in a 28-day clinical trial involving 30 amyotrophic lateral sclerosis (ALS) subjects taking the same dose as MS patients (0.5 mg/day)[3]\ , which constitutes precedence to repurpose S1PR modulators across NDD that have a strong inflammation component. The major adverse event (AE) findings in fingolimod vs. placebo-treated subjects were: 1 - A greater heart rate slowing (three participants had transient bradycardia but without clinical symptoms) in the hours after first dose, but not beyond 24h; 2 - Lower levels of circulating CD4+ and CD8+ T-lymphocytes, and CD19+ B-cells; 3 - A moderate increase in alkaline phosphatase levels in the first 4 weeks of treatment. Besides these AEs, which are similar to those reported in MS patients, there were no clinically significant abnormalities on routine blood counts, chemistries, urinalysis, and liver function tests[3]. Of note, two participants who were randomized despite being on QT-prolonging medications (sertraline and amitriptyline) completed the study without any severe AE[3]. Moreover, and very pertinent for AD, in 1 month-old 5XFAD mice (i.e. before plaque deposition onset, which starts at 2 months of age in this model) treated with 1 and 5 mg/kg/day of fingolimod for a period of two months were found to have significantly lowered Aβ42 loads when measured by ELISA as well as lower levels of neuroinflammation when assessed by densitometric analysis of GFAP-positive astrocytes and Iba1 immuno-positive microglia in the hippocampus[2]. Thus, the pleiotropic activities of S1PR modulators in AD mice and MS patients, combined with good safety and tolerability in other NDD, are hugely attractive in consideration of repurposing these agents for AD.

An important consideration when repurposing drugs is the degree of risk vs. potential benefit to patients. A major AE for fingolimod is the induction of transient bradycardia at dosing onset[29]. This risk can be mitigated via a dose-escalation regimen[24] and by selecting subjects not prone to bradycardia. Because MS patients are younger than AD patients, we believe that cardiac risks are too high to test fingolimod in AD subjects. However, **Siponimod generates much fewer and less severe cardiac AEs** than fingolimod[19], likely due to higher target selectivity for S1PR1 and 5[5]. It has been administered at **high doses** (10 mg/day for 18 days) in an up-titration regimen **without significant heart AEs[38]**. This motivated us to repurpose Siponimod rather than fingolimod for AD. That said, Siponimod is ~80% metabolized by the hepatic cytochrome P450 enzyme, CYP2C9 and thus acts as a competitive inhibitor for these enzymes. Critically, the gene for CYP2C9 has several alleles. While individuals with *CYP2C9*1* are relatively safe, those with *CYP2C9*2* and **3* alleles experience higher inhibition of the enzyme by Siponimod (see section D.1.a. for more details)[15]. Altogether, AEs induced by S1PR modulators led MS experts and the FDA to recommend the following regimens <u>for MS patients</u> (drug label): 1 – Implementation of a daily escalation dose regimen for this class of drugs to manage the risk of heart block[27]; and 2 – Set the maximum drug dose at 2 mg/day in *CYP2C9*1* subjects, but reduce the maximum dose to 1 mg/day for *CYP2C9*2* and **3* individuals[15]. In summary, the pleiotropic properties and manageable AEs of **Siponimod** make it a **very appealing candidate to repurpose** with the goal of altering both neuroinflammation and neurodegeneration, and thus the disease trajectory **of AD** if administered at an optimum regimen.

Our multidisciplinary group has pioneered the testing of immunomodulators for AD. First, we have evaluated the anti-cancer, a potent TNF-α modulator, thalidomide. Unfortunately, this drug proved too toxic for mild to moderate AD subjects, with many requesting to withdraw from the study early[8]. Currently, we are investigating lenalidomide in amnestic mild cognitive impairment subjects (NCT #04032626). For the present project, we have brainstormed with our multiple sclerosis (MS) specialty colleagues because MS is an NDD with a strong inflammatory component and with a pharmacopeia capable of reducing disease

severity[39]. This inter-disciplinary teamwork resulted in a great interest to explore sphingosine-1-phosphate receptor (S1PR) modulators, particularly Siponimod, because they show both neuroprotective and immunomodulatory properties (see section C.). This collaboration led us to formulate the **central hypothesis that Siponimod could lower the rate of brain atrophy in AD subjects**.

To test our central hypothesis, we will carry out an 18-month Phase II, double-blind, randomized, two-armed, placebo controlled, proof-of-concept clinical study in early AD subjects (i.e. mild AD) who will be receiving an escalating dose of Siponimod or placebo in the ratio 2:1 for 12 months, followed by a 6-month washout period. The primary outcome measures are safety and tolerability of Siponimod in mild AD subjects. The secondary outcome measures are the rates of brain atrophy derived from volumetric MRI (vMRI) as a proxy for neurodegeneration conducted at baseline, 6, 12, and 18 months. The tertiary outcome measures are the changes in cognition and the levels of AD-associated (e.g., Aβ and tau) and inflammatory biomarkers in CSF after Siponimod exposure. In an exploratory effort, we will also measure plasma inflammatory markers during the entire duration of the study to investigate whether one or more of these markers can be used as dynamic surrogate markers of treatment response. Using our unique experience with the repurposing of immunomodulatory drugs for AD (and NCT #04032626), in the present project we are using elements of clinical trial design that we believe were successful and made some adjustments to fit the pharmacologic and toxic properties of Siponimod.[8]

3. RISK/BENEFIT ASSESSMENT

3.1.1 KNOWN POTENTIAL RISKS

Siponimod (formerly known as BAF312 and completed trial NCT #01665144) has been FDA approved since 2019 (IND #076122) for the treatment of multiple sclerosis. Most adverse events have been reported from MS patients, and very little is known about possible adverse events in subjects without MS. Most risks are associated with doses >2 mg/day. Most common adverse reactions (reported in >5%) are headache, hypertension, and transaminase increase (see Table 2).

Table 2*: *Summary of Adverse Events Reported in ≥5% of Mayzent treated patients at a rate of at least 1% higher than in patients receiving placebo

Adverse reaction	MAYZENT 2 mg (N=1099) %	Placebo (N=546) %
Headache[a]	15	14
Hypertension[b]	13	9
Transaminase increased[c]	11	3
Falls	11	10
Peripheral edema[d]	8	4
Nausea	7	4
Dizziness	7	5
Diarrhea	6	4
Bradycardia[e]	6	3
Pain in extremity[f]	6	4

Terms were combined as follows:

a. Headache, tension headache, sinus headache, cervicogenic headache, drug withdrawal headache, and procedural headache.
b. Hypertension, blood pressure increased, blood pressure systolic increased, essential hypertension, blood pressure diastolic increased.
c. Alanine aminotransferase increased, gamma-glutamyl transferase, increased hepatic enzyme increased, aspartate aminotransferase increase, blood alkaline phosphatase increase, liver function test increased, hepatic function abnormal, liver function test abnormal, transaminases increased.
d. Peripheral edema, joint swelling, fluid retention, swelling face
e. Bradycardia, sinus bradycardia, heart rate decreased
f. Pain in extremity and limb discomfort

POTENTIAL SERIOUS ADVERSE EVENTS

+ Bradycardia, Bradyarrhythmia, and Atrioventricular Conduction Delays

Siponimod may cause the heart rate to slow, especially after taking the first dose. This issue will be mitigated by administering an EKG post dose at the 6-hour mark to assess for any potential adverse events. In addition, BP/HR will be checked hourly for 6 hours to assess for potential bradyarrhythmia or adverse events.

+ Infections

Siponimod lowers the number of lymphocytes, which may increase the risk of serious infections. Lymphocyte levels will be monitored by measuring a Complete Blood Count with differential at each in-clinic study visit.

+ Liver Injury

Elevations of transaminases may occur in Siponimod-treated patients. Caution should be exercised when using Siponimod in patients with a history of significant liver disease. We will mitigate potential for significant liver injury by routinely measuring levels via Comprehensive Metabolic Panel (CMP) which will be drawn at every in-clinic study visit.

+ Increased Blood Pressure

Siponimod-treated patients may experience an increase in systolic and diastolic pressure within 1 month of treatment initiation and persisting with continued treatment. During initial clinical trials of Siponimod it was noted that subjects experienced an increase in systolic pressure averaging 3 mmHg and diastolic pressure averaging 1.2 mmHg. Blood pressure will be monitored after each initial dose in titration schedule and at each subsequent visit. Blood pressure exceeding 160/95 after repeated testing will require investigator approval to continue in study.

+ Macular Edema

Siponimod may cause macular edema, which usually starts within the first 1 to 4 months after treatment onset. Macular edema may appear as early as 3 months following initiation of treatment with Siponimod, so subjects will be required to have baseline eye exam (within 12 months pre-dose initiation) and eye exam at scheduled intervals post-dose initiation per the study schedule of events. In clinical trials it was noted that macular edema occurred within the first four months of therapy.

3.1.2 KNOWN POTENTIAL BENEFITS

Potential benefits for subjects suffering mild AD dementia are unknown. However, Siponimod can reduce inflammation, mainly by sequestering CD8+ T memory cells in peripheral lymphoid organs, which limits their infiltration in brain tissues. Siponimod also modulates the expression of cytokines in several cell types, including in microglia[5]. Very recently, the group of Wyss-Coray showed that CD8+ T cells likely play a critical role in the adaptive immune response in the CNS of AD patients[17,42]. In addition, although the molecular mechanisms are unclear, Siponimod was able to lower the rate of brain atrophy in remitting relapsing MS (RRMS)[7]. Very interestingly, Siponimod was found to reduce brain atrophy at later stages of MS called secondary progressive MS (SPMS), making it the first drug with a demonstrated neuroprotective effect in SPMS.[12] More precisely, at month 12 the percentage of total brain volume change from baseline adjusted mean were −0.28% and −0.46% ($p<0.0001$) in Siponimod vs placebo-treated SPMS subjects, respectively[25].

In summary, the potential benefits of Siponimod in mild AD dementia would be to reduce both neurodegeneration and inflammation, which are two major contributing mechanisms involved in the cognitive decline processes associated with dementia.

3.1.3 ASSESSMENT OF POTENTIAL RISKS AND BENEFITS

Given Siponimod has been FDA approved since 2019 (and its predecessor, fingolimod, was FDA approved in 2010), a significant amount of information is available from MS studies outlining the benefits and adverse events for this drug.
In this current clinical trial, our design incorporates specific strategies to enhance benefits while mitigating risks. We are selectively enrolling patients with mild AD dementia. Our approach involves initiating treatment at a conservative dose of 0.25 mg, gradually increasing to a final dose of 1.0 mg based on a predetermined titration schedule. Regular visits will include comprehensive safety assessments, encompassing procedures such as blood draws and EKGs, to diligently monitor for potential adverse events through the trial period.

+ Fetal Risk
Based on animal studies, Siponimod may cause fetal harm. For this reason, we will confirm that all female subjects are not pregnant at screening and are not of child-bearing potential (i.e. surgically sterile or LMP >12 months ago).

DRUG INTERACTIONS
In clinical trials it was noted that the following drug interactions can occur in participants. Due to this it was determined that the possible risks do not outweigh the benefits of potential treatment, and as such, the following drug classes will be deemed exclusionary.

+ Anti-Neoplastic, Immune-Modulating, or Immunosuppressive Therapies
- Siponimod has not been studied in combination with anti-neoplastic, immune-modulating, or immunosuppressive therapies. Caution should be used during concomitant administration because of the risk of additive immune effects during such therapy and in the weeks following administration.
- When switching from drugs with prolonged immune effects, the half-life and mode of action of these drugs must be considered to avoid unintended additive immunosuppressive effects.
- Because of the characteristics and duration of alemtuzumab immune suppressive effects, initiating treatment with Siponimod after alemtuzumab is not recommended.

