Study Protocol

Spironolactone Safety in African Americans with Mild Cognitive Impairment and Early Dementia

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PROTOCOL TITLE: Spironolactone Safety in African Americans with Mild Cognitive Impairment and Early Dementia.

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REVISION HISTORY

Revision #	Version Date	Summary of Changes
1	12/08/2021	1. Blood pressure criteria revised 2. Inclusion and exclusion criteria revised 3. Study measures revised 4. Optional lumbar puncture added
2	09/28/2023	1. The title was updated to be inclusive of participants with early stage dementia from vascular and neurodegenerative conditions like Alzheimer's disease 2. Inclusion and exclusion criteria revised to include participants with MoCA $\geq$13 3. Study visit windows updated 4. Study drug dose assignment to remain at 25mg/day 5. Compensation per visit adjusted to more equitably account for time involved per visit. Total compensation for participation remains up to $150. 6. Minor administrative revisions

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1. Study Summary

Study Title	**S**pironolac**T**one Safety in **A**frican Americans with Mild Cog**N**itive Impairment and early **D**ementia (**STAND**)
Study Design	A double-blind randomized clinical trial comparing spironolactone to placebo.
Primary Objective	The primary objective of the study is to investigate the safety and tolerability of spironolactone in AA adults with MCI and early dementia.
Secondary Objective(s)	The secondary objective is to estimate the effect size difference with treatment of spironolactone vs. placebo on cognitive performance in AA with MCI/early dementia.
Research Intervention(s)/Interactions	Spironolactone 25 mg
Study Population	AA over aged ≥ 50 with MCI/early dementia
Sample Size	30
Study Duration for individual participants	12-months
Study Specific Abbreviations/ Definitions	AA: African Americans AD: Alzheimer's disease AE: adverse event DBP: diastolic blood pressure EES: estimated effect size FDA: Food and Drug Administration GADRC: Goizueta Alzheimer's Disease Research Center HVLT-T: Hopkins Verbal Learning Test-Revised MCI: mild cognitive impairment mg: milligrams MoCA: Montreal Cognitive Assessment PI: principal investigator SBP: systolic blood pressure TMT A and B: Trail Making Test parts A and B
Funding Source (if any)	Alzheimer's Association Goizueta Alzheimer's Disease Research Center

2. Objectives

2.1. The primary objective of this study is to investigate the safety and tolerability of spironolactone in AA with MCI/early dementia.

2.2. The secondary objective is to estimate the effect size difference with the treatment of spironolactone vs. placebo on cognitive performance in AA with MCI/early dementia.

2.3. The hypothesis to be tested is that compared to placebo, there will not be a significant difference in AE counts and attrition with spironolactone.

3. Background

3.1. Introduction

Alzheimer's disease (AD) impacted 5.5 million older adults in 2018[1] and is predicted to impact 15 million people by 2060.[2] Older African Americans (AA) are more likely to be diagnosed with AD as its prevalence is two times greater than non-Hispanic Whites.[3-6] AD is also associated with a racial difference in cost as AA have higher costs compared to Whites.[7] National costs for AD and associated dementia exceeded 270 billion dollars in 2018 of which of which nearly 70% was covered by Medicare and Medicaid.[1] AD thus represents a significant public health challenge that is associated with racial disparity in prevalence and cost. Exploring ethnoracial differences in biological risk[8] may offer insight into the underlying factors of the racial disparity in the prevalence of AD. Studies in people with hypertension suggest the renin angiotensin aldosterone system (RAAS) is implicated with impaired cognition.[9] Moreover, many RAAS tissue effects are mediated by aldosterone, a mineralocorticoid receptor (MR) agonist. Elevated aldosterone has been shown to be related to cognitive impairment that may reflect effects from MR in the hippocampus and altered cerebral blood flow.[10,11] Multiple studies demonstrate AA can have inappropriately elevated aldosterone,[12,13] which can increase cardiovascular disease risk, more so than Whites.[14-17] It follows that elevated aldosterone possibly impacts cardiovascular and cerebrovascular function, which are both associated with cognitive decline.[10,11,18,19] Despite evidence implicating RAAS with cognitive impairment and AD, [20,21] its specific role in AA is unclear. Results from cardiovascular studies suggest superior outcomes in AA with an aldosterone antagonist compared to other RAAS modulators.[22,23]

3.2. Pertinent literature and data

A. *Aldosterone and cognition:* Aldosterone activates MR, which are abundant in the hippocampus that is critical to memory and learning.[18] Moreover, brain RAAS and aldosterone in particular, are shown to influence cognitive function.[9,24,25] Animal studies indicate that MR activation by aldosterone and its precursor, corticosterone, can lead to cognitive impairment.[26-30] In addition to central effects, aldosterone is involved in peripheral vascular regulation with the vascular endothelium as a key target.[31-33] Vascular endothelial dysfunction is more prevalent in AA [34] and may contribute to cognitive decline that may be linked to AD.[35,36]. Peripheral vascular effects of aldosterone may also offer prognostic value for AD.[37] Elevated aldosterone can thus promote impaired cognition through MR actions and vascular alterations including cerebral blood flow.[11,38-43]

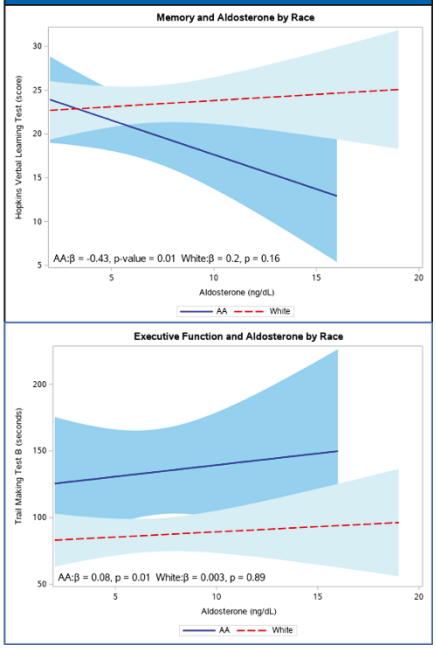
3.2.2. *Potential role for spironolactone in AA*: Hypertension studies demonstrate that AA have aldosterone elevation[12,13] that correlates with increased cardiovascular disease, more so than

whites.[14-17] RAAS activation in AA is characterized by low renin-elevated aldosterone, which alters vascular hemodynamics.[16,44] The low renin-high aldosterone pattern is attributed to a MR mutation that is associated with enhanced activation by aldosterone.[45] Despite a lack of demonstrated efficacy by some RAAS agents to treat cardiovascular disease in AA due to low renin, aldosterone antagonists are shown to have beneficial effects.[46] Studies of treatment for PTSD[47,48] and depression [49,50] demonstrate beneficial effects from RAAS modulator actions at brain MR receptors. Moreover, data from animal and human studies show that aldosterone antagonists also have beneficial cognitive effects.[51,52] Investigation of the relationship between spironolactone and MCI might offer a new, repurposed indication for MR antagonist therapy in AD, especially in AA given the low renin-high aldosterone RAAS pattern. The largest percentage increase in AD between 2018 and 2025 is expected to occur in the Western and Southeast regions of the United States (US),[1] for which the Southeast has the highest AA population in the US.[53] There is growing awareness that unrecognized individual and environmental factors can confound the outcomes of AD clinical trials.[54] Thus, the exploration of ethnoracial risk in a vulnerable population (AA with MCI) living in a high-risk region may yield important insight on AD risk and disease progression. This clinical trial will explore the safety and tolerability and will estimate the effect size of the administration of spironolactone (aldosterone antagonist) on cognitive performance in AA with MCI.

