

Amyloid PET imaging in self-identified non-Hispanic Blacks from the Anti-Amyloid in Asymptomatic Alzheimers Disease (A4) Study

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Amyloid PET imaging in self-identified non-Hispanic Blacks from the Anti-Amyloid in Asymptomatic Alzheimer's Disease (A4) Study

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Abstract

Objective: To examine whether amyloid PET in CN individuals that were screened for the Anti-Amyloid in Asymptomatic AD (A4) study differed across self-identified, non-Hispanic White and Black (NHW and NHB) groups.

Methods: We examined 3689 NHW and 144 NHB that passed initial screening for the A4 study and underwent amyloid PET. The effect of race on amyloid PET was examined using logistic (dichotomous groups) and linear (continuous values) regression controlling for age, sex, and number of *APOE* $\epsilon 4$ and *APOE* $\epsilon 2$ alleles. Associations between amyloid and genetically determined ancestry (reflecting African, South Asian, East Asian, American, European populations) were tested within the NHB group. Potential interactions with *APOE* were assessed.

Results: NHB had lower rates of amyloid-positivity and lower continuous amyloid levels compared to NHW. This race effect on amyloid was strongest in the *APOE* $\epsilon 4$ group. Within NHB, those with a lower percentage of African ancestry had higher amyloid. A greater proportion of NHB did not pass initial screening compared to NHW, suggesting potential sources of bias related to race in the A4 PET data.

Conclusion: Reduced amyloid was observed in self-identified non-Hispanic Blacks that passed initial eligibility criteria for the A4 Study. This work stresses the importance of investigating AD biomarkers in ancestrally diverse samples as well as the need for careful consideration regarding study eligibility criteria in AD prevention trials.

Introduction

The ability to measure Alzheimer's disease (AD) biomarker abnormalities among older clinically normal (CN) individuals has accelerated attempts to identify at-risk individuals and implement clinical trials that focus on prevention ¹. Large cohort studies consistently report that amyloid+ CN are at elevated risk of cognitive decline ² and dementia ^{3,4}. Further, studies have shown that approximately 30% of CN in their 70's are amyloid-positive ⁵, highlighting that a large proportion of CN may be good candidates for intervention.

Despite progress understanding the impact of AD pathology among CN, cohort-based studies are largely comprised of non-Hispanic White (NHW) individuals and little is known regarding how AD biomarker findings translate to other populations. Greater representation of minority populations is needed, especially considering that self-identified non-Hispanic Black (NHB) are at greater risk of clinical AD^{6,7}. The mechanisms underlying this heightened risk of clinical AD in NHB remain unknown, but may be related to several risk factors that differ between NHW and NHB (genetics, vascular disease, social determinants of health etc.) ⁸⁻¹¹. Interestingly, the effect of *APOE4*, the most established genetic risk factor for AD^{12,13} as well as a robust predictor of abnormal amyloid in the preclinical stage of the disease^{14,15}, has been shown to have a weaker effect on AD risk ^{13,16-19} and cognitive decline ⁸ in NHB relative to NHW. Thus, despite an overall greater risk of dementia in NHB, it is possible that established risk factors of AD in NHW (such as *APOE4*) do not have a similar impact in non-European populations and require further investigation.

To this end, we examined the effect of NHW and NHB race on amyloid in CN individuals that were screened for the Anti-Amyloid in Asymptomatic AD (A4) study²⁰, and whether these effects were influenced by *APOE* and/or African ancestry.

Methods

Participants

The Anti-Amyloid Treatment in Asymptomatic Alzheimer's Disease (A4) Study is a Phase III clinical trial that enrolled CN older adults with evidence of brain amyloid as measured by ^{18}F -Florbetapir positron emission tomography (PET) ²⁰. The overall goal of this clinical trial is to understand whether intervention with an anti-amyloid antibody treatment (solanezumab) in amyloid+ CN will slow cognitive decline²¹. Participants age 65-85 were assessed to be CN, 25-30 on the Mini-Mental State Examination (MMSE), Global Clinical Dementia Rating (CDR) scale=0, and 6-18 on the Logical Memory II test. Additionally, enrollment into the clinical trial (into either the treatment or placebo arm) required amyloid-positivity using a hybrid qualitative/quantitative algorithm. Thus, a number of individuals were screened for the A4 clinical trial to determine eligibility for enrollment into treatment or placebo arms. The first round of screening was performed on 6763 individuals and involved neuropsychological testing to determine clinical status and medical assessment to exclude individuals with co-morbid conditions²⁰, with 4371 passing all initial screening and completing an amyloid PET scan. After the amyloid PET scan, amyloid+ CN were then randomized into treatment or placebo arms. The data presented herein incorporates all available screening data for individuals that self-identify as either NHW or NHB, enabling the examination of the full range of amyloid PET values among CN as well as potential selection biases introduced after the initial screening based on neuropsychological testing and medical assessment that precluded individuals from obtaining an amyloid PET scan (Data available from Dryad; S. Figure 1: doi:10.5061/dryad.7h44j0zsh).

Race self-identification was collected across these categories: American Indian/Alaskan Native; Asian; Native Hawaiian/Other Pacific Islander; Black/African American; White; or Unknown. For this study, we examined all participants that self-identified as non-Hispanic Black (NHB) or White (NHW). Participants that identified as more than one category were excluded

(resulting in 325 NHB and 5753 NHW). Those that continued to meet study criteria after completing all initial screening visits then underwent amyloid PET (153 NHB and 3818 NHW). We excluded individuals missing APOE or had the APOE2/4 genotype, resulting in a final sample of 144 NHB and 3689 NHW with amyloid PET data (Data available from Dryad; S. Figure 1: doi:10.5061/dryad.7h44j0zsh).