+Anti-Arrhythmic Drugs, QT Prolonging Drugs, Drugs That May Decrease Heart Rate
- Siponimod has not been studied in patients taking QT prolonging drugs.
- Class Ia (e.g., quinidine, procainamide) and Class III (e.g., amiodarone, sotalol) anti-arrhythmic drugs have been associated with cases of Torsades de Pointes in patients with bradycardia.
- Because of the potential additive effects on heart rate, treatment with Siponimod should generally not be initiated in patients who are concurrently treated with QT prolonging drugs with known arrhythmogenic properties, heart rate lowering calcium channel blockers (e.g., verapamil, diltiazem), or other drugs that may decrease heart rate (e.g., ivabradine, digoxin).

+ Beta-Blockers
- Caution should be applied when Siponimod is initiated in patients receiving treatment with a beta-blocker because of the additive effects on lowering heart rate.

+ Vaccination
- During and for up to one month after discontinuation of treatment with Siponimod, vaccinations may be less effective.

- The use of live attenuated vaccines may carry the risk of infection and should therefore be avoided during Siponimod treatment and for up to 4 weeks after discontinuation of treatment with Siponimod.

+ CYP2C9 and CYP3A4 Inhibitors

- Because of a significant increase in exposure to Siponimod, concomitant use of Siponimod and drugs that cause moderate CYP2C9 and moderate or strong CYP3A4 inhibition is not recommended. This concomitant drug regimen can consist of a moderate CYP2C9/CYP3A4 dual inhibitor (e.g., fluconazole) or a moderate CYP2C9 inhibitor in combination with a separate - moderate or strong CYP3A4 inhibitor.
- Concomitant use of Siponimod and moderate (e.g., modafinil, efavirenz) or strong CYP3A4 inducers is not recommended for patients with CYP2C9*1/*3 and*2/*3 genotype.

4. OBJECTIVES AND ENDPOINTS

OBJECTIVES	ENDPOINTS
PRIMARY	
The primary objective of the study is to assess the safety and tolerability of Siponimod in mild AD dementia subjects.	Safety will be assessed at each study visit through completion of comprehensive tests and procedures aimed at monitoring for potentials adverse events (AEs). Study staff will also gather direct reports from subjects regarding any adverse events.
SECONDARY	
The secondary objective of the study is to assess the effects of Siponimod on AD-associated neurodegeneration by deriving annual brain atrophy rates from volumetric MRI (vMRI).	Siponimod was observed to slow rates of brain atrophy in both RRMS and SPMS patients. Thus, the aim is to assess the medical benefits of the drug in mild AD dementia subjects by assessing whether brain atrophy slows with Siponimod treatment. Subjects will undergo volumetric MRI (vMRI) measurements of the hippocampus, cortical thickness, and global thickness as indicators of neurodegeneration.
TERTIARY	
The tertiary objective of study is to assess the effects of Siponimod on cognition following administration of study drug vs. placebo.	Cognition will be assessed by comparing the changes in two assessments: ADCS-ADL and ADCOMS from baseline to various visit timepoints, including Week 78 (End of Study).
EXPLORATORY	
Exploratory objectives will include: i) measurements of markers found in the CSF fluid that are known to be associated with mild AD dementia and inflammation before and after treatment ii) assessment of blood inflammatory markers before and after treatment iii) to assess levels of Aβ found in saliva samples collected before and after treatment with Siponimod vs. placebo	i) CSF biomarkers that will be assessed include amyloid beta 40 & 42, Tau, and Phospo-Tau. ii) Blood inflammatory markers include CRP, TNF-α, IL-1β, IL-6, IL-8, and IL- 10 (54-plex measured on the Meso Scale Discovery [MSD] ELISA platform). Plus, we will count the number of white blood cells (B and T lymphocytes, macrophages, etc) in the blood to estimate the impact of Siponimod on peripheral markers of the immune system. iii) Saliva markers that will be assessed are Aβ40 and Aβ2. Saliva Aβ levels is a recent biomarker that has been identified to study AD. Thus, we propose to explore the effects of Siponimod on Aβ40 and Aβ2 throughout the course of the study.

5. STUDY DESIGN

OVERALL DESIGN

In the current project we aim to test the **central hypothesis that Siponimod could lower the rate of neurodegeneration as measured by brain atrophy in mild AD dementia subjects**. To test our central hypothesis, we will carry out an 18-month Phase II, double-blind, randomized, two-armed, placebo controlled, proof-of-concept clinical study in 105 mild AD dementia subjects who will receive an escalating dose of Siponimod (0.25-1 mg/day) or placebo at a randomization ratio of 2:1 for 12 months, followed by a 6-month washout period. For patients who meet inclusion/exclusion criteria, 35 will be randomly assigned to the placebo group, and 70 will be randomly assigned to the drug arm. A single site will manage the entire study, i.e. the Barrow Neurological Institute (BNI) in Phoenix, AZ. No patient stratification and no interim analysis are planned. The decision to pursue dosing will depend on safety data which is included in the inclusion/exclusion criteria.

6. SCIENTIFIC RATIONALE FOR STUDY DESIGN

This is a randomized, double-blind, placebo-controlled, parallel group study. The use of placebo is appropriate to minimize bias related to treatment expectations of the subject, study partner, and site investigator, as well as bias related to changes in the relationship between the subject and study partner that might occur with the initiation of treatment and expectation of improvement in motor symptoms or cognition. Changes in subject /study partner interactions can impact subject mood and might introduce biases that cannot be quantified. The double-blind use of placebo will also prevent bias in the clinical and scientific assessments.

Most monotherapy clinical trials with a focus on disease modification completed to date have failed to meet the clinical endpoint of significantly slowing cognitive decline in dementia due to Alzheimer's disease (AD), including BACE1 inhibitors[40], γ-secretase inhibitors and modulators[18], and active and passive immunization against monomeric, oligomeric, and protofibril Aβ[26]. Recently the mABs, aducanumab and lecanemab, were approved on the biomarker endpoints of removing amyloid. This emphasizes the urgent need for novel therapeutic approaches able to reduce several AD neuropathologies simultaneously.

Inflammation is prominent in many neurological disorders, yet no clinical trial has demonstrated the efficacy of anti-inflammatory agents for AD. Interestingly, chronic peripheral low-grade inflammation is associated with aging and increases the risk for disease and mortality, including the development of AD[20]. Accumulating evidence indicates that nuclear factor-κB (NF-κB), tumor necrosis factor alpha (TNF-α), interleukins (e.g. IL-1β, IL-2, and IL-6), and chemokines (e.g. IL-8) were found elevated both in the blood and central nervous system (CNS) of AD patients[20]. These data confirm that inflammation plays a critical role in the cause and effect of AD neuropathology[42]. Interestingly, Siponimod is capable of modulating inflammation by preventing egression of lymphocytes from lymphoid organs, and the modulation of cytokine expression, including in microglia[5].

The use of the NIA-AA criteria[32] is well-established and accepted for the diagnosis of mild AD. Nonetheless, we will use eventual updates released by the NIA for the diagnosis of AD throughout the duration of our clinical study.

Safety will be assessed by standard measures throughout the study.

The endpoints are appropriate measures of treatment effect in this study. Because no cognitive data has been reported in MS subjects, the main proxy for Siponimod efficacy is a slowing of neurodegeneration assessed by calculating annual brain atrophy rates. However, it is widely accepted in the neuroscience community that altering neurodegeneration will likely slow cognitive decline. To date, the main measure for an effective treatment of AD is cognitive performance. Indeed, while they may be useful to help with the diagnosis of AD, there is no biomarker that has demonstrated theragnostic value yet. Therefore, any inflexion of cognitive decline, as measured by the several tests we have selected, will be the only clinical validation of disease improvement. Thus, we have included the ADCOMS scale to assess cognition during our study.

Blood, saliva and CSF biomarkers were chosen for this study because they are expected to provide insights into inflammations status before, during and after dosing. As previously noted, the biomarkers being evaluated have been linked as being indicators for the progression of mild AD dementia.

JUSTIFICATION FOR DOSE

Dosing of Siponimod in AD is limited by two parameters. First, although no bradycardia was observed in a chronic high dose regimen in healthy subjects[38] and at therapeutic doses in MS patients undergoing a daily dose-titration regimen[27], these data were collected on young to middle-aged adults (age range 18-60). However, there is no literature on the possible cardiac AEs induced by Siponimod in older adults (age >60), which favors using a low dose in such individuals. Second, the *CYP2C9*2* and *CYP2C9*3* alleles are known to metabolize Siponimod more slowly than the *CYP2C9*1* genotype (half-lives of ~51 [double] and 126 hours [quintuple], respectively). *CYP2C9*2* allele is observed in 4-18% of Caucasians and Hispanics, which constitute a large proportion of the AD patients seen at BNI, and *CYP2C9*3* allele has a frequency of 2-20% in the same ethnicities, while Asians, African decedents, and Native Americans have lower frequencies for both genotypes[33,43]. These observations of moderate frequencies for *CYP2C9*2* and *CYP2C9*3* alleles, and the recommendations from the FDA-approved package insert encourage us to administer the suggested low dose of 1 mg/day in *CYP2C9*1/x* and *CYP2C9*2/x* subjects, and to exclude *CYP2C9*3/3* subjects (0.4-0.5% of the population) for the present study. Consequently, and after discussing these issues with our MS specialty colleagues, for safety reasons we have selected to administer a maximum dose of 1 mg/day to the mild AD subjects that we will enroll in this proof-of-concept study. Further, instead of the daily titration regimen recommended in MS, we will apply a biweekly dose increment of 0.25 mg/day over the course of 6 weeks, with the first dose given at each increased strength (dose increases by0.25 mg at a time) taken during in-person visits at BNI to monitor for any eventual AEs that could emerge with dose increases.

END OF STUDY DEFINITION

A participant is considered to have completed the study if he or she has completed all phases of the study including the last visit or the last scheduled procedure shown in the Schedule of Events (SOE).

7. STUDY POPULATION

INCLUSION CRITERIA

See table on page 5 for *inclusion*/exclusion criteria.

EXCLUSION CRITERIA

See table on page 6 for inclusion/*exclusion* criteria.

LIFESTYLE CONSIDERATIONS

Because of its ability to inhibit CYP enzymes, study subjects will be recommended to not eat grapefruit or drink grapefruit juice.

SCREEN FAILURES

Screen failures are defined as participants who consent to participate in the clinical trial but are not subsequently randomly assigned to the study intervention or entered in the study. The collection of a minimal set of screen failure information is required to ensure transparent reporting of participants who are deemed as a screen failure, to meet the Consolidated Standards of Reporting Trials

(CONSORT) publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demographics and eligibility criteria.

Because of inherent daily variations in cognitive performance observed in the clinic, if MMSE score is outside of the range 20-26 at the initial screening, the PI may request MMSE retesting after a two week period.

Patients who do not meet one or more of the entry criteria will not be randomized and will be considered screen failures. Screen failures may be re-screened once per PI discretion for eligibility.

STRATEGIES FOR RECRUITMENT AND RETENTION

The clinical trial aims to recruit a total of 105 subjects from the study site, Barrow Neurological Institute (BNI). Study goal is to achieve a balanced enrollment, targeting approximately 50% male and 50% female participants. These study subjects will be outpatients diagnosed with mild AD dementia at BNI.

In Arizona, there are an estimated 150,000 cases of AD[1]. Annually, BNI sees approximately 2,000 patients with memory disorders. Most patients are Caucasians, but our clinics have representative samples of URGs including LatinX and African Americans, though we consistently work to reach other members of Underrepresented Groups as well. Recently, for the New IDEAS (Imaging Dementia-Evidence for Amyloid Scanning) study, we enrolled 27 participants, all of whom belong to URGs (underrepresented racial and ethnic groups). This patient population has eagerly engaged in clinical trials, and BNI has approximately 30 clinical trials on-going at any given time. The majority of the current clinical trial participants are referred by BNI physicians, with a small percentage referred by community physicians and agencies. The BNI research team has consistently met or exceeded their patient enrollment targets over the past ten years with over 600 clinical trial participants in the current database. Patients followed at BNI are continuously offered opportunities to be screened for clinical trials.