3.3. Preliminary Data

Elevated aldosterone is associated with cognitive performance in AA (**Figure 1**). Elevated aldosterone has cardiovascular and cerebrovascular effects that are associated with cognitive impairment. The objective of the study was to explore the relationship between aldosterone and cognitive performance in a biracial cohort. A secondary data analyses from the Antihypertensives and Vascular Endothelial and Cognitive Function (AVEC) trial was performed. Executive function was measured by Trail Making Test (TMT) parts A and B. Memory was assessed by the Hopkins Verbal Learning Test-Revised (HVLT-R). Regression models accounting for demographics and body mass index (BMI) were performed for estimates of aldosterone effects on cognitive tests. Data for 49 people (30.6% AA, 59.2% female, average BMI 28.8 kg/m^2, and mean age 71.7 years) was analyzed. AA had a trend for higher aldosterone (AA: 6.98 ng/dL vs. Whites: 6.89 ng/dL, p = 0.95), but poorer executive performance per longer TMT-A (46.2 seconds vs. 33.7 seconds, p < 0.002) and TMT-B (129 seconds vs. 87.6 seconds, p < 0.002). Regression models accounting for age, BMI, sex, and education demonstrated that higher aldosterone was associated with poorer memory (HVLT-R: β = -0.43, p = 0.01) and executive function (TMT-B: β = 0.08, p = 0.01) in AA but not in Whites (Figure 2). These results indicate that elevated aldosterone may have a significant clinical relationship with cognitive performance in AA, which suggest that blocking aldosterone could have a unique cognitive impact in AA.

Figure 1. Aldosterone and Cognition



4. Study Endpoints *

4.1. The primary study endpoints are the safety measures of AE counts and attrition rate.

4.2. The secondary study endpoints are efficacy measures that include change in TMT A and B, HVLT-R, and Digit Span from baseline to the 12-month visit.

5. Study Intervention/Investigational Agent

5.1. **Randomization**

Participants will be randomized to either spironolactone or placebo in a 1:1 fashion. Randomization and study intervention will occur after baseline data collection. Participants will then be started on the study medication. For all study related testing, the test results will not be disclosed to participants unless deemed necessary by the PI.

5.2. **Drug description**

Spironolactone is approved by the Food and Drug Administration for the treatment of primary hyperaldosteronism; edema in persons with congestive heart failure, cirrhosis, and nephrotic syndrome; essential hypertension; hypokalemia; and severe heart failure. It is well tolerated by young and elderly individuals and has been used extensively in clinical practice. Risks of the study drug are described in detail in section 15.3, Drug Risks.

5.3. **Drug intervention, administration, and duration**

Study participants will receive a 25 mg dose of spironolactone or placebo. Blood pressure will be monitored during regularly scheduled follow-up visits to ensure the following criteria are present: 1) systolic blood pressure (SBP) > 100 mmHg, 2) diastolic blood pressure (DBP) > 60 mmHg, or 3) no reported symptoms of hypotension (dizziness or weakness) for SBP < 100 or DBP < 60. Investigations using spironolactone up to 200 mg per day for non-hypertensive conditions have established its effectiveness without the occurrence of symptomatic hypotension.[55-60] The 100 mmHg cut-off is based on evidence that a non-symptomatic decline of SBP by 5 to 10 mm Hg can occur in persons receiving spironolactone.[60,61] Therefore, a 100 mm Hg cutoff for SBP decreases the risk of developing symptomatic hypotension (defined as SBP < 100 mm Hg along with symptoms of pre-syncope or syncope). Treatment will be provided in similar capsule format to be taken once a day by mouth. The study investigators and personnel will be blinded to drug assignment. Participants will be blinded to drug assignment during their study participation but will be unblinded once they complete the study as detailed in section 17.1.3. Blinding. Participants will be treated for 12-months.

5.4. **Drug handling**

The study drugs will be stored in the medication room of the Investigational Drug Service (IDS) Pharmacy, located at Emory Clinic Bldg. A, Suite AT111, 1365 Clifton Road, NE, Atlanta, Georgia 30322. Access to the medication room is controlled by badge swipe and limited to the IDS Pharmacists. Accountability records are maintained for all investigational product (IP). Prescriptions will be shipped directly by IDS, picked up by study staff, or delivered by a courier to the study location to be given to participants.

5.5. **Permitted interventions**

Participants will continue to receive their usual care from their primary care provider (PCP). In the event a participant becomes hypertensive during the trial, they will be referred to their PCP for further hypertension management. Participants will be allowed to use any class of antihypertensive medication except RAAS modulating medications: ACEi, ARB, renin inhibitor, and aldosterone antagonists. The participant's PCP will be notified of the potential that the subject might be receiving spironolactone to avoid overlap.

5.6. **Prohibited interventions**

The additions of an ACEi, ARB, renin inhibitor, and aldosterone antagonist are prohibited during the study period. If a participant needs to be on a RAAS modulator then they will have their study intervention discontinued but will continue in the study if they already had more than 6 months of spironolactone. Participating in another interventional clinical trial with antihypertensive drugs is also prohibited. However, participation in an observational study is allowed.

5.7. **Medication adherence assessment**

Participants will be asked to bring their study medication bottles to the study center at each visit. Medication compliance will be assessed using pill count during the follow-up periods. We define a compliance rate for a time period (t):

$$\frac{\#\ used\ pills\ prescribed\ for\ the\ number\ of\ days(t) - \#\ pills\ remaining\ or\ unused\ for\ time(t)}{\#\ pills\ prescribed\ for\ time(t)} \times 100$$

6. Procedures Involved*

6.1. Study schematic

Evaluation	Assessments
Screening	
	A. Informed consent
	A. ADCS-MCI ADL B. Functional Assessment Questionnaire C. Logical Memory Delayed Paragraph Recall D. Montreal Cognitive Assessment E. Subjective Memory Questionnaire F. Functional Assessment Staging Test G. University of California San Diego Brief Assessment of Capacity to Consent
	Exclusion criteria: A. Heart failure, cirrhosis, renal disease (creatinine > 2.0 mg/dL) B. Hyperkalemia (K^+ > 5.5 mEq/dL) C. Clinical evidence of volume depletion (dehydration) D. HTN with uncontrolled blood pressure > 160/90 mmHg E. RAAS modulating drugs (ACEi, ARB, renin inhibitor, aldosterone antagonist)
Study	
Interview	A. Demographics B. History 1. Medical 2. Social 3. Family
Questionnaires	A. Patient Health Questionnaire-8 B. Physical Activity Scale for the Elderly C. Life's Simple 7 D. Stress & discrimination: PSS, RES, EDS, & Perceived Hostility E. Brief Trauma Questionnaire F. Pittsburg Sleep Quality Index G. Connor-Davidson Resilience Scale
Cognitive assessments	A. Quick Dementia Rating System B. Trail Making Test parts A & B C. Rey Auditory Verbal Learning Test D. Number Span E. MoCA (6-months, 12-months) F. Verbal Fluency (F & L)
Physical assessment	A. Blood pressure (AHA procedure), Body mass index, Short Physical Performance Battery
Laboratory assessment	A. Creatinine B. Electrolytes C. Liver function tests D. Blood glucose E. Inflammation panel
Vascular assessment	A. PWV B. PWA

ADCS-MCI ADL: Alzheimer's Disease Cooperative Study Mild Cognitive Impairment Assisted Daily Living Inventory, **PSS**: Perceived Stress Scale, **RES**: Race-related Events Scale, **EDS**: Everyday Discrimination Scale, **PWV/PWA**: pulse wave velocity/analysis, **AHA**: American Heart Association (mean systolic and diastolic blood pressure)

6.2. **Study overview**

There are six encounters/visits in this study: a screening visit and five evaluation visits. The study measures and procedures performed at each visit are summarized in **Table 1**. At least 1 follow-up phone call will also occur between 6 and 12 months. An optional modified phone/virtual visit may be completed as needed for any visit to collect data that can be reasonably collected by phone.