Amyloid PET

Amyloid PET data was acquired 50-70 minutes post-injection of 10 mCi of ^{18}F -Florbetapir. We examined mean global cortical standardized uptake value ratio (SUVR, whole cerebellar reference region). We used a data-driven gaussian mixture modeling approach ²² to classify participants into amyloid groups using a cut of ≥ 1.17 (since amyloid group assignment using the A4 criteria is not publicly available²⁰). We additionally examined global amyloid levels continuously.

Genotyping and Ancestry determination

All participants were genotyped on the Illumina Global Screening Array at Columbia University. No participants had autosome missingness exceeding 5%. SNPs reporting a call rate $>95\%$ and a minor allele frequency (MAF) $>1\%$ were considered for ancestry determination. Approximately 18,000 ancestry informative markers from the 1000 Genomes Consortium were used to determine individual ancestry using SNPweights v2.1^{23, 24}. This resulted in percentages for each participant that reflect ancestry across five super populations (Africans, Europeans, Americans, South Asians, and East Asians) with these five percentages summing to 100 percent for each participant. 28 NHB were missing genotyping data, resulting in a subset of 116 NHB for

the analysis with ancestry. 2915 of the 3689 NHW had genotyping data available for ancestry calculations.

Lifestyle Risk Factors and Medical Conditions

Lifestyle factors and medical conditions were available for the majority of individuals that received an amyloid PET scan, as well as a smaller subset of individuals that screen-failed (Data available from Dryad, S. Figure 1 and S. Table 1: doi:10.5061/dryad.7h44j0zsh). Notably, this data is missing for the majority of individuals that screen-failed due to low neuropsychological testing scores (Data available from Dryad, S. Figure 1: doi:10.5061/dryad.7h44j0zsh). Thus, examination of these variables was restricted to the subset of individuals that meet initial study criteria (neuropsychological and medical assessment) and completed the amyloid PET scan.

Statistical Analysis

Analyses were performed using R v3.4.1. Differences in demographics across race were examined with an analysis of variance (ANOVA) for continuous variables and Fisher exact tests for categorical variables. The effect of *APOE* in all analyses was examined by modeling the number of *APOE4* and *APOE2* alleles (0, 1, or 2). Given demographic differences across race, propensity score matching was used for causal inference with racial group (NHB vs. NHW) on age, sex, and education using the nearest neighbor matching method in the MatchIt package in R v4.9-3²⁵ (using a ratio of 2:1 for NHW to NHB). This method calculates a propensity score (ie. the probability that a NHW participant will have the same characteristics of a NHB participant, thus reducing the effects of confounding variables) estimated using logistic regression to create the matched NHW group.

Raw continuous amyloid SUVR was examined using linear models, whereas dichotomous amyloid-positivity group was examined with logistic regression. All models investigated the effect of self-ID race (NHB versus NHW), number of *APOE2* and *APOE4* alleles, age, and sex on amyloid outcomes. The interaction between race and *APOE4* and *APOE2* was also examined in separate regression models. To determine the impact of African ancestry on amyloid within the NHB group, we investigated the association between percentage of African ancestry and amyloid with linear regression. Post-hoc contrasts were run to further demonstrate potential interactions between race and *APOE* genotypes (ie. to demonstrate the effect of race across genotype, we contrasted NHB E4+ with NHW E4+, NHB E3/3 with NHW E3/3, and NHB E2+ with NHW E2+; to further understand the effect of *APOE2* within race, we contrasted NHB E2+ with NHB E3/3, and NHW E2+ with NHW E3/3). Finally, additional sensitivity analyses were run to examine potential effects between amyloid and lifestyle as well as self-reported medical conditions that differed by race (these lifestyle and medical conditions were included in models predicting amyloid to determine whether effects of race on amyloid remained significant).

Data Availability Statement

Data used in this article is available for download from the Laboratory of NeuroImaging (LONI; loni.usc.edu). All variables were extracted from spreadsheets posted on LONI, with the exception of ancestry measures that were processed from genotyping data posted on LONI.

Standard protocol approvals, registrations, and patient consents

All analyses were performed on data collected as part of the A4 study (NCT02008357). This study was approved by the Institutional Review Boards of all of the participating institutions. Informed written consent was obtained from all participants at each site.

Results

Participant Characteristics

The full NHW group had higher education and more males compared to NHB (Table 1). The distribution of *APOE* genotype by race was also significantly different ($\chi^2=19.69$, $p<0.0001$), such that there was a higher proportion of the *APOE2/3* genotype in NHB (21.5% in NHB compared to 10.3% in NHW). Given demographic differences, we additionally examined patterns of amyloid PET in a demographically matched group of 288 NHW individuals (Table 1).

Effect of Amyloid across Race

NHW groups were more likely to be amyloid+ relative to NHB (Figure 1A, Table 2A), as well as have greater continuous levels of amyloid (Figure 1B, Table 2B). NHW were ~2 times more likely to be amyloid-positive than NHB, and had ~0.05 SUVR units more on average than NHB. Similar results were observed when examining regional rather than global amyloid (Data available from Dryad, S. Table 2: doi:10.5061/dryad.7h44j0zsh), and after additionally controlling for MMSE (Data available from Dryad, S. Table 3: doi:10.5061/dryad.7h44j0zsh).