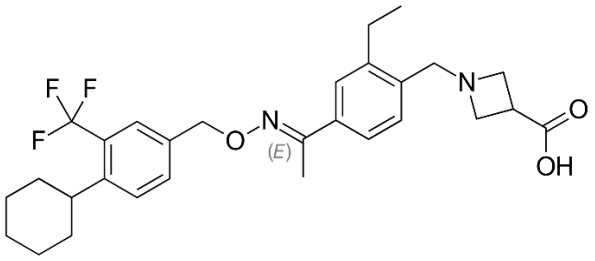
To maximize retention, we will use several methods to contact participants (phone, email, family contact) and give visit reminders. Our study design was also adapted to improve retention by offering flexibility for the scheduled in person visits (±7 days), and by including longer in person visit intervals in the final 6 months of dosing. We have consulted MS specialists to identify the minimum dose possible to retain efficacy while minimizing adverse events, which we hope will prevent early withdrawals.

8. STUDY INTERVENTION

STUDY INTERVENTION(S) ADMINISTRATION

STUDY INTERVENTION DESCRIPTION

Siponimod is marketed by Novartis Pharmaceuticals Corporation under the name “Mayzent” registered under FDA IND #076122 (initially named “BAF-312”). The empirical formula is C29H35F3N2O3, and the gram molecular weight is 516.6. Its chemical structure is shown below.



The formulation used in the present study will be provided by Novartis and is the same as indicated in IND #076122:
“Siponimod is provided as 0.25 mg film-coated tablets for oral use. Each film coated tablet contains 0.25 mg Siponimod, equivalent to 0.2 mg as Siponimod fumaric acid co-crystal. Siponimod film-coated tablets contain the following inactive ingredients: microcrystalline cellulose, crospovidone, glyceryl behenate, lactose monohydrate, colloidal silicon dioxide with a film coating containing lecithin (soy), iron oxides (red and black iron oxides for the 0.25mg), polyvinyl alcohol, talc, titanium dioxide, xanthan gum. “

Similarly, the placebo formulation will be provided by Novartis as explained in IND #076122:
“The placebo tablets used in the studies described in this NDA have the same composition and configuration as the 0.25 mg drug products but do not contain Siponimod.”

DOSING AND ADMINISTRATION

The primary objectives of our project are the examination of drug safety and tolerability. This is an 18-month Phase II, double-blind, randomized, two-armed, placebo controlled, proof-of-concept clinical study. In total, we will enroll 105 early symptomatic AD subjects (i.e. mild AD) who will receive an escalating dose of Siponimod (0.25-1 mg/day) or placebo at a randomization ratio of 2:1 for 12 months (52 weeks), followed by a 6-month washout period (month 18). Study medications will be given orally, preferably in the morning. The patients and study staff will be blind to the randomization schema. For patients who meet all inclusion and exclusion criteria for enrollment, 35 will be randomly assigned to the placebo group, and 70 will be randomly assigned to the drug arm. There is no visual, taste, or smell difference between the placebo and Siponimod formulations. The randomization schedule will be developed by the study biostatistician.

For safety reasons, a gradual dose escalation regimen will be implemented for mild AD dementia subjects receiving Siponimod/Placebo. The medication will be administered in 0.25 mg increments biweekly reaching the maximum dose of 1 mg. As a precaution, the initial higher doses will be given in a clinical setting, with subjects observed for six hours by study staff who are blinded to the study drug. This allows for prompt intervention in case of any distress, such as issues with heart or blood pressure. During these in-person visits, blood samples will be collected to monitor for signs of toxicity changes. Tests will include a complete blood count with differential, comprehensive metabolic panels, and liver function.

All test results will be reviewed by the principal investigator (or sub-investigators) to determine whether toxicity is developing in study subjects. Moreover, we will carry out cognitive tests every 3 months and volumetric MRI every 6 months (Table 1) to detect eventual brain toxicities. Finally, in case of AEs outside of the clinic, Study subjects will be provided contact information for both clinic and study coordinator.

The investigator or his designee is responsible for explaining the correct use of the investigational agents to the patient/caregiver/study partner, verifying that instructions are followed properly per drug manufacturer. Study staff will maintain accurate records of investigational product dispensing and collection and returning all unused medication to the Study Site at each in-person visit. Patients will be instructed to contact the investigator or the study coordinator as soon as possible if he or she has a complaint or problem with the investigational product so that the situation can be assessed.

If toxicities are identified, we will apply a special toxicity protocol. Per clinical trials, hepatotoxicity was primarily noted in controlled trials of Siponimod in patients. Notably, AST and ALT values were elevated above 3 times the upper limit of normal. If hepatotoxicity is noted, the subject will be withdrawn from the study and not replaced if this occurs after Visit 7.
Please refer to the section, Procedure Used in Case of Toxicity.

PREPARATION/HANDLING/STORAGE/ACCOUNTABILITY

ACQUISITION AND ACCOUNTABILITY

For this study, we will acquire a clinical trial form of 0.25 mg Siponimod from Novartis, the same formulation as used in clinical trials for multiple sclerosis (IND #076122). During the time between reception from the drug manufacturer to the delivery to study subjects, the study drug and placebo will be managed solely by licensed pharmacists from the investigator's institution (i.e., BNI). The shelf life of Siponimod being 18 months per manufacturer's recommendation, several batches of investigational products (Siponimod and matching placebo) are planned to cover the need of study drug for the entire duration of the study.

To maintain the blind of the blinded staff including the blinded monitor, there will be a separate unblinded pharmacy monitor to review the unblinded pharmacy records. Jeffrey Burmeister, PharmD, from the Barrow Neurological Institute will serve as the unblinded pharmacy monitor for this study.

The unblinded pharmacy monitor will have access to the unblinded pharmacy records and will perform accountability of the investigational products at each onsite monitoring session. Verification of the investigational products includes documenting any discrepancies between the total amounts of study drug shipped to the study site with the total amount of study drug dispensed to subjects along with unused study drug. The monitor will verify that correct study drug was administered by comparing study record against pharmacy documentation.

The unblinded pharmacy monitor will perform the following checks and verifications:

- Confirm study drug was administered only to qualified subjects who provided written Informed Consent.
- Confirm all study drug used for each subject and verify dispense dates.
- Check for site pharmacy personnel changes. If changes occur, ensure new personnel will have access to receive the study drug during the trial.
- Review temperature logs. Ensure the correct temperature is maintained.
- Ensure the receipt of drug shipments has been acknowledged by the Pharmacy on Record, and packing lists are filed.
- Verify that the investigational products are stored in a securely locked area.

At the end of the study, all unused study drugs will be reviewed by the unblinded pharmacy monitor and disposed of as directed by standard local procedures. A final drug inventory log will be retained at the study site.

FORMULATION, APPEARANCE, PACKAGING, AND LABELING

Formulation

Siponimod is an analogue of fingolimod. It is an immunomodulatory agent that binds with high affinity to the sphingosine-1-phosphate (S1P) receptors S1P1 and S1P5 subtypes of the five known S1P receptors. The chemical name is 1-[[4-[(E)-N-[[4-cyclohexyl-3-(trifluoromethyl)phenyl]methoxy]-C-methylcarbonimidoyl]-2-ethylphenyl]methyl]azetidine-3-carboxylic acid. The empirical formula for Siponimod is C29H35F3N2O3, and the gram molecular weight is 516.6.

Active ingredient: Each 0.25 mg tablet contains 0.2 mg of Siponimod.

Excipients: The excipients are microcrystalline cellulose, crospovidone, glyceryl behenate, lactose monohydrate, colloidal silicon dioxide with a film coating containing lecithin (soy), iron

oxides (red and black iron oxides for the 0.25mg strength), polyvinyl alcohol, talc, titanium dioxide, xanthan gum.

Appearance

Siponimod is white to almost white powder. It is soluble in warm water up to 100 mg/mL.

Packaging

Siponimod and matching placebo tablets will be manufactured and packaged in USP compliant child-resistant bottles by Novartis with 14 tablets per bottle. Bottles will be directly shipped by Novartis to the BNI pharmacy at regular intervals.

Placebo

Matching placebo tablets used in this study will have the same composition as the 0.25 mg drug products but do not contain Siponimod. The matching placebo will be of identical appearance to the study medication to maintain the double-blind design of the study.

Labeling

Study drug will be labeled in accordance with GCP and local regulations. Each bottle containing study drug or placebo will be labeled with the following information:

- Protocol number
- [Subject name], [Subject number], [DOB]
- [Date of dispensation]
- Siponimod 0.25 mg or Placebo Tablets
- Instructions on taking medications as directed by physicianQuantity
- Lot number/kit number
- Expiration date
- Store at refrigerated at 4°C (with excursions permitted to 2-10°C)
- Name, address, and phone number of the principal-investigator
- “Investigational Drug Use Only”

PRODUCT STORAGE AND STABILITY

Manufacturer shelf-life is 18 months. We will be acquiring the study drug from Novartis in small batches over a period of 5 years. Study drug will be stored in a refrigerator (4°C) with excursions permitted to 2-10°C. The tablets should not be exposed to high temperature and humidity. Tablets should be protected from light. Temperature will be monitored and recorded to ensure the stability of study drug. Expiry dates will be monitored, and any expired products will be pulled from the active stock.

PREPARATION

Siponimod and matching placebo tablets of the same size and color to the active drug will be compounded by Novartis in accordance with GCP (IND #076122). The matching placebo will be of identical appearance to the study medication to maintain the double-blind design of the study.

MEASURES TO MINIMIZE BIAS: RANDOMIZATION AND BLINDING

This is a randomized, double-blind, placebo-controlled, parallel group study. The use of placebo is appropriate to minimize bias related to treatment expectations of the subject, study partner, and site investigator, as well as to changes in the relationship between the subject and study partners that might occur with the initiation of treatment and expectation of improvement in motoric symptoms or cognition. Changes in subject/study partner interactions can impact subject mood and might introduce biases that

cannot be quantified. The double-blind use of placebo will also prevent bias in the clinical and scientific assessments.

Randomization will be carried out by our study biostatistician and the medication or placebo prepared accordingly by our clinical pharmacist. This procedure was developed to ensure double blinding, i.e. neither the investigators, nor the study subjects will know the medication taken by study subjects until the completion of the study and the acquisition of all data.

Emergency unblinding for AEs will be performed by the unblinded pharmacy team if the subject's well-being requires knowledge of the subject's treatment assignment. The subject's treatment status will remain blinded to other study personnel and Principal Investigator.

The investigator should make every effort to contact the medical monitor prior to unblinding a subject's treatment assignment. If a subject's treatment assignment is unblinded, the medical monitor and DSMB must be notified immediately. All unblinding events will be recorded in the CRF.

At the baseline visit (Visit 2), subjects who have met all the eligibility criteria for enrollment will be randomly assigned to one of two treatment groups:

1. Placebo (n=35) or
2. Siponimod 1 mg/day (n=70)

Randomization will be 2:1 and designed by our biostatistician. Randomization will take place during the Baseline visit.

In case of toxicity finding, the blinding will remain in effect as neither the Investigator, nor site staff will be informed whether the participants receive Siponimod or placebo. However, the study pharmacist will be informed in case there is need to adjust dosing if participants are treated with investigational product.

An independent statistician supporting the DSMB will be unblinded and may unblind the DSMB if required for proper study monitoring. All staff and research personnel involved in this study will remain blinded until all subjects have completed the trial and scientific data acquired.

STUDY INTERVENTION COMPLIANCE

Patient compliance with study medication will be assessed at each visit. Compliance will be assessed by direct questioning and counting returned tablets. Deviation(s) from the prescribed dosage regimen should be recorded in the CRF.

Patients who are significantly noncompliant will be instructed to discontinue medication from the study. A patient will be considered significantly noncompliant if he or she misses more than 21 consecutive days of study medication during the study without extenuating circumstance, for example: development of an SAE. Patients who stop dosing due to development of an SAE will have their case reviewed by investigator, site, medical monitor, DSMB, and Principal Investigator for prescribing appropriate actions. Similarly, a patient will be considered significantly noncompliant if he or she is judged by the investigator to have intentionally or repeatedly taken more than the prescribed amount of medication. Drug accountability will be emphasized at the start-up meeting, and drug accountability will be monitored throughout the study.