6.3. **Screening visit**

A. Obtain and document informed consent from the potential participant.
B. Demographic information (age, sex, self-identified race/ethnicity), education level, and the participant's contact information will be collected.
C. The identity and contact of the study partner will also be collected.
D. Assess inclusion/exclusion criteria
E. Medication inventory and medical record review
F. Laboratory for screening and safety: TSH, B12, complete blood count (CBC), and comprehensive metabolic panel (CMP)

6.3.1 Neuropsychological screening

The screening cognitive assessment that determines study eligibility includes:

A. Alzheimer's Disease Cooperative Study-Mild Cognitive Impairment Activities of Daily Living (ADCS-MCI ADL) is a 24-item scale using activities of daily living to distinguish MCI from normal cognition.[62]
B. Functional Assessment Staging in Alzheimer's disease (FAST) is a validated tool that measures functional deterioration in persons with AD throughout the disease course.[63]
C. Subjective Memory Questionnaire (SMQ) is a report of subjective memory loss per the participant or the study partner.[64]
D. Logical Memory (Delayed paragraph recall, part A) is part of the Wechsler Memory Scale-Revised with a maximum score of 25.[65]
E. Montreal Cognitive Assessment (MoCA) is a 30-point scale that assesses multiple cognitive domains and is equivalent to the Mini-Mental Status Examination in the detection of cognitive deficits. [66]
F. Functional Assessment Questionnaire (FAQ) will be used to gauge functional abilities related to performance ratings in 10 complex higher-order activities.[67]
G. University of California San Diego Brief Assessment of Capacity to Consent (UBACC) is a 10-item questionnaire assessing key areas of the informed consent process. A score greater than 14.5 correlates with a score greater than 16 on the MacArthur Competency Assessment Tool for Clinical Research (MacCAT-CR).[68] For the STAND study, participants who are unable to answer the capacity assessments questions correctly (UBACC score of <14.5) may still qualify for the study but will need to have a LAR and a study partner.

6.4. **Study visits**

The timing for the collection of study measures and procedures is shown in **Table 1**.

6.4.1 Baseline visit

A. Random group assignment: spironolactone vs. placebo
B. The following data will be collected during the participant's interview with study personnel:
    1. Demographics, social history, and medical history

Medication inventory: participants will be asked to bring all prescribed medication bottles. The medication inventory will be done by inspecting the participant medication and recording the name, dose, frequency and indication if known.[69]

C. Questionnaires:
   1. Patient Health Questionnaire-8 (PHQ-8) is an 8-item questionnaire that assess depressive symptoms over the preceding two-week period with higher scores indicating more depressive symptoms.[70] The PHQ-8 is comparable to the PHQ-9 for diagnosing and assessing major depressive disorder.[71]
   2. Physical Activity Scale for the Elderly (PASE) assesses occupational, household, and leisure activities during a one-week period and has high reliability in older adults.[72,73]
   3. Additional questionnaires measuring psychosocial stress and discrimination will be collected during various study visits, including the Perceived Stress Scale (PSS),[74] NIH Toolbox Perceived Stress and Hostility Scale,[75,76] Race-Related Events Scale (RES), and Everyday Discrimination Scale (EDS).[77]

D. Cognitive assessment:
   1. Trail Making Test (TMT) parts A and B assess executive function in patients with cerebrovascular and cardiovascular risk.[78,79]
   2. Number Span is a brief test that assesses immediate memory and attention.[80,81]
   3. Rey Auditory and Verbal Learning Test (RAVLT) is a 15-item neuropsychological measure of attention, concentration, verbal learning, and memory that can identify persons at risk for cognitive decline and progression to dementia over time.[82,83]

E. Physical assessments:
   1. BP will be measured in accordance with guidance from the American Heart Association (AHA). The participant will be in the sitting position, rested for 5 minutes, wearing the appropriate cuff size (covering 60% of upper arm length and 80% of arm circumference), and correct cuff placement (1-2 inches above brachial pulse on bare arm). Measurement will be done manually using the bell of the stethoscope or with a validated and calibrated automatic machine. Blood pressure will be measured in the non-dominant arm unless contraindicated. The dominant hand is based on the hand used for writing. The non-dominant arm will be used throughout the study. We will obtain 2 seated readings followed powerby 2 standing blood pressure measurements at 1 and 3 minutes during each visit.
   2. Weight and height for each participant will be recorded using a stadiometer to measure height and a balance beam scale to measure weight without shoes.
   3. Body mass index (BMI) for each participant will be calculated based on the following formula, $\boldsymbol{weight(kg) \div height(m)\text{^}2}$

F. Laboratory measures
   1. Safety: CBC and CMP if they were not collected at the screening visit.
   2. Biospecimen collection: inflammation panel

G. Vascular measures: vascular stiffness/elasticity will be assessed with the carotid-femoral pulse wave velocity (PWV) and radial pulse wave analysis (PWA) using the SphymoCor device (SphygmoCor®, Atcor Medical, Australia).[86-88] An augmentation index (AI) that is normalized for a heart rate of 75 beats per min (AI@75 bpm) will be derived.

H. Lumbar puncture (optional): A lumbar puncture (LP) will be performed if participants are eligible based on the inclusion criteria. The LP will be performed per the ADNI procedure manual at the L3-4 or L4-5 interspace with a 24-gauge Sprotte atraumatic spinal needle. The Sprotte needle decreases the frequency of post LP headaches to < 5%.[89] Approximately 15 to 20 mL of cerebrospinal fluid (CSF) will be collected into polypropylene transfer tubes followed by freezing on dry ice within 1 hour after collection. Aliquots (0.5ml) will be prepared from these samples

after thawing (1 hour) at room temperature and gentle mixing. The aliquots will be stored in bar code–labeled polypropylene vials at −80°C. In addition to routine assessment (cells, proteins, albumin, glucose), Aβ1-42, t-Tau, and p-Tau181p will be measured.

6.4.2 Follow-up visits

A. Adverse events
B. Medication inventory
C. Questionnaires: PHQ-8, Life's Simple 7, PASE, PSS, RES, EDS, Perceived Hostility, BTQ, PSQI, Q-DRS.
D. Cognitive measures: TMT A and B, Number Span, RAVLT, and MoCA
E. Physical measures: BP, weight and height, and BMI
F. Laboratory measures
G. Safety (BMP, CBC) and biospecimen collection: inflammation panel
H. Vascular measures: PWV/PWA
I. Lumbar puncture (optional)

Table 1. Clinical trial measures and timing of measurement or procedures							
Phase:	Screen	Baseline	RANDOMIZATION	Follow-up			
Number of Visits	1-2	1-2		1	1	1	1-2
Month(s)	-1	0		0.50-1	3	6	12
Informed consent	X						
Eligibility	X						
Medication inventory	X	X		X	X	X	X
Weight/Height~	X	X		X	X	X	X
Blood pressure	X	X		X	X	X	X
Screening Labs (PT/INR, TSH, B12, CMP, CBC)	X						
Cognitive Screening	X						
Cognitive Battery		X				X	X
Questionnaires (refer to section 6.4, Study Visits)		X					X
Vascular measures (PWV/PWA)		X					X
Functional battery (FAQ, ADCS-MCI ADL)		X*					X
LP for CSF for biomarker and molecular analysis		X**					X
Safety Labs (CMP & CBC)	X			X#	X#	X#	X^
Laboratory for biospecimen collection: inflammation panel		X				X	X
Time* (minutes)	120	150-210		30	30	120	150-210

^ Safety labs at 12 months include BMP + CBC.
If subject started taking any blood thinners during the study, PT/INR will be repeated before LP at 12 months.
~ Height collected only at screening
** If subject has had a lumbar puncture (LP) within the 6 months prior to their screening/baseline visit and samples are available for measuring CSF outcomes, then no LP will be done at baseline. Otherwise, LP will be done at baseline to obtain CSF for outcome measures.
# Safety labs at 1, 3, and 6 months include BMP (BUN, creatinine, & electrolytes)
*FAQ will be skipped at BL if already completed at screening

7. Data and Specimen Banking* N/A ☐

7.1. Biospecimens including CSF from LP procedures that are collected at the baseline and follow-up visits will be stored for future analysis. The samples will be stored at 6 Executive Park. Vials containing the samples will be labeled with the participant ID numbers, name of the study, and date collected. They will be stored at the cryogenic storage facility until time of processing.