Race effect strongest in *APOE4* carriers

Linear models predicting continuous amyloid levels were repeated with an *APOE4* allele number x race interaction term, as well as an *APOE2* allele number x race term (in separate models). The interaction between *APOE4* and race was significant ($p=0.01$), whereas the *APOE2* and race term was near significant ($p=0.06$). Although *APOE3/4* and *APOE4/4* genotypes were associated with higher amyloid across both races (Figure 1), the difference between NHB and

NHW groups on continuous amyloid was greatest within the *APOE4+* group (*APOE3/4* and *APOE4/4* combined). This was confirmed with a significant post-hoc contrast between NHB and NHW *APOE4+* groups, revealing that *APOE4+* NHB had 0.094 SUVR units less than *APOE4+* NHW (Table 3). There was a marginal difference, although not significant, between NHB and NHW among the *APOE3/3* genotype (0.034 SUVR less in *APOE3/3* NHB), while no significant race effect was found among the *APOE2* group (*APOE2/2* and *APOE2/3* combined, Table 3).

Interestingly, *APOE2* did not appear to be protective within NHB (Figure 1), such that levels of amyloid were not significantly different between *APOE2/3* NHB compared to *APOE3/3* NHB (Table 3). In fact, values were qualitatively higher in *APOE2/3* NHB compared to *APOE3/3* NHB, though this increase was not significant. As expected, the *APOE2/3* group had significantly lower levels of amyloid than the *APOE3/3* group within NHW (Table 3).

Ancestry Estimation and effect on Amyloid

Genome-wide SNP array data to compute continuous ancestry was available for 2915/3689 NHW and 116/144 NHB. This subset analysis revealed that those who self-identified as NHW largely had European ancestry above 89% (mean=91%, SD=0.07; Figure 2A) whereas those who self-identified as NHB had a wide range of African ancestry that was largely admixed with European ancestry (mean=0.63, SD=0.14) (Figure 2B). There were two NHB outliers with African ancestry at 2.1% and 4.2% that were excluded from further analysis (resulting in N=114 NHB for this analysis).

Within NHB, higher African ancestry was associated with less amyloid and this effect was independent of number of *APOE4* alleles (% African ancestry: $\beta = -0.239$, SE= 0.103, $p=0.023$; *APOE4* allele count: $\beta = 0.083$, SE= 0.021, $p=0.0002$) (Figure 3). The interaction between ancestry and *APOE4* ($p=0.99$) or *APOE2* allele count ($p=0.45$) was not significant.

Higher exclusion rate in NHB

In order to undergo amyloid PET for the A4 study, participants first underwent an initial screening visit to determine eligibility based on having a clinical diagnosis of CN, be CDR=0, within cut off values for the MMSE (25 or above), Logical Memory delayed recall (between 18 and 6; thus both low and high scoring participants were excluded based on this test), as well as lack of co-morbid health conditions. Of all the participants that underwent initial screening (S. Figure 1), NHB were more likely to screen-fail at this stage relative to NHW (53% versus 33%; $\chi^2=54.9$, $df=1$, $p<0.0001$). We then compared the proportions across three categories of screen-failure criteria: (1) exclusion due to low testing scores, (2) high Logical Memory scores, or (3) other reasons (Data available from Dryad, S. Figure 1: doi:10.5061/dryad.7h44j0zsh). Among those that screen-failed during this initial stage, the reasons for exclusion across these three categories significantly differed by race ($\chi^2=31.5$, $df=2$, $p<0.0001$). Specifically, NHB were more likely to screen-fail due to low testing scores compared to NHW (51.1% versus 34.6%) and less likely to screen-fail due to high Logical Memory scores (1.1% versus 10.5%). Exclusion for other reasons was similar across race (47.7% versus 49.0%). Other possible exclusion criteria such as self-reported medical conditions were not collected on the majority of screen-fail participants, precluding further assessment of whether ineligibility due to medical reasons differed by race during initial screening (Data available from Dryad, S. Figure 1: doi:10.5061/dryad.7h44j0zsh).

Impact of lifestyle factors and self-reported medical conditions on amyloid

Among individuals that underwent amyloid PET, we examined whether there were differences between lifestyle factors and/or self-reported medical conditions by race (Data available from Dryad; S. Table 1, S. Figure 2: doi:10.5061/dryad.7h44j0zsh). Overall, NHB had

lower self-reported alcohol consumption (Data available from Dryad; S. Figure 2A: doi:10.5061/dryad.7h44j0zsh), greater cardiovascular risk factors (higher blood pressure, higher BMI, and higher self-reported cardiovascular conditions) (Data available from Dryad; S. Figure 2B-E: doi:10.5061/dryad.7h44j0zsh), and *lower* self-report of multiple medical conditions (specifically, Psychiatric, Dermatologic-Connective Tissue, Gastrointestinal, and Head, Eyes, Nose, and Throat) (Data available from Dryad; S. Figure 2F-I: doi:10.5061/dryad.7h44j0zsh). Further, summation across total self-reported medical conditions revealed that NHW had a greater total burden of self-reported conditions than NHB (Data available from Dryad; S. Figure 2J-K: doi:10.5061/dryad.7h44j0zsh).