Temporary discontinuation of treatment with study drug due to ARIA-E should not be considered as non-compliance.

CONCOMITANT THERAPY

All concomitant medications taken during the study must be recorded on the concomitant medications case report form (CRF). Patients and their study partners/ caregivers will be instructed to consult the investigator or other appropriate study personnel at the site before initiation of any new medications or supplements and before changing dose of any current concomitant medications or supplements.

Medications that may affect cognitive function should be discussed with the medical monitor to determine if the patient should remain in the study.

- Allowed Concomitant Therapy
Patients on stable standard of care (SoC) AD treatment will be allowed in the study. The patient can be on stable doses of a cholinesterase inhibitor and/or memantine if it is stable for at least 90 days prior to the screening visit and is expected to remain on a stable dose for the remainder of the study period; or have demonstrated intolerance to or lack of efficacy from these medications.

- Prohibited Concomitant Therapy
Any therapy that does not fit with the SoC will be prohibited, including, but not limited to, cancer chemotherapy, narcotics, antimanic, and any anticonvulsants/antiseizures taken specifically for seizure disorders or epilepsy, and drugs listed in the exclusion criteria.

9. STUDY INTERVENTION DISCONTINUATION AND PARTICIPANT DISCONTINUATION/WITHDRAWAL

DISCONTINUATION OF STUDY INTERVENTION

A subject may discontinue study drug under any of the following circumstances:

- The subject wishes to discontinue study drug treatment for any reason.
- The subject experiences AEs that necessitate discontinuing study drug treatment. The Principal Investigator, medical monitor, and DSMB should be notified immediately.
- The investigator or DSMB decides to discontinue treatment for medical reasons. The reason for termination of study drug before study completion should be recorded in the CRF. At the time of study drug discontinuation, the End of Study Drug Visit should be administered. If the subject is not willing or able to return for all remaining study visits, they should be asked to return for the Week 52 and Week 78 visits, as well as returning all unused study drug. If this is not possible, they should be contacted by phone at Week 52 for the End of Study assessments (adverse events, and concomitant medications). The reason for withdrawal should be recorded in the case report form (CRF). The subject should complete the procedures for the Early Termination Visit, provided written consent to do so has not been withdrawn.

Subjects who discontinue or are withdrawn from the study following randomization:

- if they stop treatment (drug or placebo) before Visit 7, they will be replaced by new subjects
- if they stop treatment (drug or placebo) after Visit 7, they will not be replaced.

LOST TO FOLLOW-UP

To be considered lost to follow up, the study coordinator will make three documented attempts to contact the subject, who must also miss two scheduled visits. If these efforts are unsuccessful, a certified letter will be sent to the subject. If no response is received after this communication, then, the subject will be officially considered lost to follow up.

10. STUDY ASSESSMENTS AND PROCEDURES

EFFICACY ASSESSMENTS

The main drug efficacy measure for this study is neurodegeneration (secondary objective). We will derive annual brain atrophy rates by comparing volumetric MRI (vMRI) values for the hippocampal, cortical, and global thickness. All 105 study participants will undergo vMRI at Baseline (Week 0) and after 6 months 12 months , and 18 months.

To demonstrate clinical utility, it is necessary to show that a possible treatment for AD can slow down or reverse cognitive decline. Thus, our tertiary objective (and secondary efficacy endpoint) will assess cognition in all study subjects. For this, we will employ the recently created, high-sensitivity

Alzheimer's Disease Composite Score (ADCOMS), which is recommended for mild AD in clinical trials of 12 months' duration or longer[41]. Briefly, ADCOMS consists of 4 Alzheimer's Disease Assessment Scale-cognitive subscale items (ADAS-Cog13), 2 Mini-Mental State Examination items (MMSE), and all 6 Clinical Dementia Rating-Sum of Boxes items (CDR-SOB)[41]. ADCOMS will be administered at specified time points throughout the study (refer to study table 1). Furthermore, to assess conversion to AD or reversion to normality, we will repeat the cognitive measurements at 18 months (Week 78). This time was selected because the calculated mean time for conversion from aMCI to AD in the literature ranges from 12-20 months[16]. Thus, measuring cognitive performance 18 months after the start of the trial should provide us with a good assessment of whether Siponimod is effective at slowing down cognitive decline. A positive effect of Siponimod on cognitive outcome measures would translate into increased ADCOMS scores at the end of dosing vs. Baseline and/or a slower decline than in placebo-treated subjects at month 18.

Additional objectives (exploratory efficacy endpoints) are (i) an additional assessment of cognitive performance via the ADCS-ADL test administered every 6 months; and (ii) the measurement of biomarkers in the CSF, blood, and saliva, to explore the possibility of using biofluid markers as proxy of drug efficacy. For example, since Siponimod is an immunomodulator that prevents the egression of T lymphocytes from peripheral lymphoid organs, we should observe a decrease in circulating T lymphocytes and inflammatory markers in the blood samples we collect during the entire course of this clinical trial.

All procedures are carried out by trained medical staff and scientists used to conduct clinical trials at the study site. Labs (CBC with differential and CMP) analyses are performed by a contracted medical laboratory, therefore follow GCP standards.

+ Neurodegeneration Measures

- Volumetric MRI

MR imaging will be conducted on a 3T MRI scanner located in Phoenix, AZ. Sequences will include a high resolution 3D T1-weighted magnetization prepared rapid acquisition gradient echo (MPRAGE), a T2-weighted image, a 3D FLAIR image, and a high-resolution T1 image of the hippocampus. Each individual scan will be less than 7 minutes in length and will match protocols developed by the Alzheimer's disease neuroimaging initiative (ADNI). Other imaging may include advanced diffusion and perfusion . Each of these scans will be less than 10 minutes in duration, and the overall scan time will be less than 1 hour.

40 min sequence (estimated 38 min 26 s)

Sequence	**Approx. run time in minutes**
T1-weighted MPRAGE	6:11
T2-weighted image	6:42
T2-weighted FLAIR	6:10
High Res Hippo	5:36
Diffusion MRI	8:05
Perfusion MRI	5:32

+ Cognitive Measures

- Alzheimer's Disease Assessment Scale - Cognitive

Alzheimer's Disease Assessment Scale - Cognitive (ADAS-cog)[36] is a psychometric instrument that evaluates memory, attention, reasoning, language, orientation, and praxis. A higher score indicates more impairment; the range is 0 to 89. We will use the ADAS-cog13 as it is the currently accepted scale used by clinicians to detect changes in mildly demented subjects.

- Clinical Dementia Rating-Sum of Boxes

The Clinical Dementia Rating - Sum of Boxes (CDR-SOB) test[34] is a commonly used instrument in MCI studies. The CDR-SOB describes five degrees of impairment in performance on each of 6 categories of cognitive functioning including memory, orientation, judgment and problem solving, community affairs, home and hobbies, and personal care. The ratings of degree of impairment obtained on each of the 6 categories of function are synthesized into one global rating of dementia (ranging from 0 to 3), with more refined measure of change available by use of the Sum of Boxes. The CDR-rater will be a trained clinician. Ideally the same CDR rater will be used throughout the course of the trial when possible.

- Mini Mental State Examination

The Mini Mental State Examination (MMSE) is a screening instrument frequently used in Alzheimer's disease drug studies.[11] It evaluates orientation, memory, attention, concentration, naming, repetition, comprehension, and the ability to create a sentence and copy two intersecting pentagons. Scores range from 0 to 30, with 30 being the score assigned to cognitively normal individuals. The range for mild AD is 21-26.

- Alzheimer's Disease Cooperative Study - Activities of Daily Living

In the Alzheimer's Disease Cooperative Study - Activities of Daily Living (ADCS-ADL) test, study partners are queried via a structured interview format as to whether subjects attempted each item in the inventory during the preceding four weeks, as well as their level of performance[14]. This will provide a reliable estimate of daily functioning. ADCS-ADL is not part of the ADCOMS computation. It will be administered every 6 months, as done routinely for our clinical trials.

- ***ADCOMS***

The AD Composite Score (ADCOMS) consists of 4 ADAS-Cog items, 2 MMSE items, and all 6 CDR-SOB items [PMID: 27010616]. The ADCOMS was derived with the intention of using it as a primary outcome measure for registration trials of disease-modifying treatments in persons with predementia and early AD-related dementia. The ADCOMS has demonstrated improved sensitivity over each individual scale comprised in the composite and requires smaller sample sizes to detect longitudinal cognitive changes. Retrospective analyses have shown that ADCOMS is superior to other measures in detecting and measuring early cognitive decline and disease progression among patients with early stages of AD, i.e. mild cognitive impairment and mild AD [PMID: 35099758]. Thus, the ADCOMS appears as the most promising cognitive composite score for the present study.

+ Biomarkers

- Exploratory Blood Biomarkers

Because CSF collection is not possible at each site visit, we explore the possibility of using blood inflammatory markers as surrogate markers of Siponimod efficacy. For this, we will collect whole blood (cubital vein); maximum 45 mL in tubes containing ethylenediaminetetraacetic acid (EDTA). After differential centrifugations, we will measure plasma C-reactive protein, TNF-α, IL-1β, IL-6, IL-8, and IL-10 by multiplex ELISA (Meso Scale Discovery 54 Plex for human inflammatory markers).

+ Exploratory Saliva Biomarkers (Optional Sub-Study)

In recent years, saliva amyloid beta levels have shown good performance at discriminating AD vs. cognitively normal subjects. For example, the lead PI has published a proof-of-concept study on a few subjects that showed increased levels of saliva Aβ42 in AD patients compared to age- and gender-matched cognitively normal controls[37]. Thus, in an exploratory effort we will offer to our study subjects the option to donate saliva at specified points in the study (see table 1) to determine whether Aβ40 and Aβ42 levels in saliva are affected by long-term treatment with Siponimod.

+ Exploratory CSF Biomarkers (Optional Sub-Study)

As there is currently no effective treatment for AD, it is unclear how the levels of CSF biomarkers associated with the disease (e.g., amyloid beta 40 and 42, total tau, phospho-tau; all measured on the

Meso Scale Discovery multiplex ELISA platform) may change after a positive outcome induced by a disease-modifying therapy. Therefore, in the present study we will analyze these biomarkers in the CSF. In addition, because Siponimod is an immunomodulator, we will assess CSF inflammatory markers as a possible proxy for drug efficacy. LPs will be carried out at Baseline (Week 0) and end of dosing (Week 52). For this, we will measure the levels of CSF C-reactive protein, TNF-α, IL-1β, IL-6, IL-8, and IL-10 by multiplex ELISA (Meso Scale Discovery 54 Plex for human inflammatory markers).

+ Appropriateness of Measurements
The efficacy measures assessed in this study are appropriate for the evaluation of AD therapies. All cognitive tests we use are validated for the assessment of AD status and progression. The imaging and plasma biomarkers we are investigating are the most likely to demonstrate an alteration of AD neuropathologies and inflammation.

SAFETY AND OTHER ASSESSMENTS

Safety will be evaluated by assessing AEs, clinical laboratory data, vital signs, and concomitant medications. The investigator is responsible for monitoring the safety of study subjects and providing appropriate medical care during the study. Any significant safety issues should be discussed with the Medical Monitor and DSMB in a timely manner.

- Protection Against Privacy Risk
Every effort will be made to ensure the privacy of participant PHI and confidentiality by utilizing secure EMR and databases. Paper copies are filed by number in accordance with professional standards of privileged information. All documentation and case reports forms are compliant with HIPAA regulations. Confidentiality is safeguarded by strict adherence to these standards.

- Safety Monitoring

An independent Data Safety Monitoring Board (DSMB) comprised of three members not associated with the study will be secured prior to starting the study. This group of experts advises NIH and the Sponsor/Investigator. During the trial, the DSMB will review cumulative study data to evaluate safety, study conduct, and scientific validity and integrity of the trial. …

A Medical Monitor has been secured to provide medical expertise to the study team including answering investigator questions about the protocol, patient eligibility and the medical management of subject AE/SAE assessment/treatment, and/ or medical emergencies.

