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RESEARCH ARTICLE

Impact of sex and *APOE ε4* on the association of cognition and hippocampal volume in clinically normal, amyloid positive adults

Kellen K. Petersen[1] | **Ellen Grober**[1] | **Richard B. Lipton**[1] | **Reisa A. Sperling**[2,3] | **Rachel F. Buckley**[4] | **Paul S. Aisen**[5] | **Ali Ezzati**[1]

[1] Department of Neurology, Albert Einstein College of Medicine, New York City, New York, USA

[2] Department of Neurology, Harvard Aging Brain Study, Massachusetts General Hospital, Harvard Medical School, Boston, Massachusetts, USA

[3] Department of Neurology, Center for Alzheimer Research and Treatment, Brigham and Women's Hospital, Harvard Medical School, Boston, Massachusetts, USA

[4] Department of Neurology, Massachusetts General Hospital/Brigham and Women's Hospital/Harvard Medical School, Boston, Massachusetts, USA

[5] Alzheimer Therapeutic Research Institute, Keck School of Medicine, University of Southern California, San Diego, California, USA

Correspondence
Kellen Petersen, Department of Neurology, Albert Einstein College of Medicine, Van Etten 3C12, 1300 Morris Park Avenue, Bronx, NY 10461,USA.
Email: kellen.petersen@einsteinmed.org

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Abstract

Introduction: Cognitive decline follows pathological changes including neurodegeneration on the Alzheimer's disease continuum. However, it is unclear which cognitive domains first become affected by neurodegeneration in amyloid-positive individuals and if sex or apolipoprotein (*APOE*) ε4 status differences affect this relationship.

Methods: Data from 1233 cognitively unimpaired, amyloid-positive individuals 65 to 85 years of age were studied to assess the effect of hippocampal volume (HV) on cognition and to evaluate differences due to sex and *APOE* ε4 status.

Results: Lower HV was linked with worse performance on measures of memory (free recall, total recall, logical memory delayed recall, Mini-Mental State Examination [MMSE]), executive functioning (digit symbol substitution, DSS), and the Preclinical Alzheimer's Cognitive Composite (PACC). Among both women and *APOE* ε4+ individuals, all cognitive measures, except MMSE, were associated with HV. DSS and PACC had the largest effect sizes in differentiating early and intermediate stage neurodegeneration.

Discussion: Despite all cognitive measures being associated with HV, cognitive tests show differences in detecting early or late signs of neurodegeneration. Differences exist in association between cognition and neurodegeneration based on sex and *APOE* ε4 status

KEYWORDS
APOE ε4, cognition, FCSRT96, hippocampal volume, logical memory, MMSE, PACC, Sex

1 | INTRODUCTION

Pathologic hallmarks of Alzheimer's disease (AD) including amyloid beta (Aβ), tau (T), and neurodegeneration (N) are thought to occur long before individuals show clinical symptoms of AD.[1–4] To optimize early interventions, the complex relationship between these pathologic changes and different clinical phenotypes needs to be unraveled. A challenge in the early detection of the association between pathologic changes and clinical symptoms in the preclinical stages of AD is using cognitive tests, which are not sufficiently optimized to

detect subtle cognitive changes in cognitively unimpaired individuals. Based on a hypothetical model of dynamic biomarkers,[5] AD-related pathologic changes happen before clinical symptoms start. However, this might be related partially to the fact that cognitive tests are not sufficiently sensitive for the detection of subtle cognitive impairment. This might partially explain the lag between time from detection of AD pathology to detection of cognitive decline. Investigating the association between subtle neurodegenerative changes and cognitive changes in cognitively normal older adults could improve our mechanistic understanding of disease.

The Anti-Amyloid Treatment in Asymptomatic Alzheimer's study (A4 Study) developed the Preclinical Alzheimer Cognitive Composite (PACC), its primary cognitive outcome measure, to explore these associations and to be sensitive to change in preclinical AD.[6,7] PACC is formed using an established normalization method made up of four cognitive score components, each chosen based on a review of the literature regarding their assessment in three key domains relevant to preclinical AD: episodic memory, executive function, and orientation.[7] Most studies to date have focused on the relationship of PACC to amyloid status.[8] The focus of this study is to better understand the effect of neurodegeneration on cognition defined by PACC and its components.