Given these differences in cardiovascular risk factors (*higher* in NHB) and self-reported medical conditions (*lower* in NHB), we performed additional analyses to explore whether these factors may be associated with amyloid. There were no significant associations between these vascular and medical factors with amyloid, and the effect of race on amyloid remained significant (Data available from Dryad; S. Table 4-5: doi:10.5061/dryad.7h44j0zsh). Finally, we explored if the total number of endorsed medical conditions was associated with amyloid, given that the total burden was less in NHB (and may account for reduced amyloid in NHB). Interestingly, we did find that a higher number of medical conditions was associated with greater amyloid levels, but the effect of race on amyloid remained significant (Data available from Dryad; S. Table 6: doi:10.5061/dryad.7h44j0zsh).

Discussion

In a large sample of older CN screened for a secondary prevention trial, we found that self-identified NHB individuals had reduced levels of amyloid measured with PET compared to self-identified NHW individuals, an effect that was strongest within *APOE4* carriers. Within the

NHB group, higher levels of continuous African ancestry were associated with lower amyloid. Finally, rates of screen-failure were higher in NHB, and this at least was partially related to eligibility criteria for neuropsychological testing scores, suggesting potential selection bias (more NHB were excluded from receiving a PET scan). Overall, our results highlight the importance of understanding race specific factors and selection bias in studies of preclinical AD.

Our main result was that NHB CN had less amyloid as measured with PET than NHW CN. This effect was consistent regardless of whether amyloid was treated as a dichotomous or continuous variable, as well as whether the entire sample of NHW was used or a demographically matched NHW group. This effect also remained significant after inclusion of potential confounds related to vascular risk factors and medical conditions. Given that abnormal levels of amyloid in CN are associated with future memory decline and progression to clinical impairment ²⁶, our finding of reduced amyloid in NHB suggests other risk factors may contribute to AD dementia in NHB. These other risk factors in NHB may include co-morbid vascular risk factors, TDP43 pathology, and other age-related pathological accumulations that are known to influence the clinical expression of dementia ^{9, 27}. Along these lines, we found evidence of greater vascular risk factors in the NHB sample, consistent with an increased role of vascular disease in NHB. Further, social determinants of health, including increased exposure to stressful events throughout the lifespan, is increased in NHB and associated with cognitive decline in aging²⁸. However, given the severe paucity of AD biomarker studies specifically within NHB CN samples, it is simply unknown whether PET measures of AD pathology hold a similar predictive value regarding future progression to dementia in NHB compared to NHW CN cohorts. This represents a critical gap in knowledge, especially as the field of AD research shifts towards preventative strategies in hopes of targeting CN individuals with initial evidence of AD pathology.

To our knowledge, one study has compared amyloid PET across NHB and NHW within a CN sample from the Atherosclerosis Risk in Communities (ARIC) study ²⁹. In contrast to our study,

Gottesman et al²⁹ found that NHB had greater amyloid-positivity and continuous levels of amyloid than NHW in a sample of CN combined with patients with mild cognitive impairment (MCI). When divided by diagnosis, the effect remained significant within their sample of 37 NHB with MCI compared to 52 NHW with MCI, such the NHB MCI group had elevated amyloid compared to the NHW MCI group. However, when examining the group of 104 CN NHB compared to 136 CN NHW, there was no significant difference across race. A few recent studies have examined cerebrospinal fluid (CSF) levels of amyloid and tau proteins across race, however these have focused on the spectrum of AD (CN, MCI, and AD dementia) and have not specifically examined the pattern of these AD biomarkers specifically within the CN group³⁰⁻³². These studies generally report no difference in CSF amyloid levels by race, but have converged to reveal less CSF tau in NHB compared to NHW. It is possible that differences in CSF versus PET measures of amyloid may be relevant for understanding these different patterns across studies (e.g. soluble versus fibrillar amyloid), and to our knowledge have not been fully investigated with respect to race. Taken together, these studies highlight that race specific differences may exist in profiles of AD biomarkers, however, the inconsistencies underscore the importance of examining these effects in larger cohorts, and the need to explore factors that may drive these patterns, such as study selection criteria, disease stage, and biomarker measurement differences.

The *APOE* genotype is the strongest genetic risk factor for late onset AD and has consistently been associated with elevated AD risk in NHW populations. *APOE4* has been related to abnormal accumulation of the beta-amyloid protein, as well as to influence the rate of cognitive decline in early stages of dementia^{14, 15, 33-35}. Interestingly, although the *APOE4* allele has been shown to have a higher frequency in NHB relative to NHW, this genetic risk variant has a weaker effect on clinical AD dementia risk^{13, 16-18, 36} and cognitive decline⁸ in NHB relative to NHW. Our analysis revealed that the reduced effect of amyloid in NHB was strongest among *APOE4* carriers. This suggests that the lower effect of *APOE4* on AD risk in NHB may be mediated by

pathways related to amyloid accumulation. Interestingly, we also found that *APOE2* had a reduced protective effect in NHB, such that amyloid levels in the *APOE2* group were non-significantly higher than the *APOE3* group within NHB (as opposed to the expected reduction in amyloid levels in *APOE2* compared to *APOE3* in NHW). Our finding is consistent with work by Farrer et al that showed a lack of protective effect of *APOE2* on risk of clinical AD dementia in NHB¹³. Other genetic risk factors, such as *ABCA7*¹⁶ and *RBFOX1*³⁷, have been identified as having a larger effect on AD phenotypes in NHB compared to NHW. Overall, our findings are consistent with a broader literature suggesting that genetic risk factors of AD have varying effects across race.