There will also be a Study Monitor who is responsible for monitoring the conduct of the clinical trial on a regular basis as per GCP and ICH guidelines. He/she will inspect the case report forms at regular intervals throughout the study to verify adherence to the protocol; completeness and accuracy of the data; and adherence to local regulations on the conduct of clinical research. The investigators agree to cooperate with the study monitor to ensure that any problems detected during these monitoring visits are resolved.

The study monitor will inspect site facilities (e.g., pharmacy, drug storage areas, laboratories) and review study-related records to evaluate the trial conduct and compliance with the protocol, GCP, and applicable regulatory requirements. Results of locally administered tests such as EKG tracings and local clinical laboratory reports will be retained as source documents. At the monitoring visits, the progress of the study will be discussed with the Investigator and/or the Study Coordinator and the informed consent forms and CRFs will be checked for completeness and accuracy. Subjects' source documents should be available for review.

The investigators will immediately inform the Medical Monitor and the DSMB in case of moderate to severe adverse events, as well as when an individual subject is temporarily removed from dosing due to hematologic signs of toxicity (e.g. thrombocytopenia and/or neutropenia). All early

terminations will also be indicated to the DSMB in a timely manner. The IRB will be notified per Barrow Neurological Institute IRB Policy regarding Adverse Events Reporting.

- Complaint Handling
The investigator or his/her designee is responsible for handling the following aspects of the product complaint process in accordance with the instructions provided for this study:
- recording a complete description of the product complaint reported and any associated AEs using the study-specific complaint forms provided for this purpose.
- informing the medical monitor within 24-48 business hours of subject complaints.

- Safety Measures
+ Routine Clinical Laboratory Tests
Routine hematology, blood chemistry will be performed at screening and subsequent follow-up visits as described in the schedule of events.

+ Electrocardiograms
For each subject, 12-lead digital electrocardiograms (EKGs) will be collected according to the Schedule of Events. The EKG at screening will be a single EKG and will be interpreted by the principal investigator, or sub-investigator. Every effort will be made to obtain EKGs around the same time of day; however, if unable to schedule for the same time, EKGs will still be completed per protocol. Patients must be supine for approximately 5 to 10 minutes before EKG collection

. EKGs will be interpreted by the principal investigator, or sub-investigator, with every effort made to have EKG results reviewed as close to time of collection as possible. Ideally EKG result review will be completed while the subject is still at site, so that any clinically relevant findings are identified with subject present.

After enrollment, if a clinically significant increase in the QT/corrected QT (QTc>500) interval from baseline, or other clinically significant quantitative or qualitative change from baseline, is present, the investigator will assess the subject for symptoms (for example, palpitations, near syncope, syncope) and to determine if the subject can continue in the study. The investigator, or qualified designee, is responsible for determining if any change in patient management is needed and must document their review of the EKG printed at the time of evaluation along with the final overread EKG and any alert reports.

+ Physical Examination
A complete physical examination (with neurological exam) will be performed at the visits listed in the study protocol and schedule of events. An abbreviated physical examination, to include an ophthalmic examination and routine examination of the heart, lungs, abdomen, skin, and oral cavity, will also be performed. Sitting vital signs (blood pressure, heart rate, respiratory rate, and temperature), after each subject sits quietly for at least 5 minutes, and body weight will be assessed at the Screening Visit and at each subsequent visit.

+ Vital Signs
Body temperature (oral or tympanic) will be measured as specified in the Schedule of Activities and as clinically indicated.
Blood pressure and pulse rate (PR) will be measured as specified in the Schedule of Activities and as clinically indicated. Blood pressure and PR should be measured after at least 5 minutes of sitting.

+ Body Weight and Height
Body weight and height will be recorded as specified in the Schedule of Activities and as clinically indicated.

+ Neurological Examination

A directed neurological examination will be performed by the investigator at the time points specified in the protocol and schedule of events.
Any clinically significant change from baseline on follow-up physical/neurological examinations should be recorded as an AE and reported in the CRF. If additional ARIA-H, ARIA-E, are detected, an additional neurological exam may be performed by the investigator.

- Toxicity Assessment
During visits at the study site, we will collect blood samples and conduct blood counts and comprehensive metabolic panels to monitor hematologic and metabolic toxicities. All lab results will be reviewed by the primary investigator, or sub-investigators, to determine if any clinically significant findings are seen that could be attributed to Siponimod related toxicities.

- Procedure Used in Case of Toxicity
If toxicities are identified, we apply a special toxicity protocol. The hematologic or chemistry values indicative of severe AEs are those listed in exclusion criteria.
If toxicity, including cardiac SAEs, is suspected, or proven, to be linked to Siponimod, subjects will stop taking the study drug for 7 days, and undergo new blood tests after at least 7 days of washout. If test results continue showing toxicity, subjects will be withdrawn from the study and not replaced if this occurs after Visit 7.
If toxicity markers improve, subjects will resume dosing at the prior lower dose (i.e., toxic dose minus 0.25 mg) and remain on this regimen until the end of the study. In case of a second toxic event, dosing will again be stopped for 7 days, and blood tests conducted. If the test results continue showing toxicity, subjects will be withdrawn and not replaced if this occurs after Visit 7. If toxicity markers improve, subjects will resume dosing at the previous lower dose (i.e., toxic dose minus 0.25 mg) and remain at this lower dose until the end of the study.
In case of a third toxic event, subjects will be withdrawn and not replaced if this occurs after Visit 7.
When study medication is withdrawn, study subjects will be recorded as “End Of Study Drug Visit” and undergo the corresponding assessments (Week 52 in Table 1). Then, the subjects will be asked to complete the corresponding washout period schedule of assessments: (Week 56 in Table 1) and 26 week “End of Study” (Week 78 in Table 1).

- Tolerability Assessment
Tolerability of Siponimod in study subjects will be derived from the frequency of AEs and study withdrawal motivated by subjects’ discomfort during the dosing period.

- Standard MRI (screening 1)
Patients who do not have an MRI scan within the past 12 months of Screening Visit Day 1 will be scheduled for an vMRI or CT during the screening period with results obtained before randomization is decided. (MRI is preferred).

>> Routine MRI Scan (*default choice*)
MR imaging will be conducted on a 3T MRI scanner located in Phoenix, AZ. Sequences will include a high-resolution 3D T1-weighted magnetization prepared rapid acquisition gradient echo (MPRAGE), a T2-weighted image, a 3D FLAIR image, and a high-resolution T1 image of the hippocampus. Each individual scan will be less than 7 minutes in length and will match protocols developed by the Alzheimer’s disease neuroimaging initiative (ADNI). Other imaging may include advanced diffusion, perfusion, spectroscopy, and functional MRI. Each of these scans will be less than 10 minutes in duration, and the overall scan time will be less than 1 hour. With the acquired T1 and T2 images, hippocampal, ventricular and whole brain volumes will be determined by automated segmentation using Freesurfer 6.0. Cortical thicknesses and subcortical volumes are also determined from this automated segmentation. With the high-resolution hippocampus T1 image, we are able to obtain the volumes of hippocampal subfields by using ASHS atlas. Voxelwise measures of regional gray matter volumes (corrected for the total intracranial volume) are determined using voxel-based morphometry in SPM8

and the longitudinal whole brain atrophy are estimated using iterative principal component analysis. Furthermore, voxel-wise whole brain morphological changes are determined using deformation-based morphometry with high resolution T1 images as input.

50 min sequence (estimated 49 min 30 s)

Sequence	Approx. run time in minutes
T1-weighted MPRAGE	6:11
T2-weighted image	6:42
T2-weighted FLAIR	6:10
High Res Hippo	5:36
Diffusion MRI	8:05
Perfusion MRI	5:32
MR Spectroscopy	2:54
Functional MRI	8:20

-Optional CSF Collection Sub-Study
Lumbar Puncture (LP) is a standard neurodiagnostic procedure for collection of CSF but may be associated with pain during the performance of the procedure. This is usually temporary and confined to the lower back. A persistent low-pressure headache may develop after LP, due to leakage of CSF. Although the frequencies of post-LP headaches have been reported to be as high as 10% using standard 20 gauge spinal needles, rates of less than 2% have been reported in elderly subjects when atraumatic (Sprotte) needles are used[35]. If a post-LP headache persists, it may need additional treatment, e.g. with fluids and analgesics. Uncommonly, a blood patch (injection of some of the subject's blood to patch the CSF leak) may be needed. Potential but rare risks of lumbar puncture include infection, damage to nerves in the back, bleeding into the CSF space, and death. The risk of these is less than 1%.

- Optional Saliva Collection Sub-Study
Saliva samples will be collected via passive drooling into plastic tubes, thus involving no risk to study subjects.

11. ADVERSE EVENTS AND SERIOUS ADVERSE EVENTS

DEFINITION OF ADVERSE EVENTS (AE)

An adverse event (AE) is any untoward medical occurrence in a clinical study participant administered pharmaceutical (investigational or non-investigational) product. An AE does not necessarily have causal relations with the intervention. An AE can therefore be any unfavorable and unintended sign (including an abnormal finding), symptom, or disease temporally associated with the use of a medicinal (investigational or non-investigational) product. (Definition per International Council on Harmonisation [ICH]).

This includes any occurrence that is new in onset or aggravated in severity or frequency from the baseline condition, or abnormal results of diagnostic procedures, including laboratory test abnormalities. The collections of AEs will begin at the Baseline visit after the first administration of study medication/placebo with the exception of study procedure related AE's that may occur during the screening process.

An unexpected adverse event is any AE, the specificity or severity of which is not consistent with the Investigator's Brochure.

A life-threatening adverse event is any AE that, in the view of the Investigator, places the subject at immediate risk of death from the reaction as it occurred. Disability is defined as a substantial disruption of a person's ability to conduct normal life functions.

DEFINITION OF SERIOUS ADVERSE EVENTS (SAE)

A serious adverse event is any AE that is fatal or life threatening (see below), results in persistent or significant disability (see below) or incapacity, requires inpatient hospitalization or prolongation of an existing hospitalization or is a congenital anomaly/birth defect or results in death. Other important medical events that may not result in death, be life threatening or require hospitalization should also be considered a serious adverse event when, based upon appropriate medical judgment, they may jeopardize the subject and may require medical or surgical intervention to prevent one of the outcomes listed above. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse. Associated with the use of the drug means that there is a reasonable possibility that the experience may have been caused by the drug.

An event is classified as serious if it meets any of the following criteria:
- Results in death.
- Is life-threatening.
- Requires inpatient hospitalization or prolongs an existing hospitalization.
- Results in persistent or significant disability/incapacity.
- Other important medical events. Medical and scientific judgment should determine whether an AE should be classified as serious in other situations, such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the subject or require intervention to prevent one of the outcomes listed in the definition above.

CLASSIFICATION OF AN ADVERSE EVENT

SEVERITY OF EVENT

The following levels of severity will be used in the evaluation of adverse events in our clinical study:
- **Mild**: Awareness of symptoms that are easily tolerated, causing minimal discomfort and not interfering with everyday activities.
- **Moderate**: Sufficient discomfort is present to cause interference with normal activity.
- **Severe**: Extreme distress, causing significant impairment of functioning or incapacitation. Prevents norma everyday activities.

The investigator must use clinical judgement in assessing the severity of events not directly experienced by the participant (e.g. laboratory abnormalities).

RELATIONSHIP TO STUDY INTERVENTION

The following definitions of relationship to study drug will be used to characterize the suspected causality of each AE, based on the Investigator's consideration of all available information:
- **Related**: There is a reasonable causal relationship between study intervention administration and the AE.
-**Probably Related**: The relationship with the use of the study drug seems relevant and/or the evet cannot be reasonably explained by another cause, but additional information may be obtained.
- **Possibly Related**: The relationship with the use of the study drug is weak but cannot be ruled out completely. Alternative causes are also possible (e.g., underlying or concurrent illness/clinical condition or/and an effect of another drug). Cases where relatedness cannot be assessed, or no information has been obtained should also be classified as possible.
- **Not Related**: No evidence that AE/SAE is related to the administration of the study drug.