7.2. The release of the specimens will follow the Emory Goizueta Alzheimer's Disease Research Center (GADRC) processes for release of biospecimens.

8. Sharing of Results with Participants*

8.1. ***Sharing participant information***

De-identified data, i.e., data that has been stripped of all information that can identify participants, may be placed into public databases where, in addition to having no direct identifiers, researchers will need to sign data use agreements before accessing the data. Any personal information that could identify participants before information is shared will be removed. This will ensure that, by current scientific standards and known methods, it would be extremely unlikely for anyone to be able to identify participants from the information that is shared.

Data [and specimens] from this study may be useful for other research being done by investigators at Emory or elsewhere. To help further science, de-identified data and/or specimens may be provided to other researchers. If data is shared, then no identifying information will be included.

In general, individual results from the study of the samples will not be provided to participants. If something of urgent medical importance is detected then participants will be informed, although it is expected that this will be a very rare occurrence.

8.2. **Safety management**

8.2.1 Clinical adverse events

Adverse events (AE) will be monitored throughout the study with inquiries at each in-person and telephonic visit. The proposed study utilizes widely used and approved procedures, including commonly used neuropsychological questionnaires and interview materials; blood pressure measurement; LP; (PWV/PWA); and blood draws.

8.2.2 Adverse event rating

During the study, any problem reported by the participants will be recorded in the participant's chart (adverse event log), along with the start and stop dates. The problem may be related to research participation, or the drug used in the study (drug reaction). All Aes will be rated on the following characteristics:

A. Related to the research: all AEs are evaluated regarding their relationship to research participation or the study drug as:
    1. Related
    2. Possibly related
    3. Probably related
    4. Unrelated
    5. Unknown

B. Severity: the severity of AE will be rated on the following scale:

1. Mild
2. Moderate
3. Severe
4. Life-threatening
5. Death

C. Anticipated or unanticipated: we use the Emory IRB definition of an unanticipated event, i.e., "unexpected, not described in the study documents, or if described before, it is now presenting with increased severity, duration, or frequency".

8.2.3 Serious adverse event definition

A serious adverse event (SAE) is defined using the Code of Federal Regulations Title 21 definition as any event that results in any of the following outcomes:

A. Death
B. Life-threatening AE
C. Inpatient hospitalization or prolongation of existing hospitalization
D. Persistent or significant incapacity
E. Substantial disruption of the ability to conduct normal life functions
F. Congenital anomaly/birth defect

8.2.4 Unanticipated problems

We follow the Office for Human Research Protections (OHRP) definition of unanticipated problem (UP) when it meets **all** the following criteria:

A. Unexpected in terms of nature, severity, or frequency
B. Related or possibly related to participation in the research
C. Suggests that the research places subjects or others at a greater risk of harm than was previously known or recognized

8.3. **Reporting procedures**

8.3.1 Reporting adverse events and deaths

A. UP that are also AE and SAE, which occur while a participant is being screened or evaluated will be reported to the Emory IRB within 10 days from the identification of the event by the PI using the Emory eIRB website.
B. All AE will be assessed for the need for prompt reporting. We follow the guidelines established by the Emory IRB for AE reporting.
C. Any AE that fulfills the criteria to be "unanticipated" (section **8.1.3**), probably or possibly related to study participation (drug effect or as a consequence of a study procedure) and involving risk for participants or others is considered an UP and will be reported to the Emory IRB within 10 days.
D. If an AE can be attributed to an underlying medical condition, then it will not be considered related and hence not reportable.
E. Any SAE (section **8.1.2**) that does not fulfill all the criteria to be an UP will be reported during continuing review.
F. Deaths of participants during the study will be promptly reported to the IRB if related to study participation and reported during continuing if they are not related to study participation. A summary of AE/SAE/Deaths will be presented to the DSMB by the study biostatistician.

8.3.2 Adverse event follow-up

The following steps will be performed to document and follow-up on all Aes:

A. All AE/SAE's will be recorded in the participant chart with start and stop dates.
B. The study physician may perform additional evaluations, visits, phone communication, repeat blood testing, or refer to the emergency room or the participant healthcare provider for medical care if necessary.
C. If the AE results in intervention discontinuation and/or study withdrawal, the participant will be followed for 2 months post-withdrawal with a monthly phone call.
D. They will also be invited to perform the final visit evaluation including all study procedures planned for that visit if the participant has been in the study more than 5-months post baseline evaluation.
E. Follow-up on reported symptoms that may be due to hypotension (dizziness, weakness, near syncope, or syncope) or report that their blood pressure is less than 100/40 mm Hg.
    1. Study physician will review the incident and determine if the subject is safe to continue in the study.
    2. Physician may discontinue the study medication immediately, ask the participant to present to the study center for a safety evaluation, and/or monitor their blood pressure.
    3. Participant may continue in the study if blood pressure is >100/40 mm Hg or they report resolution of their symptoms.

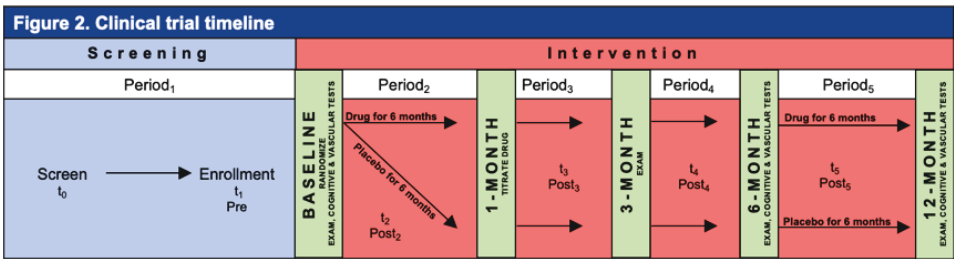
8.3.3 Incidental findings

Incidental findings (lab tor vascular testing) identified during the conduct of the study that do not require immediate medical care will be reported to the subject with instructions to seek medical care from the participant's healthcare provider. Those with incidental findings requiring urgent care will be referred to an emergency room in a facility of their choosing.

9. Study Timelines*

9.1. Study timeline

Each person that is enrolled in the study will participate for 12-months. There are six study encounters/visits: one screening visit and five evaluation visits (**Figure 2**). An enrollment period of 18 months to achieve the study sample size is anticipated. The estimated date of study completion is March 2025.



9.2. Study visit windows

The screening and baseline visit may occur at the same encounter/day or on separate encounters/days to be separated by no more than 60 days. The timing windows for the baseline and subsequent study visits are shown in **Table 2**.

Table 2. Study visit windows	
1-month	+/- 2 weeks of scheduled visit
3-month	+/- 4 weeks of scheduled visit
6-month	+/- 4 weeks of scheduled visit
12-month	+/- 4 weeks of scheduled visit

10. Inclusion and Exclusion Criteria*

10.1. **Inclusion criteria**

A. Signed and dated informed consent form.

B. Willingness to comply with all study procedures and availability for the duration of the study.

C. Self-identified AA (male or female) aged ≥ 50 years.

D. Blood pressure: normotensive and hypertensive participants will be eligible.

1. Normotensive criteria: BP 110-140/60-90 mmHg and not receiving any treatment for hypertension.
2. Hypertensive criteria: controlled hypertensives are eligible and is defined as a BP within the normotensive range of 110-140/60-90 mmHg. The study team will review medications for hypertension. Persons currently treated for hypertension with a RAAS modulator are eligible if they are willing to taper off the medication. To facilitate the taper, a study investigator will contact their primary care provider about tapering the RAAS modulator and subsequent blood pressure management. We will taper RAAS modulating medications in a stepwise manner using a seven-day washout period. In addition to blood pressure checks at study visits and study investigator discussions with primary care providers, participants will be instructed to also follow up with their primary care provider regarding changes to their antihypertensive medications. Blood pressure, safety labs, and potential adverse events are monitored during study visits (section 6.4) for all participants.
3. If the normotensive or controlled hypertensive criteria are not clear for any participant, then the study physicians will discuss and review their physical examination, medical history, and clinical data for study inclusion or exclusion.