Although we identified an association between higher African ancestry and reduced levels of amyloid, it is important to emphasize that race is a social construct that is typically based on visible characteristics and predominantly used in the United States³⁸. The current dataset is ill-equipped to examine how the social construct of race influences risk of AD and amyloid burden. Recent studies suggest that coming from a disadvantaged neighborhood is associated with increased odds for post-mortem AD neuropathology³⁹ and having more life-stressors has been associated with cognitive decline in NHB²⁸. It is becoming increasingly recognized that other pervasive factors are important to consider when examining race and dementia risk. Nevertheless, examination of genetic ancestry is an approach that allows quantification of genetic loci that have specific allele frequencies based on geographical origin⁴⁰. Previous studies have shown that NHB in the United States have admixed genetic African and European ancestry^{41, 42}. This is in stark contrast to those Americans of European descent, who have very little genetic contribution from non-European ancestral backgrounds. Thus, the continuum of African ancestry may be relevant when understanding differences in AD risk factors in NHB, given that multiple genetic factors related to AD vary across race^{13, 16-19, 36}. Along these lines, Nigerian Yoruban individuals have been shown to have a lower incidence rate of AD dementia than NHB that live

in the United States^{43, 44}. Our results are consistent with this, and suggest that within the NHB group prominently from the United States, higher levels of African ancestry were associated with lower levels of amyloid. Thus, it is possible that genetic differences, such as ancestral origin of *APOE4*⁴⁵, captured by ancestry measures may influence amyloid accumulation, and that in turn is protective against AD dementia. It is also possible that discrepant findings across studies of NHB on AD risk more broadly reflect different amounts of African ancestry between study populations.

In addition to finding overall lower levels of amyloid PET in NHB, we also found significant differences in screen-failure rates by race that precluded more NHB individuals from receiving an amyloid PET scan. Specifically, NHB were more likely to screen-fail than NHW before the stage of receiving an amyloid PET scan, and one reason for this exclusion was due to worse scores during neuropsychological testing (NHB were more likely to perform under the pre-defined cut offs for these tests). NHB often perform lower at baseline on many standardized neuropsychological tests used in clinical trials of AD, but this intercept effect does not translate to greater decline over time⁴⁶. The lower performance on neuropsychological tests is thought to represent socioeconomic and demographic issues such as limited educational opportunities⁴⁷. This pattern implies that baseline neuropsychological tests may not accurately capture true cognitive performance in NHB, and instead reflect biases in these tests and inadequate norming procedures⁴⁸. Such a selection bias related to neuropsychological screening within the A4 study could drive the apparent reductions in amyloid PET in NHB compared to NHW by excluding NHB participants that are more likely to be amyloid+ and creating a more “resilient” NHB group that was included in the PET dataset. Consistent with this interpretation, we did find that among those that underwent a PET scan, NHB had less self-reported medical conditions than NHW. Interestingly, we found that a greater burden of self-reported medical conditions was related to higher levels of amyloid. However, this effect did not account for the association between race

and amyloid (both aggregated medical conditions and race significantly predicted amyloid levels independently). Nevertheless, we were restricted to self-reported medical conditions, so it remains possible that selection bias present in the A4 dataset may have resulted in a more resilient and healthier NHB group relative to NHW that underwent PET, and that factors associated with this resilience are directly related to reduced amyloid levels. Unfortunately, we were unable to assess if there was a higher degree of medical conditions in the screen-failure group since these variables were not collected across all excluded participants (in particular, the majority of individuals that screen-failed based on cognitive testing scores did not have this data available). Along these lines, factors related to survival bias have been shown to influence race differences in stroke⁴⁹, and it's possible that similar confounds are present in the A4 screening dataset. However, even if the observed effect of reduced amyloid in NHB is driven by a selection bias, the pattern of results remains highly relevant for clinical trial design and recruitment. As a field it is critical to be cognizant of the biases introduced in the recruitment of individuals into clinical trials and the degree to which resulting cohorts accurately reflect our target populations.

In addition to differential rates of screen-failure by race during the initial screening phase, there are also known biases related to general participation in clinical trials, and it is possible that these biases systematically vary by race. For instance, recent work from the cancer field has shown specific bias in the actions of health care professionals when recruiting minority participants into clinical trials (including negative perceptions held by health care professionals related to minorities as unsuitable participants, ultimately leading to reduced recruitment of minority participants)⁵⁰. Implicit biases present during the recruitment stages may further contribute to a selection bias within the NHB group that does enroll. Further, older CN individuals that volunteer for AD prevention trials likely represent a biased group of educated individuals that are aware of these opportunities and have greater healthcare access. It is unclear how these motivations vary by race and result in different degrees of selection bias. Overall, as the field aims

to reduce health disparities in AD, it will be critical to address confounds that influence the recruitment of NHB individuals into AD clinical trials.

Our study has limitations. First, the current study lacks data on social health determinants such as socioeconomic status, lifetime stress exposures, etc. Likewise, our examination of vascular factors was limited to a small list of variables, and there are undoubtedly additional vascular measures that may impact amyloid in CN cohorts. Next, longitudinal measures of clinical progression would allow us to estimate whether the effect of amyloid on future clinical progression varies by race. Finally, replication of our findings in a larger CN NHB population is warranted, especially given the mixed findings across pre-existing literature²⁹.

Taken together, we found that CN NHB that were screened for an AD prevention trial had lower levels of fibrillar amyloid as measured with PET compared to CN NHW. Higher levels of African ancestry within the NHB group were associated with reduced amyloid levels, which may partially explain the group level effect of lower levels of amyloid in NHB. Finally, significant differences in eligibility and screen-failure rates existed across race that may influence our findings. Overall, the presence of a race effect on a central AD phenotype (amyloid PET) highlights the need to understand disease mechanisms and potential selection bias in diverse populations.