An Emergency Code Break procedure will be available to the investigator to allow for unblinding of a patient, i.e. informing the investigator to which treatment group the patient has been assigned. Such procedures should only be utilized in emergency situations when the identity of the subject must be known by the investigator in order to provide appropriate medical treatment, i.e. unblinding is not required and should not be performed if knowledge of the patient's treatment group would have no influence on how the patient is being treated in the emergency situation.

EXPECTEDNESS

The Principal Investigator will be responsible for determining whether an adverse event (AE) is expected or unexpected. An AE will be considered unexpected if the nature, severity, or frequency of the event is not consistent with the risk information previously described for the study intervention.

TIME PERIOD AND FREQUENCY FOR EVENT ASSESSMENT AND FOLLOW-UP

The occurrence of an adverse event (AE) or serious adverse event (SAE) may come to the attention of study personnel during study visits and interviews of a study participant presenting for medical care, or upon review by a study monitor.

All AEs including local and systemic reactions not meeting the criteria for SAEs will be captured on the appropriate case report form (CRF). Information to be collected includes event description, time of onset, clinician's assessment of severity, relationship to study product (assessed only by those with the training and authority to make a diagnosis), and time of resolution/stabilization of the event. All AEs occurring while on study must be documented appropriately regardless of relationship. All AEs will be followed to adequate resolution.

Any medical condition that is present at the time that the participant is screened will be considered as baseline and not reported as an AE. However, if the study participant's condition deteriorates at any time during the study, it will be recorded as an AE.

Changes in the severity of an AE will be documented to allow an assessment of the duration of the event at each level of severity to be performed. AEs characterized as intermittent require documentation of onset and duration of each episode.

Study Coordinator will record all reportable events with start dates occurring after the first administration of study medication/placebo with the exception of study procedure related AE's that may occur during the screening process within 48 hours starting from when site became aware of AE and/or SAE. At each study visit, the investigator will inquire about the occurrence of AE/SAEs since the last visit. Events will be followed for outcome information until resolution or stabilization.

ADVERSE EVENT REPORTING

The condition of each study subject will be monitored throughout the study. Signs and symptoms of possible AEs may be observed by the staff, elicited by asking the patient and/or study partner/informant an open or indirect question (e.g. How have you felt since your last clinic visit?) or volunteered by the subject and/or study partner. All adverse events, whether observed by the investigator or staff, elicited from the subject or study partner, or volunteered by the subject or study partner, will be recorded in the

case report form. Data will include start and end dates, investigator-specified severity and relationship to study drug, and action taken. Whether the event resulted in death, required (or prolonged) hospitalization with persistent or significant disability/incapacity, required intervention to prevent any of the above outcomes, and/or whether it was reported as serious to the medical monitor, DSMB, IRB, and Study Sponsor will also be recorded.

Knowledge of a serious adverse event occurring or worsening in a study subject at any time during the trial must be reported by telephone within 24 hours (if physically possible) to the investigator who will immediately inform the medical monitor, DSMB and IRB in accordance with the IRB's specific requirements. Reports relative to the subject's subsequent course must be submitted to the DSMB until the event has subsided or, in the case of permanent impairment, until the condition stabilizes. At any time following the study, the investigator should immediately notify medical monitor, DSMB, and IRB if he learns of the occurrence of any malignancy or pregnancy involving the participant of a clinical trial, of any serious adverse event that could possibly be related to study drug, or of any congenital anomaly in an offspring of a participant.

SERIOUS ADVERSE EVENT REPORTING

All SAEs (regardless of suspected causality) should be communicated within 24 hours of discovery of the event to the medical monitor and DSMB members, and to the local IRB within 10 working days.

Any SAEs occurring any time after study participation that are considered by the investigator to be possibly related to study drug must also be reported as outlined above.

The study site will be responsible for notifying the Food and Drug Administration (FDA) of any unexpected fatal or life-threatening suspected adverse reaction as soon as possible, but in no case later than 7 calendar days after the sponsor's initial receipt of the information. In addition, the site must notify FDA and all participating investigators in an Investigational New Drug (IND) safety report of potential serious risks, from clinical trials or any other source, as soon as possible, but in no case later than 15 calendar days after the sponsor determines that the information qualifies for reporting.

REPORTING EVENTS TO PARTICIPANTS

Participants will be informed to discontinue the study drug if blood or cardiac toxicity is discovered. The hematologic values indicative of severe AEs are those listed in exclusion criteria. Refer to section, Procedure Used in Case of Toxicity.

EVENTS OF SPECIAL INTEREST

Occurrences of acute overdose should be reported for tracking purposes. In this study, an overdose is defined as a dose greater than the specified dose and requires medical attention.

REPORTING OF PREGNANCY

Because Siponimod is a suspected teratogenic agent based on animal studies, females must be surgically sterile (bilateral tubal ligation, oophorectomy, or hysterectomy) or of non-childbearing potential.

Although it is currently unknown whether Siponimod present in semen can affect the growth of fetuses, male participants will be asked to use contraceptive methods preventing the exposure of fetuses to semen.

If, despite the use of contraceptives as described in this protocol, the partner of a subject becomes pregnant during the study, she will be followed through the end of her pregnancy. A Pregnancy Notification/Outcome Consent Form and Pregnancy Notification Form will be completed and faxed to the Drug Safety Department at Research Pharmaceutical Services Inc. (RPS). The outcome of the pregnancy and the medical condition of any resultant offspring will be reported on a Pregnancy Outcome Form.

UNANTICIPATED PROBLEMS

The Office for Human Research Protections (OHRP) considers unanticipated problems involving risks to participants or others to include, in general, any incident, experience, or outcome 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 protocol-related documents, such as the Institutional Review Board (IRB)-approved research protocol and informed consent document; and (b) the characteristics of the participant population being studied.
- Related or possibly related to participation in the research ("possibly related" means there is a reasonable possibility that the incident, experience, or outcome may have been caused by the procedures involved in the research); and
- Suggests that the research places participants or others at a greater risk of harm (including physical, psychological, economic, or social harm) than was previously known or recognized.

UNANTICIPATED PROBLEM REPORTING

The investigator will report unanticipated problems (UPs) to the reviewing Institutional Review Board (IRB), NIH, the NI and to the DSMB. The UP report will include the following information:

- Protocol identifying information: protocol title and number, PI's name, and the IRB project number.
- A detailed description of the event, incident, experience, or outcome.
- An explanation of the basis for determining that the event, incident, experience, or outcome represents an UP.
- A description of any changes to the protocol or other corrective actions that have been taken or are proposed in response to the UP.

To satisfy the requirement for prompt reporting, UPs will be reported using the following timeline:

- A Major Protocol Deviation – a deviation, based on the clinical judgment of the PI or their designee, determined to significantly impact the scientific soundness of the research plan or suggest the research placed participants or others at a greater risk of harm (physical, psychological, economic, or social harm) not previously recognized. A major deviation meets all criteria for an UP involving risk. Major Protocol Deviations /UPs will be reported to local IRB within 10 days of investigator becoming aware. These will be reported to the DSMB and to the study sponsor within 24h of the investigator becoming aware of the event.
- Any other UP will be reported to the DSMB and to the study sponsor within 5 days of the investigator becoming aware of the problem. These UPs will be reported to the site's Institute QA Monitor within 7 days, and to the local IRB at continuing renewal.

REPORTING UNANTICIPATED PROBLEMS TO PARTICIPANTS

The study site will report UPs to participants who are directly involved in the UP, or if the UP identifies risks that may impact their willingness to participate. Appropriate actions for the continuation or withdrawal from the study will be discussed with the involved participants.

12.STATISTICAL CONSIDERATIONS

STATISTICAL HYPOTHESES

- Primary and Exploratory Efficacy Endpoint(s):

The null hypothesis for the primary efficacy endpoint is that biomarker measures (primary endpoint: vMRI; tertiary and exploratory endpoints: CSF biomarkers, blood inflammatory markers, saliva amyloid beta levels) will be the same in Siponimod- vs. placebo-treated study subjects after 12 months (end of dosing).
The alternative hypothesis is that Siponimod will improve biomarker measures at the same time point.

- Secondary Efficacy Endpoint(s):

The null hypothesis for the secondary efficacy endpoint is that cognitive measures (ADCOMS) will be the same in Siponimod - vs. placebo-treated study subjects after 12 (end of dosing) and 18 months (6 months follow up).
The alternative hypothesis is that Siponimod will improve cognitive measures at the same time points.

SAMPLE SIZE DETERMINATION

Mild AD subjects will receive an escalating dose of Siponimod up to 1 mg/day (N=70) or placebo (N=35) for 12 months (Table 2). We conducted a power analysis for sample size determination using the reported data from brain volume changes in SPMS subjects treated for at least 12 months with Siponimod or placebo[25]. We wished to fit a repeated measures ANOVA model with two treatment levels in drug vs. placebo, and 4-time measures to receiving the drug (i.e. vMRI at baseline, 6, 12, and 18 months). We defined $Y_{ij} = \mu + \alpha_i + \tau_j + (\alpha\tau)_{ij} + \varepsilon_{ijk}$, where Y_{ijk} denotes the measure on brain atrophy, α_i denotes the treatment effect $i = 1, 2$; τ_j denotes the time effect at time j=1, 2, 3, 4; $k = 1,\ n$; $(\alpha\tau)_{ij}$ is the interaction between time and treatment; and ε_{ijk} is the error term[21]. We study the effect of the drug on brain atrophy. The between-subject factor in our model is treatment, with two levels (study drug and placebo), and we allocate each treatment in the ratio 2:1. The within-subject factor is time, with four levels (0, 6, 12, and 18 months). The number of patients needed is determined to achieve a power of 0.90 at significance level $\alpha = 0.05$ for the test of the interaction between time and treatment, where the contrast over time contains all pairwise comparisons. We also intend to generate a plot of power versus sample size that covers the power range of 0.05 to 0.90.

The default Hotelling-Lawley F test is appropriate for this study, especially because it is the same as the Wald test in PROC MIXED with the DDFM=KR Kenward-Roger degrees-of-freedom method and an unstructured covariance model. We conjecture that the mean measure of brain atrophy will decrease by 20-30% (GLMPOWER Procedure SAS/STAT User's Guide 9.2). To characterize the variability, we specify a set of parameters that defines the entire covariance matrix of the residuals. We conjecture that the error standard deviation is the same at all four time points, with a value approximately 4-11, and we account for uncertainty by including both the lower and upper ends of this range in the sample size analysis. The power analysis is given in Table 1. A power of 90% is obtained with 90-95 subjects to determine a difference in brain atrophy over placebo. To account for a possible ~10% attrition rate, we will use 105 subjects in total.

Table 4: ***Power Analysis Versus Sample Size****.*

Index	Transformation	Effect	Actual Power	N Total
1	Time	Time	0.903	90
2	Mean (Dep)	Treatment	0.906	94

We chose to study mild AD subjects for the major reason that these patients do suffer continuous but limited/slow brain atrophy, which is measurable by vMRI[22], vs. mild cognitive impairment patients (MCI), whose brain atrophy may not progress enough on an annual basis for robust quantification. Furthermore, the functional disabilities in mild AD patients are not too pronounced yet vs. moderate and severe AD. Thus, if Siponimod is able to slow down brain atrophy, mild AD subjects will likely benefit the most by, hopefully, also slowing down functional decline.

POPULATIONS FOR ANALYSES

All subjects receiving at least 1 dose of study drug will be included in the safety analyses.
For efficacy analyses, subjects will be included only if they reach Visit 12 (Week 8) of the dosing period (i.e., patients withdrawn before Visit 12 and replaced will not be included in this efficacy analysis). Patients withdrawing or withdrawn from the study before Visit 12 are unlikely to demonstrate a change in the biomarkers investigated here (cognitive performance) given the effects of Siponimod in the CNS take several weeks to be measurable.