F. MCI defined as:

1. Abnormal memory function per the Logical Memory subscale (Delayed Paragraph Recall, Paragraph A only) from the Wechsler Memory Scale-Revised (the maximum score is 25):
    a. Score <11 for 16 or more years of education
    b. Score <9 for 8-15 years of education
    c. Score <6 for <7 years of education
2. MoCA score 13-25
3. Preserved general functional performance per FAQ ≤ 9

If the diagnosis of MCI is not clear, then a review of the participant's cognitive and non-cognitive data will be performed by the study physicians and Neuropsychologist. The diagnosis of MCI will then be based on the clinical judgment of the study physicians and Neuropsychologist.

10.2. **Exclusion criteria**

Potential participants who meet any of the following criteria will be excluded:

A. History of stroke in the previous three years.

B. MoCA < 13 or the inability to follow instructions to perform cognitive testing.

C. Inability to participate in the study procedures: cognitive tests (communication limitations from language or other factors).

D. Active medical problem/symptom that interferes with the study procedures, e.g., chest pain, shortness of breath, or abdominal pain.

E. Deemed ineligible by study investigators due to safety issues or non-compliance.

F. History of the inability to tolerate treatment with an aldosterone antagonist.

G. Hypertension

    a. Uncontrolled defined as a BP > 140/90 mmHg

b. Current therapy with RAAS modulating medications, i.e., angiotensin II receptor blocker (ARB), angiotensin converting enzyme inhibitor (ACEi), renin inhibitor, or aldosterone antagonist such as eplerenone (Inspra) due to the risk of hyperkalemia and the interference with intrinsic activity of the RAAS in the body.

H. Renal disease (baseline creatinine > 2.0 mg/dL)
I. Hyperkalemia (K^+ > 5.5 mEq/dL)
J. Other neurological or psychiatric conditions that could impact cognition, e.g., Parkinson's disease and related movement disorders, multiple sclerosis, epilepsy, or schizophrenia, untreated major depressive disorder, and other psychotic disorders.
K. Abnormal thyroid stimulating hormone (TSH) (>10 mU/L) or vitamin B12 (<250 pg/mL) as they can impact cognitive function. If the potential participant's TSH and/or vitamin B12 levels are abnormal, then the PI may enroll that person if it is thought the lab abnormalities are unrelated and do not affect cognitive function.
L. Active medical or psychiatric disease, which in the judgment of the study investigators affects participant safety or the scientific integrity of the study.
M. Uncontrolled congestive heart failure reflected by poor exercise tolerance and shortness of breath.
N. Women of childbearing potential, i.e., non-menopausal.
O. When potential participants are not capable of providing informed consent due to their cognitive impairment and a study partner/LAR is not available to sign consent forms on their behalf.

10.3. Inclusion of women and minorities

Our recruitment efforts will focus on older self-identified AA individuals of both sexes to aim for equal representation of men and women in our sample. We will approach recruitment of minorities using known, effective techniques, thus meeting our desired sample size while maintaining our ability to study minority groups. The techniques include faith-based institutions where recruitment in minority population has been successful and screenings at minority health fares. Recruitment and retention strategies are further described in section 13.0.

11. Vulnerable Populations* N/A ☐

11.1. Inclusion of children

This research is in older adult AA with MCI/early dementia and will not include children.

11.2. Cognitively impaired adults

This research involves cognitively impaired adults thus there may be situations wherein participants, due to their cognitive impairment, may not understand the study procedures and hence will not be capable of providing informed consent. A study partner is required for all participants with MCI or those who are unable to answer the capacity assessment questions correctly (UBACC score <14.5). These persons may still qualify for the study but will need to have a surrogate and a study partner. Older AA with MCI/early dementia, based on standardized testing are targeted because this population may be more responsive to interventions preventing further cognitive decline. Participants enrolled in the study will be closely monitored for adverse events at visits and telephonic follow-up. An investigator may terminate participation in the study if any medical condition, event, or situation occurs such that continued participation would not be in the best interest of the subject. Participants are also free to withdraw from the study at any time.

12. Local Number of Participants

12.1. We aim to accrue 30 participants from the local, metropolitan Atlanta area.

13. Recruitment Methods

13.1. **Recruitment and retention strategies**

Potential participants will be identified through community activities, health fairs, advertisements and mail out announcements in periodicals and local newspapers. Recruitment will also occur during BP screening events in community barbershops and beauty salons, where information about the study will be provided. The GADRC team has previously used these venues for the recruitment of AA into clinical trials with significant success. Additional recruitment venues include the GADRC Clinical Core registry of research participants and screen-fail participants for other research studies at Emory University. Local physicians (primary care or specialty) will be informed of the study, its requirements, and provided with information about referrals. In addition, flyers for the study will be posted at outpatient areas in the following facilities: Emory University Hospital, Emory Clinic, and Grady Memorial Hospital.

13.2. **Telephone Screening**

A telephone screening form is utilized to assess the eligibility of potential study participants prior to scheduling a screening visit. A study team member will first contact potential participants to discuss the study and confirm their interest. If interested in study participation, the telephone screening form is reviewed. No identifiable health information is recorded during this phone interview.

13.3. **Study advertisement**

A one-page flyer, newspaper advertisement and tri-fold brochure with information about the purpose of the study, target population, and study team contact information will be used for participant recruitment. Copies of these documents are submitted with the IRB application.

13.4. **Participant compensation**

Subjects will receive a payment for their participation in this project in the amount of $150 if they complete all portions of the study (screening and five visits). Payments will be provided in the form of gift cards:

A. $10 for the Screening visit
B. $50 for the Baseline visit
C. $10 for the 1-month visit
D. $10 for the 3-month visit
E. $20 for the 6-month visit
F. $50 for the 12-month visit

Subjects may also derive a sense of satisfaction from contributing to an increase in overall knowledge about vascular disease and the brain.

14. Withdrawal of Participants*

14.1. **Withdrawal reason**

Subjects are free to withdraw from the study at any time. An investigator may also terminate a study subject's participation in the study if:

A. Any medical condition, event, or situation occurs such that continued participation in the study would not be in the best interest of the subject.
B. There is a lack of adherence to study treatment or visit schedules.

14.2. **Handling of participant withdrawal**

Every effort will be made to maintain every participant in the study until all visits are completed. In the event a participant withdraws, they will be asked to have a "withdrawal/termination" visit and followed for 2-months post-withdrawal with a monthly phone call.

14.3. **Premature termination or study suspension**

This study may be suspended or prematurely terminated due to, but not limited to, determination of unexpected, significant, or unacceptable risk to subjects. If the study is prematurely terminated or suspended, the PI will promptly inform the IRB and will provide the reason(s) for suspension or termination.

15. Risks to Participants*

15.1. **Potential study participation risks**

Some of the potential risks for study participation are minor, such as boredom or loss of time. Other risks from participation are more serious and include adverse effects from the drug, spironolactone, such as confusion and low blood pressure. A full list of expected risks from the study procedures and interventions is described below.

15.2. **Study procedure risks**

Cognitive testing assessment may be accompanied by anxiety, frustration, and overall fatigue. BP measurement and the assessment of vascular stiffness requires the attachment and removal of a BP cuff for venous occlusion and placement of an ultrasound probe on the neck, which may cause mild discomfort.

Risks associated with the LP include pain, bleeding, infection, and headaches. Topical anesthesia is applied to the skin around the LP site to minimize discomfort. If the participant becomes distressed at any point during the procedure, the procedure will be terminated. The risk for excess bleeding is evaluated by history (e.g., bleeding diathesis and use of therapeutic anticoagulation). The LP will not be performed if there is any concern for an increased bleeding risk. Post-LP headache has been reported to occur at various rates. For this study, 24-gauge Sprotte atraumatic spinal needles are used for which the incidence of post-LP headache has been less than 5%.[66] The risk of headaches is higher in younger individuals and is less in those with MCI/AD compared to other populations.[66] Each participant will be counseled on the signs and symptoms of post-LP headaches and given information on management (acetaminophen, caffeine, rest). Bacterial meningitis and a CSF leak are very rare LP complications. A blood patch is rarely needed to heal CSF leaks. All subjects will be instructed to promptly contact the study team if the headache persists for more than three days. If the study physician determines that a blood patch is required, the cost will be billable to the grant.