Figures legends and Tables

Figure 1. Proportion of amyloid positivity (A) and continuous amyloid SUVRs (B) across *APOE* genotypes for the NHB group. The E2/E4 group is shown in this plot but was excluded from statistical analyses. Sample sizes for each NHB genotype are listed, with the subset of individuals classified as amyloid+ in parentheses. Bars are missing for NHB *APOE*2/2 since there were no NHB individuals with this genotype. Likewise, bars are missing for the NHW Matched *APOE*2/4 since there were no NHW Matched individuals with this genotype. Given there was just one E2/2 subject in the NHW Matched group, there is no error bar drawn.

Figure 2. Genetic ancestry measures for all A4 participants (A) and NHB (B). Data are ranked by African ancestry, with each row representing the ancestry composition of a single individual participant. The NHW group shows little variance beyond the European ancestry (A), whereas examination of the NHB group reveals a continuum of African ancestry (B). Triangles = Non-Hispanic Whites; Stars = Non-Hispanic Blacks.

Figure 3. Plot of continuous amyloid PET SUVRs compared to percent African ancestry, with amyloid residualized by age, sex, number of *APOE*4 alleles, and number of *APOE*2 alleles.

	NHB	NHW	NHW-M
N	144	3689	288
AGE (YEARS)	70.77 (4.87)	71.24 (4.67)	70.72 (4.44)
EDUCATION (YEARS)	16.10 (2.76)	16.65 (2.83)	16.27 (2.63)
MINI MENTAL STATE EXAM	28.54 (1.34)	28.84 (1.19)	28.99 (1.11)
% FEMALE	105 (72.9)	2199 (59.6)	215 (74.7)
AMYLOID SUVR	1.04 (0.13)	1.10 (0.19)	1.12 (0.22)
% AMYLOID +	25 (17.4)	1030 (27.9)	89 (30.9)
% APOE*	-	-	-
APOE2/3, APOE2/2	31 (21.5)	403 (10.9)	27 (9.4)
APOE3/E3	68 (47.2)	2036 (55.2)	156 (54.2)
APOE3/4, APOE4/4	45 (31.2)	1250 (33.9)	40.5

Table 1: Demographics. Mean (standard deviation) are listed for continuous values. NHWm matched to NHB on age, sex, education using 2:1 propensity matching to the NHB group. *E2/E4 excluded.

A) Logistic Regression Predicting Amyloid group. Odds ratio (95% CI)		
	Full NHW Sample vs NHB	Matched NHW Sample vs NHB
Race (NHW v NHB)	1.94 (1.22-3.19) p=0.007	2.33 (1.35-4.13) p=0.003
Gender (Female v Male)	1.18 (1-1.39) p=0.044	1.37 (0.78-2.47) p=0.284
Age (years)	1.11 (1.09-1.12) p<0.0001	1.09 (1.04-1.15) p=0.0007
APOE2 (0, 1, 2)	0.69 (0.51-0.91) p=0.012	1.12 (0.45-2.49) p=0.791
APOE4 (0, 1, 2)	4.74 (4.09-5.5) p<0.0001	5.66 (3.62-9.13) p=<0.0001
B) Linear Regression Predicting Continuous Amyloid SUVR. Beta estimate (standard error)		
	Full NHW Sample vs NHB	Matched NHW Sample vs NHB
Race (NHW v NHB)	0.046 (0.015) p=0.002	0.066 (0.018) p=0.0003
Gender (Female v Male)	0.015 (0.006) p=0.008	0.036 (0.019) p=<0.0001
Age (years)	0.007 (0.001) p<0.0001	0.005 (0.002) p=0.006
APOE2 (0, 1, 2)	-0.020 (0.008) p=0.016	-0.016 (0.026) p=0.523
APOE4 (0, 1, 2)	0.133 (0.005) p<0.0001	0.137 (0.016) p=<0.0001

Table 2. Regression models summarizing effect of race on amyloid.

<i>Effects of race by genotype</i>	
NHB E4+ vs. NHW E4+	-0.094 (0.033) p=0.004
NHB E3/3 vs. NHW E3/3	-0.034 (0.019) p=0.070
NHB E2+ vs. NHW E2+	0.009 (0.026) p=0.723
<i>Effect of E2 within race</i>	
NHB E2+ vs. NHB E3/3	0.024 (0.024) p=0.315
NHW E2+ vs. NHW E3/3	-0.025 (0.008) p=0.002

Table 3: Post-hoc contrasts showing effect of race on amyloid SUVR for *APOE4* and *APOE3/3* genotypes, as well as effect of *APOE2* within race. All contrasts were performed with the full NHW group, controlling for age and sex. Beta estimate (standard error).