STATISTICAL ANALYSES

GENERAL APPROACH

All intra and inter-subject outcome measures collected during our study will be sent to Dr. Wilson at Arizona State University for statistical processing.

The data will be summarized using professional standards. The distributions of patients completing each study visit and the reason for early terminations will be tabulated for each treatment group. The duration of double-blind treatment within each treatment group and compliance rates will be summarized by descriptive statistics (mean, standard error, standard deviation, median, minimum, maximum and sample size) for days of treatment and for total days of exposure and by the distributions of patients exposed to various durations of treatment categories. Demographic and baseline characteristics will be summarized by descriptive statistics for all patients in each treatment group.

Volumetric MRI data measures will be computed to determine eventual brain volume changes between pre- and post-treatment, which will validate or refute our central hypothesis. Data from Aims 2-3 will also be used in correlational analyses for blood vs. AD central markers to investigate whether minimally invasive tests that use peripheral markers could be used as proxies to track drug performance. We wish to fit a repeated measures ANOVA model with two treatment levels (drug and placebo) and 13 time measures (for blood markers). We define $Y_{ij} = \mu + \alpha_i + \tau_j + \varepsilon_{ijk}$, where Y_{ij} denotes the measure on siponimod, α_i denotes the treatment effect i=1,2 , τ_j denotes the random effect at subject j=1, 2,..., 70, k=1,,,13 and ε_{ijk} is the error term. We assume that the random effect is distributed with mean zero and σ2. Paired data tests will be used to assess eventual biomarker changes between pre- and post-dosing datasets. Correlational statistics and linear regressions will be employed to determine possible dose-response for siponimod on the biomarkers investigated. If siponimod has an effect on inflammatory and AD biomarkers (either in blood, or in CSF), the current study should demonstrate an amelioration of these biomarker profiles. Also, we will use the results of cognitive tests to calculate the average conversion rate, continuous variables, and time to event analysis in our two groups.

Missing data will not be imputed, and all summary statistics will be reported based upon observed data. For limited analyses where formal inferential analyses are specified, rank-based methods will be used to incorporate subjects missing follow-up assessments.

ANALYSIS OF THE PRIMARY EFFICACY ENDPOINT(S)

Statistical analysis of the biomarkers and clinical assessments will be carried out by our biostatistician. Comparisons will assess the change from baseline for the following markers assessed at the frequency indicated in the schedule of visits Table 1):
- vMRI, which is used to estimate the annual rate of brain atrophy.

ANALYSIS OF THE SECONDARY ENDPOINT(S)

Statistical analysis of the biomarkers and clinical assessments will be carried out by our biostatistician. Comparisons will assess the change from baseline for the following markers assessed at the frequency indicated in the schedule of visits Table 1):

- Alzheimer's Disease Assessment Scale - Cognitive (ADAS-Cog13);
- Clinical Dementia Rating-Sum of Boxes (CDR-SOB);
- Mini Mental State Examination (MMSE);
- ADCOMS, which is calculated using components of the three cognitive tests indicated above.

SAFETY ANALYSES

- Concomitant Medications

Concomitant medications will be coded by the World Health Organization (WHO) Drug Dictionary and tabulated by the treatment group.

- Adverse Events

The incidence of all treatment-emergent AEs (TEAEs) and treatment-related AEs will be tabulated by treatment received. These AEs will be classified by system organ class and preferred term using the Medical Dictionary for Regulatory Activities (MedDRA). For incidence reporting, if a subject reported more than one AE that was coded to the same system organ class or preferred term, the subject will be counted only once for that specific system organ class or preferred term. A TEAE is defined as an event that first occurs or worsens in intensity after the administration of study drug. Events recorded between the time the informed consent is signed and the first study drug administration will be listed.

An overview of AEs, which includes subject incidence of TEAEs, treatment-related AEs, SAEs, deaths, and AEs leading to discontinuation, will be presented. For AEs presented by severity, the worst severity during the study will be presented for each subject.

The subject incidence of TEAEs and treatment-related AEs will be summarized by system organ class and preferred term.

- Serious Adverse Events

All SAEs will be listed and summarized in a similar manner to AEs.

- Clinical Laboratory Results

Clinical laboratory values will be measured by a central laboratory.
Summary statistics for actual values and for changes from baseline will be tabulated for laboratory results by scheduled visit. Subjects with clinical laboratory values outside of the normal reference range at any post-baseline assessment will be summarized. Shifts from baseline laboratory values (to worst severity per subject) will be tabulated.

- Electrocardiogram Results

The EKG parameters (heart rate, PR interval, QRS interval, QT interval, and QTc interval) at each time recorded (scheduled for screening and study end) as well as the change from screening will be summarized with descriptive statistics.

The overall EKG assessment will be reported as “Normal” or “Abnormal” with respect to relevant abnormalities by the investigator. A shift table comparing the EKG assessment over the treatment period (every abnormal) to screening will be presented.

- Vital Signs

The observed data at baseline and change from baseline for each measurement day will be summarized with descriptive statistics.

BASELINE DESCRIPTIVE STATISTICS

The distributions of patients completing each study visit and the reason for early terminations will be tabulated for each treatment group. The duration of double-blind treatment within each treatment group and compliance rates will be summarized by descriptive statistics (mean, standard error, standard deviation, median, minimum, maximum, and sample size) for days of treatment, for total days of exposure, and by the distributions of patients exposed to various durations of treatment categories. Demographic and baseline characteristics will be summarized by descriptive statistics for all patients in each treatment group.

PLANNED INTERIM ANALYSES

Formal interim analyses are not planned. Data will be reviewed by the DSMB for their assessment of safety and risk/benefit considerations.

SUB-GROUP ANALYSES

No sub-group analyses are planned.

TABULATION OF INDIVIDUAL PARTICIPANT DATA

Participant data will be listed by measure and time point.

EXPLORATORY ANALYSES

Statistical analysis of the exploratory CSF, blood, and saliva biomarkers will be carried out by our biostatistician. Comparisons will assess the change from baseline for the following markers assessed at the frequency indicated in the schedule of visits Table 1):

- Alzheimer's Disease Cooperative Study- Activities of Daily Living (ADCS-ADL)
- blood inflammatory markers (e.g., TNF-α, IL-1β, IL-6)
- CSF and plasma C-reactive protein
- CSF and plasma TNF-α
- CSF and plasma IL-1β
- CSF and plasma IL-6
- CSF and plasma IL-8
- CSF and plasma IL-10
- CSF Amyloid Beta 40, 42, total Tau and Phospho-Tau
- saliva Amyloid Beta 40 and 42.

13. SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

REGULATORY, ETHICAL, AND STUDY OVERSIGHT CONSIDERATIONS

INFORMED CONSENT PROCESS

CONSENT/ASSENT AND OTHER INFORMATIONAL DOCUMENTS PROVIDED TO PARTICIPANTS

This study will be conducted in accordance with the protocol and ethical principles stated in the Declaration of Helsinki or the applicable GCP, and all applicable federal, state, and local laws, rules, and regulations.

All data recorded in the CRF for subjects participating in this study will be transcribed from medical records, though the CRF may serve as the source document in some cases. When the CRF is used as the source, it will be documented in the study file.

Consent forms describing in detail the study intervention, study procedures, and risks are given to the participant and written documentation of informed consent is required prior to starting intervention/administering study intervention.

CONSENT PROCEDURES AND DOCUMENTATION

Each patient must be assessed for capacity to consent by the principal or sub-investigators in order to provide informed consent. If the patient is deemed unable to provide informed consent, they must have a legally authorized representative (LAR), and the LAR must review and sign the informed consent form. If the patient does not have an LAR, the patient must appear able to provide informed consent and must review and sign the informed consent form. If the patient is deemed unable to provide informed consent and does not have a LAR, they cannot participate in the study. In addition, the patient's study partner/informant (as defined in the study inclusion criteria) must sign the informed consent form. If the LAR and the patient's study partner/informant are the same individual, he/she should sign under both designations.

The investigator is responsible for presenting the risks and benefits of study participation to the subject (or legally appointed representative, as appropriate) in simple terms using the informed consent

document. The investigator will ensure that written informed consent is obtained from each subject (or legally appointed representative, as appropriate) and /study partner (if not the legally appointed representative) by obtaining the appropriate signatures and dates on the informed consent document prior to the performance of protocol evaluations or procedures, i.e. prior to or at the beginning of Visit 1 (screening). If the ICF is revised, subjects (or legally appointed representative, as appropriate) and study partner (if not the legally appointed representative) must be re-consented as per the Ethics Committee's recommendations.

STUDY DISCONTINUATION AND CLOSURE

This study may be temporarily suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be provided by the suspending or terminating party to investigator, funding agency, the Investigational New Drug (IND) sponsor and regulatory authorities. If the study is prematurely terminated or suspended, the Principal Investigator (PI) will promptly inform study participants, the Institutional Review Board (IRB), and sponsor and will provide the reason(s) for the termination or suspension. Study participants will be contacted, as applicable, and be informed of changes to the study visit schedule.

Circumstances that may warrant termination or suspension include, but are not limited to:

- Determination of unexpected, significant, or unacceptable risk to participants
- Insufficient compliance to protocol requirements
- Data that are not sufficiently complete and/or evaluable
- Determination of futility

Study may resume once concerns about safety, protocol compliance, and data quality are addressed, and satisfy the sponsor, IRB and/or Food and Drug Administration (FDA).

CONFIDENTIALITY AND PRIVACY

Participant confidentiality and privacy is strictly held in trust by the participating investigators, their staff, and the sponsor(s) and their interventions. This confidentiality is extended to cover testing of biological samples in addition to the clinical information relating to participants. Therefore, the study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study, or the data will be released to any unauthorized third party without prior written approval of the sponsor.

All research activities will be conducted in as private a setting as possible.

The study monitor, other authorized representatives of the sponsor, representatives of the Institutional Review Board (IRB), regulatory agencies or pharmaceutical company supplying study product may inspect all documents and records required to be maintained by the investigator, including but not limited to, medical records (office, clinic, or hospital) and pharmacy records for the participants in this study. The clinical study site will permit access to such records.

The study participant's contact information will be securely stored at the clinical site for internal use during the study. At the end of the study, all records will continue to be kept in a secure location for as long a period as dictated by the reviewing IRB, Institutional policies, or sponsor requirements.

Study participant research data, which is for purposes of statistical analysis and scientific reporting, will be transmitted to and stored at the study biostatistician. This will not include the participant's contact or identifying information. Rather, individual participants and their research data will be identified by a unique study identification number. The study data entry and study management systems used by clinical sites and by research staff will be secured and password protected. At the end of the study, all study databases will be de-identified and archived at BNI/Dignity Health, Phoenix, AZ.

FUTURE USE OF STORED SPECIMENS AND DATA

Data collected for this study will be analyzed and stored at BNI, Phoenix, AZ. After the study is completed, the de-identified, archived data will be transmitted to and stored at BNI, Phoenix, AZ, for use by other researchers, including those outside of the study.

With the participant's approval and as approved by local Institutional Review Board (IRB), de-identified biological samples (blood and CSF) will be stored for 15 years at the Translational Neurodegenerative Research Lab for SIPO1-AD with the same goal as the sharing of data with BNI in Phoenix, AZ. With consent from patients, these samples could be stored indefinitely and used to research the causes of Alzheimer's disease, its complications, and other conditions for which individuals with Alzheimer's disease are at increased risk, and to improve treatment. The Translational Neurodegenerative Research Lab will also be provided with a code-link that will allow linking the biological specimens with the phenotypic data from each participant, maintaining the blinding of the identity of the participant.

During the conduct of the study, an individual participant can choose to withdraw consent to have blood and/or CSF samples stored for future research. However, withdrawal of consent regarding biosample storage may not be possible after the study is completed.

When the study is completed, access to study data may be requested through the Principal Investigator.

KEY ROLES AND STUDY GOVERNANCE

Provide the name and contact information of the Principal Investigator and the Medical Monitor.