15.3. **Drug risks**

Participants may experience adverse effects from spironolactone of which the most common is gynecomastia, or enlargement of a man's breasts (9%). *The frequency of the less common adverse*

reactions of spironolactone has not been defined by the drug manufacturer. These have been reported within each body system in order of decreasing severity:

A. Digestive: gastric bleeding, ulceration, gastritis, diarrhea and cramping, nausea, vomiting.
B. Reproductive: decreased libido, inability to achieve or maintain erection, irregular menses or amenorrhea, postmenopausal bleeding, breast, and nipple pain.
C. Hematologic: leukopenia (including agranulocytosis) and thrombocytopenia.
D. Hypersensitivity: fever, urticaria, maculopapular or erythematous cutaneous eruptions, anaphylactic reactions, and vasculitis.
E. Metabolism: hyperkalemia (K^+>5.5meq/dL), electrolyte disturbances, hyponatremia (Na^+< 135 mEq/dL), and hypovolemia.
F. Musculoskeletal: leg cramps.
G. Nervous system/psychiatric: lethargy, mental confusion, ataxia, dizziness, headache, and drowsiness.
H. Hepatobiliary: a very few cases of mixed cholestatic/hepatocellular toxicity, with one reported fatality, have been reported with spironolactone administration.
I. Renal: renal dysfunction including renal failure (serum creatinine > 2.5 baseline value).
J. Skin: Stevens-Johnson Syndrome (SJS), toxic epidermal necrolysis (TEN), drug rash with eosinophilia and systemic symptoms (DRESS), alopecia, and pruritis.

16.Potential Benefits to Participants*

16.1. **Potential benefits**

This study is not designed to directly benefit participants. It is possible that participants can experience an improvement in cognition, but they may not, and they could also become worse. The goal of the study is to learn more about the safe administration and tolerability of spironolactone in older AA adults with impaired cognition.

17. Data Management* and Confidentiality

17.1. **Statistical considerations**

17.1.1 Study design

The study is a 2-arm double-blind randomized clinical trial. The statistical analyses will follow the intention-to treat (ITT) approach and pre-protocol analyses will be performed.

17.1.2 Randomization

The study blinded biostatistician will provide oversight of randomization fidelity and blinding. Randomization will be stratified by **spironolactone** vs. **placebo** (yes vs. no) using a computerized random number generator (SAS, V9.4) so that participants with and without a diagnosis of hypertension are represented in both groups. Only the pharmacy will have access to the randomization lists. As the pharmacist is notified that an individual subject is eligible for randomization, the pharmacist prepares the appropriate blinded study product. The pharmacist will enter the subject ID, date of product randomization, and the unique allocation sequence number on a web-based data form.

17.1.3. Blinding

The study team and biostatisticians will remain blinded to the treatment while the study is open. Study participants will remain blinded to the treatment during their participation in the study. At the end of a participant's study participation, Emory's Investigational Drug Service (IDS) pharmacy

will provide the study team with letters to the participant and the participant's primary care physician (PCP) which disclose the participant's study group assignment. These letters will be provided to the study team in sealed envelopes. Once the participant completes the study, they will receive their letter in a sealed envelope and their PCP will also receive this disclosure by mail. The study team takes measures to ensure participants do not unintentionally disclose their study group assignment by reminding them of the importance of the study team remaining blinded to their study group assignment during their initial consenting into the study; and this reminder is repeated to them during their final study visit. Study team members may also periodically remind participants of the importance of the study team remain blinded should this question come up during an individual's participation in the study. Participants may also be unblinded due to safety considerations. The study team works with the Emory pharmacy to facilitate safety related disclosures, should there be a need.

The designated study biostatistician receives blinding information during the data analysis phase of the study. All other study team members remain blinded until after completion of data analysis to minimize bias of study results.

17.1.4 Data analyses

The baseline characteristics of the two groups including demographic, social, and cognitive measures will be compared to assess randomization fidelity. Dependent variables include cognitive test scores and vascular assessments (PWV/PWA) at baseline and percent change at 12-months. Independent variables include group allocation (spironolactone vs. placebo). Analyses will account for variables that are recognized confounders in evaluating cognitive performance such as age, sex, BMI, and education level. The study measures are presented in **Table 1**. We will compare the change in outcomes over the study period between the two groups with mixed models.

17.1.5 analyses

We design our power analysis to detect the difference in change (slopes) between spironolactone and placebo in outcomes: AE counts, attrition rates, and cognitive test scores. We use $\alpha = 0.05$ and $\beta = 0.80$ to estimate differences after 12-months. A sample of 24 people will have an effect size difference of 1.20. Accounting for 20% dropout rate by 12-months we aim to enroll six extra subjects for a total sample of 30. A sample of 30 people will have an effect size difference of 1.06. To meet our target sample of 30 people, we expect to screen at least 36 people, which accounts for a 20% screen failure rate.

17.2. **Data management**

17.2.1 Media

Data collected during interviews and exams will be documented on trial-specific paper-based forms. Vascular assessment and CSF data will be saved in digital formats on a HIPAA-compliant server.

17.2.2 Management

Once a subject is enrolled into the study, he/she will be assigned a unique identifier number and be referred by initials and the study number only. Only research team members will have access to the files. Data will be entered on a web-based secure trial data system; the trial database will include all variables and an electronic data audit of data edits (who, when, and why). A data query report (including missing, out of range, and logic checks) will be generated by the trial statistician. The investigators will keep subjects' medical records private as far as the law allows. The IRB and officials

of the sponsor/funding agency will have access to these records as needed within legal guidelines. If study results are published in journals or presented at meetings, we will not use the participant's names.

18. Provisions to Monitor the Data to Ensure the Safety of Participants*

18.1. **Quality assurance**

18.1.1 Quality training
Research personnel will be trained by the PI and study physicians. The process of training on data forms completion, neuropsychological assessment, and subject evaluation will be documented in a training log for each study personnel.

18.1.2 Quality control
To assess protocol compliance and data collection quality, the study physicians along with another investigator will randomly and at intervals review collected data. In addition, an assessment of personnel competencies in obtaining data will be performed.

18.1.3 Protocol deviations
Every attempt will be exercised to maintain compliance with the approved study protocol. In the event a deviation is noted, the Emory IRB will be notified as required by IRB Policies & Procedures if the deviation affects the rights or welfare of subjects, safety of subjects, willingness of subjects to continue with study participation, or the integrity of the research data. The PI or designated personnel will investigate the setting, reasons, and potential remedies that need to be instituted to rectify the deviation and prevent similar future occurrences.

18.2. **Data safety monitoring plan**
18.2.1 Participant safety

18.2.1.1 Adherence and Monitoring Statement
The Data Safety and Monitoring Plan (DSMP) outlined below will adhere to the protocol approved by the institutional review board (IRB). Unless a waiver of consent documentation has been approved by the local institutional IRB, an IRB-approved written informed consent will be obtained from each subject at entry into the study; elements of informed consent will include: (a) having the subject and/or their LAR review the study consent form; (b) having the investigator(s) or study staff meet with the subject and/or guardian/proxy to review the consent, confirm understanding, and answer any questions; and (c) having the consent signed once the investigator(s) or study staff are convinced that the protocol is understood and that there is agreement to participate. A witness signature to the consent will be obtained if required by the study sponsor or the institutional IRB. A copy of the signed consent form will be provided to the subject and/or their legally authorized representative (LAR). Documentation of the informed consent process will be made in the research and/or medical records as appropriate.

An appointed designee will review a 10% sample of all data collection forms once per quarter for completeness and accuracy of the data in comparison to source documents, as well as for compliance with the protocol. Any data inconsistencies will generally be resolved by the study team within 2-weeks. The study team will review this protocol on a continuing basis for subject safety and protocol deviations or study noncompliance. Reportable Events will be communicated to the IRB as

required. The results of the review will also be included in annual progress reports submitted to the IRB or any other regulatory agency or sponsor, as required.