Appendix 1

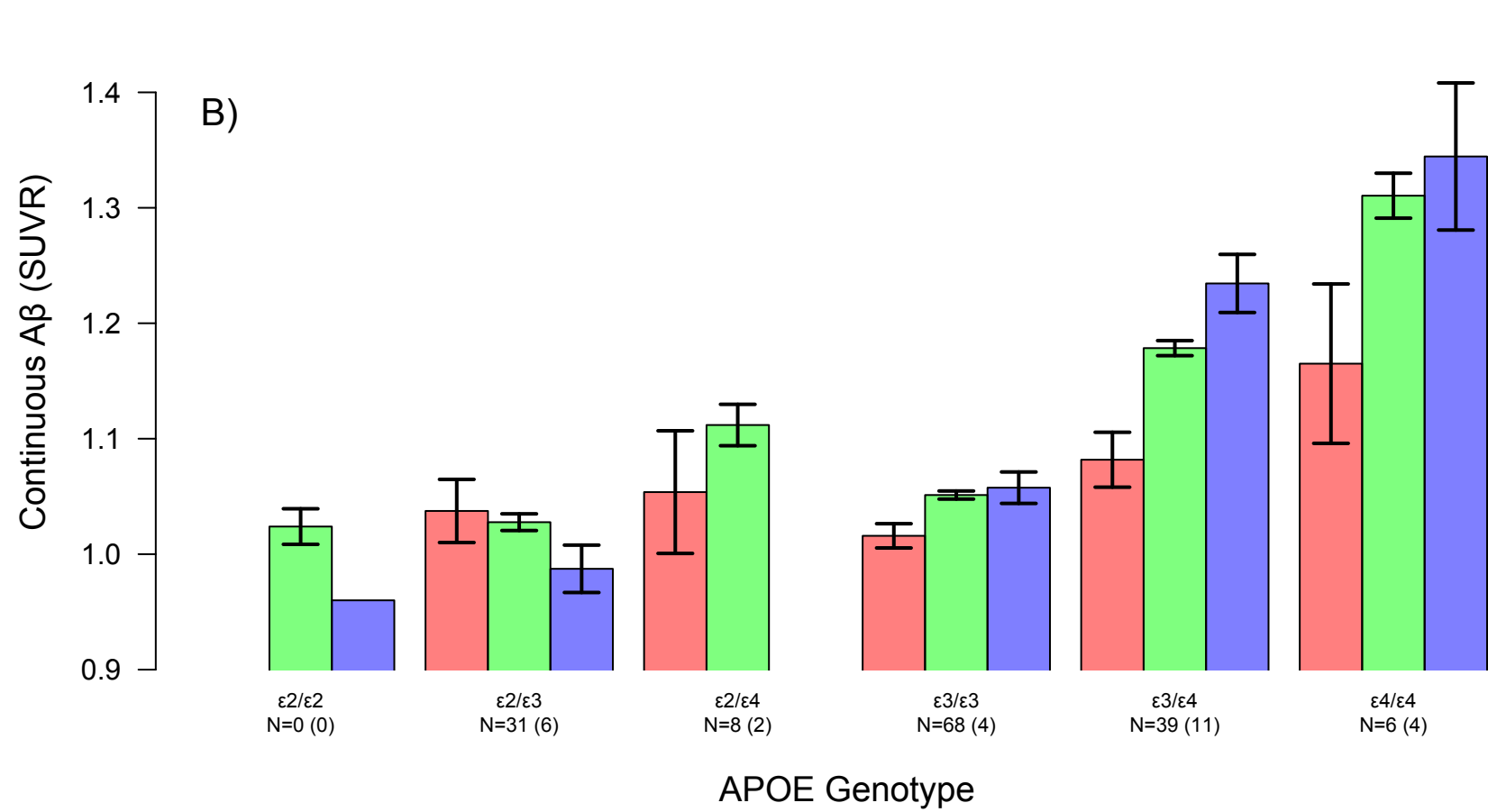
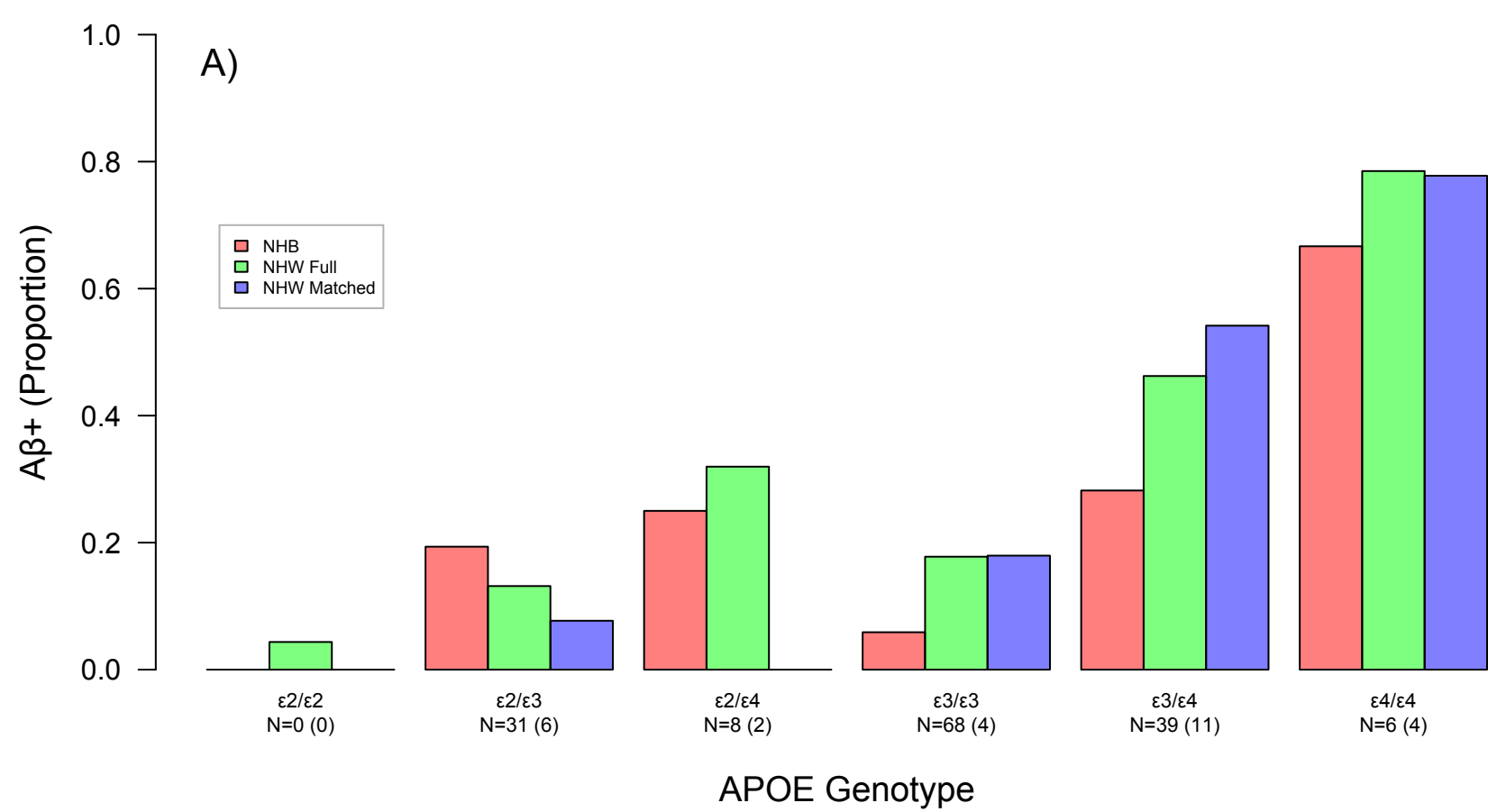
Name	Location	Contribution
Kacie Deters	Stanford Univ., Stanford, CA	Design and conceptualized study; drafted manuscript; data interpretation; data analysis; revised manuscript
Valerio Napolioni	Stanford Univ., Stanford, CA	data analysis; revised manuscript; data interpretation
Reisa A. Sperling	Brigham and Women's Hospital, Boston, MA	Acquisition of data; revised manuscript
Gabriel Kennedy	Stanford Univ., Stanford, CA	Data analysis
Michael D. Greicius	Stanford Univ., Stanford, CA	Data interpretation; manuscript revision; interpretation of data
Richard Mayeux	Columbia University Medical Center, New York, NY	Data interpretation; revised manuscript
Timothy Hohman	Vanderbilt Genetics Institute, Nashville, TN	Data interpretation; revised manuscript
Elizabeth C. Mormino	Stanford Univ., Stanford, CA	drafted manuscript; data interpretation; data analysis; revised manuscript