We anticipate that the relationship between neurodegeneration and cognition will be affected by sex differences and apolipoprotein E (*APOE*) ε4 status. Mounting evidence suggests that women are at higher risk of developing AD pathophysiology[9–12] and therefore show clinical progression at a higher rate.[9,13,14] Although sex differences in Aβ burden alone have not been reported in cognitively normal older adults,[15,16] some research has found that women show less cognitive decline and hippocampal volume (HV) loss despite male-equivalent amyloid burden in aging.[17] Other work has found that Aβ- and *APOE* ε4-positive females exhibited faster cognitive declines than males[18] and potential increased susceptibility for clinical AD in women.[19]

The *APOE* ε4 allele is the strongest genetic risk factor for sporadic AD.[20,21] *APOE* ε4 carriers have higher levels of AD pathology and lower cognitive function, even in preclinical stages of the disease.[9,22] Until recently, that the association of *APOE* ε4 and AD is more pronounced in women had been commonly overlooked.[11,17] A meta-analysis confirmed a sex and *APOE* ε4 interaction such that ε3/ε4 women had up to a four-fold increased risk of progression to AD when compared to women homozygote for the risk-neutral ε3 allele, whereas men with one *APOE* ε4 allele had little to no increase in risk.[23]

In this study we used data from cognitively normal, amyloid-positive participants enrolled in the A4 Study to investigate how HV, a proxy for neurodegeneration, was related to the PACC and the cognitive tests, each relevant to preclinical AD, that comprise PACC. Specifically, we evaluated: (1) the relationship of specific cognitive tests to neurodegeneration; (2) the sex and *APOE* ε4 interaction and its effect on the relationship between neurodegeneration and performance on different cognitive domains; and (3) the difference in cognitive scores for individuals in early, intermediate, and advanced stages of neurodegeneration. Because neurodegeneration occurs before cognitive decline, and can be measured more precisely, it is important to understand the relationship between them. We hypothesized that even subtle neurodegenerative differences are related to cognitive performance overall and within different domains using the composite score PACC and its components, respectively. We found the relationship between neurodegeneration and cognition to be more pronounced in women and *APOE* ε4-positive individuals.

RESEARCH IN CONTEXT

1. **Systematic Review**: A literature review was conducted using traditional sources (eg, PubMed). Evidence suggests women that develop Alzheimer's disease (AD) pathophysiology, and therefore clinical progression, at a higher rate than men. In addition, the association between apolipoprotein E (*APOE*) ε4 and AD has been found to be more pronounced in women.
2. **Interpretation**: Our results indicate that different cognitive tests may have different sensitivities for early versus late signs of neurodegeneration and that differences exist in association between cognition and neurodegeneration based on sex and *APOE* ε4 status.
3. **Future Directions**: Our findings suggest that further work is needed to understand differences more fully in sex and *APOE* ε4 status on cognition and neurodegeneration, which may include: (1) further analysis using a data sample more representative of the general population; (2) additional analysis utilizing longitudinal information not available in this study; and (3) investigation of the effect of AD pathology on other brain regions.

HIGHLIGHT

- All cognitive measures found to be significantly associated with neurodegeneration
- Sensitivity of cognitive tests appear to differ by stage of neurodegeneration
- Differences found between cognition & neurodegeneration based on sex/APOE ε4 status

2 | METHODS

2.1 | Subjects/Participants

Data were obtained from the A4 Study, an ongoing clinical trial conducted at 67 sites in four countries among cognitively normal participants, from 65 to 85 years of age.[24] Among the reasons that an individual may have been excluded from the A4 study was that they had a non-zero Clinical Dementia Rating (CDR) score; they were taking a

prescribed Alzheimer's medication; they lived in a skilled nursing facility or nursing home; or they had a history or issue with a particular illness or disease, alcohol use, major depression, or suicidal risk.[6] A total of 6763 individuals completed the initial screening visit that included obtaining informed consent from all participants in addition to a collection of information such as demographics, family history, lifestyle habits, cognitive testing, functional questionnaire, and medical screening. A total of 4486 individuals were deemed eligible to participate in a second screening visit during which amyloid positron emission tomography (PET) imaging was obtained. A total of 1323 participants with abnormal amyloid were eligible to continue in screening for the A4 Study. Of those, 1265 received magnetic resonance imaging (MRI) and continued in the A4 Study. When our study was restricted to individuals with complete data profiles, our analysis included data from 1233 of these participants (Figure S1).