18.2.1.2 Participant monitoring
This will be performed by the PI, co-PI or co-investigators, and research coordinator(s).

18.2.1.3 Participant safety data examination and monitoring procedures
During the study, any problem reported by participants will be recorded in their chart (adverse event log), along with start and stop dates. The event may be related to their research participation, or the drug used in the study (drug reaction). All *adverse events* (AEs) will be rated on seriousness, severity, if related to the research, and if anticipated or unanticipated.

A serious adverse event (SAE) is defined using the Code of Federal Regulations Title 21 definition as any event that results in any of the following outcomes: death, a life-threatening adverse event, inpatient hospitalization, or prolongation of existing hospitalization, a persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions, or a congenital anomaly/birth defect.

The *severity* of an adverse event will be rated as mild, moderate, severe, life-threatening, or death. All adverse events will be judged regarding their relationship to the research participation or the study drug as related, possibly related, probably related, unrelated, or unknown.

Based on the Emory IRB definition, an *unanticipated event* is an event that is "unexpected, not described in the study documents" or if described in the study documents, is "presenting with increased severity, duration or frequency" than what is documented in the protocol, informed consent, or investigator's brochure.

18.2.1.4 Potential ("anticipated") adverse events

Safety parameters related to the use of spironolactone
The proposed study will utilize spironolactone, a drug approved by the Food and Drug Administration for the treatment of primary hyperaldosteronism; edema in persons with congestive heart failure, cirrhosis, and nephrotic syndrome; essential hypertension; hypokalemia; and severe heart failure. It is well tolerated by young and elderly individuals and has been used extensively in clinical practice. Risks of the study drug are described in detail in section 15.3, Drug Risks.

Safety parameters related to the conduct of study procedures
Neuropsychological assessment may be accompanied by anxiety, frustration, and overall fatigue. The attachment and removal of a BP cuff for venous occlusion and ultrasound probe on the neck may cause mild discomfort. Vascular ultrasound to measure PWV/PWA is safe and associated with only mildly uncomfortable due to tightness in the arm when the cuff is inflated. We will use ultrasound to painlessly bounce sound beams off the carotid artery using a pencil-like sensor.

18.2.1.5 Plan for detecting and minimizing subject risk
At each encounter, standardized adverse event (AE) monitoring forms will be completed. At any point during the study period an AE is identified, it will be recorded in the participant's research record along with the time/place of the event. The PI/study physician will be immediately notified, and the participant evaluated either by phone (if they are not on site) or face-to-face (if they are on site). Depending on the physician's assessment, further evaluation may be performed such as additional lab work, exam, or referral to an emergency department. Details about the safety parameters will be recorded and entered in a database. Comparisons between the two groups will be conducted to assess overall and between-group differences.

18.2.1.6 Reporting adverse events and death
All AEs will be assessed for the need for prompt reporting. We follow the guidelines set forth by the Emory IRB for AE reporting. Any event that fulfills criteria such as being unanticipated (as defined in section 18.2.1.3), probably or possibly related to study participation (drug effect or because of a study procedure) and involving risk to participants or others is considered an unanticipated problem and will be reported to the Emory IRB within 10 days. If an event could be explained by an underlying medical condition, then it will not considered related and not reportable. Any SAE as defined in section 18.2.1.3 that does not fit all the criteria of being an unanticipated problem will be reported during a continuing review. Deaths of participants during the study will be promptly reported to the IRB if related to study participation and reported during continuing review if they are not related to the study.

18.2.1.7 Protection against study risks

Consenting procedure
A signed consent form will be obtained from each participant or their LAR. A single informed consent form will describe both the screening and study procedures. The consent form will describe the purpose of the study, the procedures to be followed, and the risks and benefits of participation. A copy will be given to each participant and this fact will be documented in the participant's study record.

Plans for transmission of temporary or permanent suspension actions
Any actions that mandate temporary or permanent suspension of study will be transmitted to the supervising institutional IRB, and if appropriate, to the study sponsor, FDA, and the National Institutes of Health.

Plans for protecting subject confidentiality
All information and materials that are obtained for research purposes only will be kept in strict confidence. Confidentiality will be assured by use of subject codes rather than personal identifiers. The study database will be secured, and information will only be entered using subject identifier codes rather than personal identifiers. Electronic communication will involve only coded, unidentifiable information.

Plans for assuring data accuracy and protocol human safety compliance
Research personnel will be trained on data collection and recording. The process of training on data forms completion, neuropsychological assessment, and vital sign evaluation will be documented in a training log for each study personnel. To assess the quality of data collected the PI along with another investigator will randomly and on intervals review data obtained. In addition, quarterly assessment of personnel competencies in obtaining data (e.g., BP checks, neuropsychological assessments) will be performed.

The above detailed plans should assure data accuracy and protocol human safety compliance for this study. These include computerized database management, IRB oversight and communication. This plan, together with proposed oversight by IRB, should be sufficient for a study that does not require a national DSMB. Additional plans for assuring data accuracy and protocol human safety compliance if the study is complex, multisite, or of significant risk will involve a DSMB.

18.3. **Interim analyses**
Due to the sample size of this study no interim analysis is planned.

18.4. **Data and safety monitoring board**
DSMB meetings will take place annually, and a summary from the meeting will be included with the continuing review submission to the IRB.

Names and affiliations of members:

Whitney L. Wharton, PhD
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18.5. **Study discontinuation**

The study may be discontinued at any time by the investigators, the Emory IRB, the OHRP or other government agencies as part of their duties to ensure that research participants are protected.

19. Provisions to Protect the Privacy Interests of Participants

19.1. **Participant confidentiality**

Only the investigators will have access to information about a particular subject. The subject's primary care physician will only be notified if the subject agrees. To maintain confidentiality, subject data will be referenced by number and stored in locked computer files and cabinets. Identifying information about a subject will not be used during the discussion, presentation, or publication of any research data. Only research team members will have access to the files. Data recorded and stored on the computer will be backed up to a disc and stored with paper files. Participants will not be given any results of the research procedures unless there is a medical necessity. This will be clearly stated in the informed consent process.

19.2. Describe what steps you will take to make the participants feel at ease with the research situation in terms of the questions being asked and the procedures being performed. "At ease" does not refer to physical discomfort, but the sense of intrusiveness a participant might experience in response to questions, examinations, and procedures.

19.3. Indicate how the research team is permitted to access any sources of information about the participants.

20. Economic Burden to Participants

20.1. **Participant costs**

There will be no cost for participating in this study, other than basic expenses like transportation. You will not be charged for any of the research activities.

21. Consent Process

21.1. **Institutional Review Board**

This protocol and the informed consent document and any subsequent modifications will be submitted and reviewed by the Emory IRB.

21.2. **Informed consent/assent and HIPAA authorization**

A single informed consent form will describe both the screening and study procedures. The consent form will describe:

A. Purpose of the study
B. Procedures to be followed
C. Risks and benefits of participation.

A copy will be given to each participant and this fact will be documented in their study record and medical record as per Emory University policy. The ability to understand the research study details/contents of the informed consent will be assessed by asking the subject a set of study-specific questions. A validated brief instrument for decisional capacity assessment will be administered. The UBACC instrument is a 10 item questionnaire that asks the participant about key areas of the informed consent.[68] A score greater than 14.5 correlates with a core of greater than 16 on the MacArthur Competency Assessment Tool for Clinical Research (MacCAT-CR).[68] We will use a score less than 14.5 to require a LAR sign/cosign the consent along with the assent of the participant. A signed consent (combined consent/HIPPA) form will be obtained from each participant or their LAR. Any subject who expresses a desire to discontinue participation in the study after providing consent will be removed from the study.