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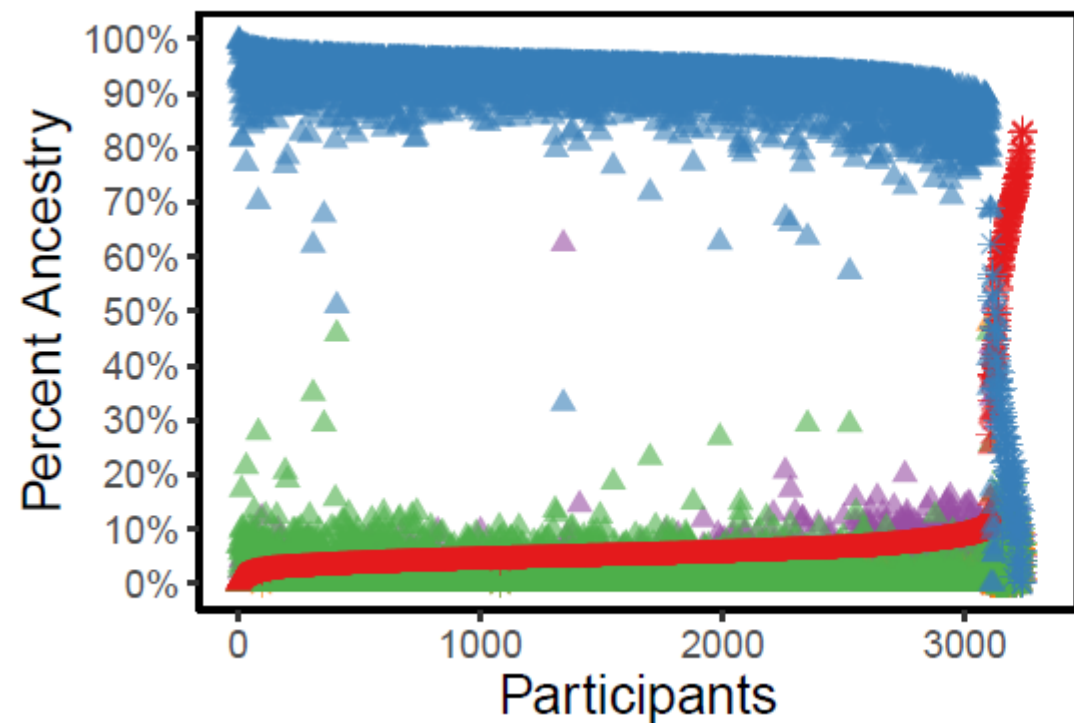
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A.



● African ● European ● American ● East Asian ● South Asian

▲ NHW ✱ NHB

B.