Principal Investigator
Marwan N. Sabbagh , MD, FAAN Staff Physician
Barrow Neurological Institute
240 W. Thomas Road, Suite 301 Phoenix, AZ 85013
Phone: 602-406-6262
E-mail: Marwan.Sabbagh@commonspirit.org

This protocol (and the attachments) will be conducted according to all stipulations of the protocol, including all statements regarding confidentiality, local legal and regulatory requirements, applicable US federal regulations and ICH guidelines. A Manual of Operations (MOP) will also be available and will identify the study team, their roles and responsibilities in conducting, managing and providing oversight of this trial.

SAFETY OVERSIGHT

An independent Data Safety Monitoring Committee (DSMB) comprised of three members not associated with the study will be secured prior to starting the study. This group of experts advises NIH and the Sponsor/Investigator. During the trial, the DSMB will review cumulative study data to evaluate safety, study conduct, and scientific validity and integrity of the trial.

CLINICAL MONITORING

A Medical Monitor has been secured to provide medical expertise to the study team including answering investigator questions about the protocol, patient eligibility and the medical management of subject AE/SAE assessment/treatment, and/ or medical emergencies.

There will also be a Study Monitor who is responsible for monitoring the conduct of the clinical trial on a regular basis as per GCP and ICH guidelines. He/she will inspect the case report forms at

regular intervals throughout the study to verify adherence to the protocol; completeness and accuracy of the data; and adherence to local regulations on the conduct of clinical research. The investigators agree to cooperate with the study monitor to ensure that any problems detected in the course of these monitoring visits are resolved.

The study monitor will inspect site facilities (e.g., pharmacy, drug storage areas, laboratories) and review study-related records to evaluate the trial conduct and compliance with the protocol, GCP, and applicable regulatory requirements. Results of locally administered tests such as EKG tracings and local clinical laboratory reports will be retained as source documents. At the monitoring visits, the progress of the study will be discussed with the Investigator and/or the Study Coordinator and the informed consent forms and CRFs will be checked for completeness and accuracy. Subjects' source documents should be available for review.

QUALITY ASSURANCE AND QUALITY CONTROL

To ensure accurate, complete, and reliable data, the investigators, helped by institutional data management experts, will do the following:

- Provide training for all raters of the functional and psychometric scales used in this study.
- Provide instructional material to site staff, as appropriate.
- Instruct the study personnel on how to complete CRFs and study procedures.
- Be available for consultation and stay in contact with the study site personnel by mail, email, telephone, and/or fax.
- Monitor the subject data recorded in the CRF against source documents at the study site.
- Review and evaluate CRF data and use standard computer edits to detect errors in data collection.

The study may be audited by the Sponsor or its representatives and/or regulatory agencies at any time.

To ensure the safety of participants in the study, and to ensure accurate, complete, and reliable data, the investigator will keep records of laboratory tests, clinical notes, and patient medical records in the patient files as original source documents for the study. If requested, the investigator will provide the sponsor, applicable regulatory agencies, DSMB, and IRB with direct access to original source documents.

DATA COLLECTION AND MANAGEMENT RESPONSIBILITIES

Data collection is the responsibility of the clinical trial staff at the site under the supervision of the site investigator. The investigator is responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported.

All source documents should be completed in a neat, legible manner to ensure accurate interpretation of data.

Hardcopies of the study visit worksheets will be provided for use as source document worksheets for recording data for each participant enrolled in the study. Data recorded in the electronic case report form (eCRF) derived from source documents should be consistent with the data recorded on the source documents.

Clinical data (including adverse events (AEs), concomitant medications, and expected adverse reactions data) and clinical laboratory data will be entered into the IND/IDE Redcap Cloud, a 21 CFR Part 11-compliant data capture system provided by BNI, Phoenix, AZ. The data system includes password protection and internal quality checks, such as automatic range checks, to identify data that appear inconsistent, incomplete, or inaccurate. Clinical data will be entered directly from the source documents.

STUDY RECORDS RETENTION

Study documents should be retained for a minimum of 2 years after the last approval of a marketing application in an International Conference on Harmonization (ICH) region and until there are no pending or contemplated marketing applications in an ICH region or until at least 2 years have elapsed since the formal discontinuation of clinical development of the study intervention. These documents

should be retained for a longer period, however, if required by local regulations. No records will be destroyed without the written consent of the sponsor, if applicable. It is the responsibility of the sponsor to inform the investigator when these documents no longer need to be retained.

PROTOCOL DEVIATIONS

A protocol deviation is any noncompliance with the clinical trial protocol, International Conference on Harmonization Good Clinical Practice (ICH GCP), or Manual of Procedures (MOP) requirements. The noncompliance may be either on the part of the participant, the investigator, or the study site staff. As a result of deviations, corrective actions are to be developed by the site and implemented promptly. These practices are consistent with ICH GCP:

- 4.5 Compliance with Protocol, sections 4.5.1, 4.5.2, and 4.5.3
- 5.1 Quality Assurance and Quality Control, section 5.1.1
- 5.20 Noncompliance, sections 5.20.1, and 5.20.2.

It is the responsibility of the site investigator to use continuous vigilance to identify and report deviations within 5 working days of identification of the protocol deviation, or within 5 working days of the scheduled protocol-required activity. All deviations must be addressed in study source documents, reported to the NIA Program Official and Barrow Neuological Institute. Protocol deviations must be sent to the reviewing Institutional Review Board (IRB) per their policies. The site investigator is responsible for knowing and adhering to the reviewing IRB requirements. Further details about the handling of protocol deviations will be included in the MOP.

PUBLICATION AND DATA SHARING POLICY

It is the intent of the investigator to publish the final results of this study as a collaborative effort using criteria for authorship established by international guidelines. Post-study, a summary of the results will be made available to all participants and study personnel.

This study will be conducted in accordance with the following publication and data sharing policies and regulations indicated below.

National Institutes of Health (NIH) Public Access Policy ensures that the public has access to the published results of NIH funded research. It requires scientists to submit final peer-reviewed journal manuscripts that arise from NIH funds to the digital archive PubMed Central upon acceptance for publication.

This study will comply with the NIH Data Sharing Policy and Policy on the Dissemination of NIH-Funded Clinical Trial Information and the Clinical Trials Registration and Results Information Submission rule. As such, this trial will be registered at ClinicalTrials.gov, and results information from this trial will be submitted to ClinicalTrials.gov. In addition, every attempt will be made to publish results in peer-reviewed journals. Data from this study may be requested from other researchers 10 years after the completion of the primary endpoint by contacting the Principal Investigators.

In addition, this study will comply with the NIH Genomic Data Sharing Policy, which applies to all NIH-funded research that generates large-scale human or non-human genomic data, as well as the use of these data for subsequent research. Large-scale data include genome-wide association studies (GWAS), single nucleotide polymorphisms (SNP) arrays, and genome sequence, transcriptomic, epigenomic, and gene expression data.

CONFLICT OF INTEREST POLICY

The independence of this study from any actual or perceived influence, such as by the pharmaceutical industry, is critical. Therefore, any actual conflict of interest of persons who have a role in the design, conduct, analysis, publication, or any aspect of this trial will be disclosed and managed. Furthermore, persons who have a perceived conflict of interest will be required to have such conflicts managed in a way that is appropriate to their participation in the design and conduct of this trial. The study leadership in conjunction with the NIA has established policies and procedures for all study group members to disclose all conflicts of interest and will establish a mechanism for the management of all reported dualities of interest.

ADDITIONAL CONSIDERATIONS

The IRB supervising the present study will be from the Barrow Neurological Institute

14. ABBREVIATIONS

Abbreviation	Term
Aβ	Amyloid Beta
AD	Alzheimer's Disease
ADAS	Alzheimer's Disease Assessment Scale
ADAS-Cog	Alzheimer's Disease Assessment Scale – Cognitive subscale
ADCS-ADL	Alzheimer's Disease Cooperative Study - Activities of Daily Living
AE	Adverse Event
ALS	Amyotrophic lateral sclerosis
ALT	Alanine aminotransferase
aMCI	Amnestic mild cognitive impairment
ANDA	Abbreviated New Drug Application
Anti HCV	Hepatitis C Antibody
ApoE	Apolipoprotein subtype E
APP	Amyloid precursor protein
ARIA-E	Amyloid-related imaging abnormalities - edema/effusions
ARIA-H	Amyloid-related imaging abnormalities - hemosiderin deposition
BACE1	Beta amyloid precursor protein cleaving enzyme 1
BP	Blood pressure
CBC w/ diff	Complete blood count with differential
CDSCO	Central Drugs Standard Control Organization
CDR	Clinical Dementia Rating Scale
CDR-SOB	Clinical Dementia Rating Scale - Sum of Boxes
Cmax	Maximum plasma concentration
CMH	Cerebral microhemmorhage
CMP	Comprehensive metabolic panel
CNS	Central nervous system
CONSORT	Consolidated Standards of Reporting Trials
CRF	Case report form
CRO	Contract Research Organization
CRP	C reactive protein
CSE	Clinically significant event
CSF	Cerebrospinal fluid
CT	Computed Tomography
CVA	Cardiovascular Accident
CVE	Cerebral Vasogenic Edema
DOB	Date of Birth
DSMB	Data Safety Monitoring Board
DSM	Diagnostic and Statistical Manual of Mental Disorders
eCRF	Electronic Case Report Form
EDTA	Ethylenediaminetetraacetic Acid
EKG	Electrocardiogram
ELISA	Enzyme-linked immunosorbent assay
EMA	European Medicines Agency
FBRP-PET	Florbetapir- Positron Emission Tomography
FDA	Food and Drug Administration
Abbreviation	Term

FLAIR	Fluid-Attenuated Inversion Recovery image
FSH	Follicular Stimulating Hormone
FTD	Frontotemporal dementia
FOV	Field of View
GCA	Global Cortical Average
GCP	Good Clinical Practice
G-CSF	Granulocyte-colony stimulating factor
GDS	Geriatric Depression Scale
GWAS	Genomic-Wide Association Studies
HC	Hippocampus
Hep	Hepatitis
HIPAA	Health Insurance Portability and Accountability Act
HIV	Human Immunodeficiency Virus
ICH	International Conference on Harmonization
IEC	Independent Ethics Committee
IFN	Interferon
IND	Investigational New Drug
IL	Interleukin
IRB	Institutional Review Board
IV	Intravenous
LAR	Legally authorized representative
LP	Lumbar puncture
MCI	Mild cognitive impairment
MDS	Myelodysplastic Syndrome
MedDRA	Medical Dictionary for Regulatory Activities
MOP	Manual of Operations
MM	Multiple myeloma
MMSE	Mini-Mental State Examination
MPRAGE	Magnetization prepared rapid acquisition gradient echo
MRI	Magnetic Resonance Imaging
mRNA	messenger Ribonucleic Acid
NCT	ClinicalTrials.gov Identifier
NEX	Number of Excitations
NI	Neurological Institute
NIA-AA	National Institute of Aging and Alzheimer's Association
NIH	National Institutes of Health
NV	Nevada
NSAIDs	Non-steroidal anti-inflammatory medications
OCT	Optical Coherence Tomography
PET	Positron emission tomography
PK/PD	Pharmacokinetics/Pharmacodynamics
POCT	Point of Care Testing
QA	Quality Assurance
QT	Interval between start of Q and end of T wave
QTc	Corrected QT corrects for heart rate extremes
RBC	Red Blood Cell Count
Abbreviation	Term
RPS	Research Pharmaceutical Services Inc.

SAC	Safety Assessment Committee
SAE	Serious Adverse Event
SoA	Schedule of Activities
SoC	Standard of Care
SPM8	Statistical Parametric Mapping software version 8
SUVr	Standard Uptake Value Ratio
T2	2 Tesla MRI image
TE	Echo Time (MRI)
Tmax	Time to maximum plasma concentration
TNFa	Tumor Necrosis Factor alpha
TSH	Thyroid-Stimulating Hormone
TST	Total time to make a horizontal saccade
ULN	Upper Limit of Normal
US	United States
UP	Unanticipated Problems
USA	United States of America
vMRI	Volumetric Magnetic Resonance Imaging
WBC	White blood cell count

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