21.3. **Study Partner**

Because of the nature of this research, we may encounter situations where the potential participant, due to impaired cognition, may not understand the procedures of the study and hence are not capable of providing informed consent. A study partner is required for all participants with MCI or those who are unable to answer the capacity assessment questions correctly (UBACC score <14.5). These persons may still qualify for the study but will need to have an LAR and a study partner. A study partner may be:

A. Participant's legal guardian (if one exists according to applicable GA law and consistent with the intent of US 45 C.F.R. 46 and 21 C.F.R. 50 & 56)
B. Next of kin
C. Family member or a friend.

The study partner will also need to consent to the study. If either party refuses, then the participant will not be enrolled. The criteria for a study partner are:

A. Ability to provide information about the participant
B. Willing to provide information about the participant
C. Have contact with the participant for at least once a month (in person or telephone).

The definition of the surrogate will be consistent with the intent of the Common Rule (45 CFR 46, Subpart A). The following are, in order, possible surrogates:

A. The person's agent designated by an advance health care directive.
B. The conservator or guardian of the person having the authority to make healthcare decisions for the person.
C. The spouse of the person.
D. The domestic partner of the person as defined in Section 297 of the Family Code
E. An adult son or daughter of the person
F. A custodial parent of the person
G. Any adult brother or sister of the person
H. Any adult grandchild of the person
I. An available adult relative with the closest degree of kinship to the person

When there are two or more available persons who are in different orders of priority pursuant to subdivision (c), refusal to consent by a person who is a higher priority surrogate shall not be superseded by the consent of a person who is a lower priority surrogate. That surrogate will need to consent to the study. If either party refuses, we do not enroll the subject. To ensure both the participant and the proxy understand the study protocol, we will ask subjects or legal next of kin to explain in their own words the nature of the study and the procedures involved.

The LAR can be the study partner if there is contact at least once a month. In some instances, such as the lack of adequate contact by the LAR or their inability to complete the tasks of the study partner, the study partner may be a different individual as long as they are willing to provide information about the participant and have contact with the participant for at least once a month (in person or telephone). The study partner will sign a separate *Study Partner Informed Consent* document. If they are not available in person, the study staff will contact the study partner to obtain needed information about the subject's cognitive symptoms. In that case, a verbal consent will be obtained.

22. Process to Document Consent in Writing

22.1. Informed consent documentation

Consent for study participation will be documented in writing. Prior to making a decision about study participation, potential participants will be required to read the form, or have it read to them, listen to an explanation of the study by the investigators or staff, and ask questions about any aspect of the study that is not clear. Potential participants will have the opportunity to take a copy of the consent form to contemplate their participation as well as discuss it with family or friends. The consent document is attached to the IRB application for review and approval.

23. Setting

23.1. Facilities

The GADRC Clinical Research Unit (CRU) occupies newly renovated space on the 2nd floor (25,000 square feet of combined clinic, office, and laboratory space) in 6 Executive Park (EP6). An additional 2,200 square feet of laboratory space for biospecimen processing, storage, and analysis is located on the 1st floor of EP6. The CRU space hosts participant visits for GADRC clinical trials and observational studies. The renovation represents a major expansion of the Brian Health Center (BHC) housed in EP6 and 12 Executive Park (EP12). EP6 is adjacent to EP12, which houses administrative offices and clinics for the Emory Brain Health Center, including the Cognitive Neurology Clinic and the Integrated Memory Care Clinic (IMCC).

23.2. Office

Offices are located on the 2nd floor of the BHC and CRU. Office space in the CRU consists of 7,000 square feet and includes offices and cubicles for affiliated faculty and staff with the GADRC. Seventy-two cubicle workstations support the teams overseeing daily research operations with 12 additional touchdown spaces available. Stations have power and high-speed internet ports. Leadership and administrators occupy the 16 private offices in the BHC and CRU space. One 8-person room and two 15-person capacity conference rooms can be reserved by Emory employees within the CRU 2nd floor office space. Each conference room is equipped with videoconference technology, integrated phone systems, and dedicated computers with projectors or televisions for screen casting.

Additionally, conference rooms are also available in the BHC space at EP12 with similar capabilities and can be reserved as needed.

23.3. **Research**

Research visits and procedures for studies conducted at the GADRC occur in dedicated research space (25,000 total square feet: exam, laboratory, and participant space plus office space) located on the 2nd floor of EP6. The CRU has 20 flex exam rooms, nine testing rooms, three cardiovascular rooms, two retinal imaging rooms, seven infusion bays, and storage space, a lab for biofluid handling, and faculty and staff offices, and a large participant reception area (approximately 6,000 square feet).

24. Resources Available

24.1. **Laboratory**

The CRU Laboratory occupies a 2,200 square feet wet lab space for specimen processing, long-term storage, and analysis. This space is located on the 1st floor of EP6 and is equipped with bench space for six individuals and all necessary equipment for the proposed work, including centrifuges, balances, microplate readers, spectrophotometers, refrigerators, hot plates, stirrers, and -20°C freezer. A dedicated freezer room will house ten -80°C freezers and two large liquid nitrogen tanks for on-site long-term biospecimen storage. Additional key CRU Laboratory equipment includes a Cobas e601 analyzer for immunoassays, Tecan liquid handler for HTP handling of liquids and more accurate aliquoting, and two mass spectrometers including a fully equipped state-of-the-art Orbitrap Fusion Lumos Tribrid mass spectrometry system.

24.2. **Computer**

All faculty and staff offices are equipped with computers connected to the Emory network and local printers. Faculty and staff have access to Emory site-licensed software (e.g., Microsoft Office), statistical packages (e.g., SAS, SPSS), and bibliographic software (e.g., Endnote). The Emory network allows access to multiple on-line databases including Medline, Embase, and PsychInfo. GADRC teams use Salesforce CRM, REDCap, and Emory enterprise-wide systems such as Nautilus LIMS and the Emory Research Subject Registry (ERSR) that have been customized for data collection, storage, and retrieval. Dedicated terminals, label printers, and bar code scanners are available for data entry and sample tracking. The application associated SQL Server, MySQL, and Oracle databases utilize GUIDs to connect the information across platforms and in the Data Mart. The GUIDs are independent of patient identifiers and meet HIPAA privacy standards. The applications and databases reside on servers in a secured zone behind the Emory University firewall that requires network and application logins to access data granted at the user role level. The firewall and servers are maintained by Emory University and comply with security policies and regulations.

Database servers are physically located in Emory's state of the art enterprise server room with continuous environmental monitoring, battery and generator backup, and advanced electronic fire suppression with keycard physical security measures. Domain controlled usernames and passwords allow for appropriate user specific application-level security privileges including project specificity, write restrictions, and system administration restrictions. The database incorporates full transactional logging allowing for point in time data restoration as well as full data audit trails. Nightly backups are made to tape and are stored off site.

24.3. **Clinical**

GADRC studies are conducted in dedicated clinical research space (25,000 total square feet: exam, laboratory, and participant space plus office space) on the 2nd floor of EP6. The unit has 20 flex exam rooms, nine testing rooms, three cardiovascular rooms, two retinal imaging rooms, seven infusion bays, and storage space, a lab for biofluid handling, and faculty and staff offices, and a large participant reception area (approximately 6,000 square feet). The examination rooms are equipped with an ophthalmoscope, otoscope, and sphygmomanometer and are supplied for phlebotomy. The rooms are also equipped with tables to perform LP and other procedures. All exam and testing rooms are specially equipped with sound masking for uninterrupted cognitive testing and voice data collection.

24.4. **Equipment**

Vascular Equipment: The equipment to perform vascular assessments includes the Sphygmocor device (SphygmoCor®, Atcor Medical, Australia) and is housed on the 2nd floor of EP6 in the CRU. The Sphygmocor device is an automated, cuff-based system that assesses peripheral arterial stiffness as carotid-femoral artery PWV and radial pulse wave analysis PWA. The device is portable and easily setup inside a private examination room. It sequentially records high-quality pressure waveforms at peripheral pulse sites using a high-fidelity tonometer by which it assesses PWV and PWA.

25. Multi-Site Research when Emory is the Lead Site*N/A ☐

25.1. N/A

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