2.2 | Volumetric MRI

Volumetric parcellations of different cortical and subcortical regions were processed for all participants using FreeSurfer 6.0. The three-dimensional (3D) T1-weighted MRI scans were processed and quality control was performed using the automated segmentation software, approved by the US Food and Drug Administration (FDA) for clinical use (NeuroQuant; CorTechs Labs, San Diego, California).[24] In this study HV was used as a proxy for neurodegeneration (N).[4] To account for differences in intracranial volume (ICV), adjusted hippocampal volume (HVa) was calculated by residual correction using the following equation:

$$\mathrm{HVa} = \mathrm{HV} - \mathrm{B} * (\mathrm{ICV} - \mathrm{mean}\,(\mathrm{ICV}))$$

where B was obtained as a regression coefficient when HV is regressed against ICV.

2.3 | Cognitive tests

Participants completed the Preclinical Alzheimer Cognitive Composite (or PACC), a composite score of global cognition calculated as the average of the standardized scores on the following tests[6]:

- Mini-Mental State Examination (MMSE), a screening tool and measure of global cognition (range, 0–30).
- Delayed Logical Memory (DLM), recall of a narrative story 15 minutes after initial recall (range, 0–25).
- Digit Symbol Substitution (DSS), primarily a measure of executive function as well as processing speed and working memory (maximum score, 91).
- Free and Cued Selective Reminding Test (FCSRT), a multi-trial word recall test. Scores includes the sum of free recall (FR) alone (range, 0–48) and combined with total recall (TR), the sum of FR and cued recall (TR96: range, 0–96).

2.4 | Statistical analysis

All statistical analyses and computational work were conducted using MATLAB (version 2021a). The associations between demographic variables, cognitive scores, and HVa were calculated using Pearson correlation coefficient, and group differences were measured using independent *t*-tests and χ^2 tests. A series of multiple linear regression (MLR) models were conducted to investigate the association between the cognitive scores and HVa in the whole population adjusting for sex, age, education, and *APOE* ε4 status. We performed hierarchical regression analysis to test our hypotheses that sex and *APOE* ε4 status influence the association between HVa and cognition. The Akaike information criterion (AIC) measures the quality of the different multiple regression models, where lower values indicate a better fitting model with the ΔAIC being the difference from the HVa-only model (Model 1).[25] In addition, we stratified our sample based on sex and *APOE* ε4 status and developed MLR models for each subsample with HVa as the independent variable, age and education as covariates, and cognitive performance as the outcome.

To further explore the relationship between neurodegeneration and cognition, and to elucidate any potential nonlinear relationship between them, we fit monotonically decreasing cubic splines (MDCS) to the z-scores of the different cognitive scores, where the spline knots were chosen at the end points of the HVa intervals.[26] Cubic splines are piecewise third-order polynomial interpolations that are both simple to implement and perform well in terms of reduced interpolation error. Specifically, we utilized cubic splines of the Hermite form. Because neurodegeneration is associated with decreased cognitive ability, we used monotonic splines to capture this relationship. Furthermore, MDCSs provide insight into the non-linear behavior between neurodegeneration and cognition that is not observed in linear regression models. In addition, sex- and *APOE* ε4-stratified groups were interpolated using MDCS to understand the separate effects on cognition and neurodegeneration.

Finally, to understand the effect of early-to-intermediate and intermediate-to-advanced stage neurodegeneration on cognition, we stratified the whole sample into three groups based on tertiles of HV. Group differences were assessed by calculating the effect size, with a corresponding *P*-value, as the mean difference between samples scaled by earlier-stage standard deviation. Similar analysis was repeated after stratification of the sample based on sex and *APOE* ε4 and included as supplementary material.

3 | RESULTS

3.1 | Cohort characteristics

Participants had a mean age of 71.99 (SD = 4.84) years and 16.6 (SD = 2.81) years of education; 58.6% were female, 94.4% were White, and 58.6% were *APOE* ε4 carriers (Table 1). In addition, the average PET standardized uptake value ratio (SUVr) was 1.33 (standard deviation, SD = 0.18) and the average HVa was 6.75 (SD = 0.74) cm^3. Figure S2

TABLE 1 Sample characteristics for the whole sample and for subgroups based on sex and *APOE* ε4 status

		Sex			*APOE* ε4 status		
	Entire sample	Men	Women	*P*-value	Negative	Positive	*P*-value
Sample size	1233	511	722		511	722	
Men/women	511/722	511/0	0/722		211/300	300/422	.925
White/other	1164/69	471/40	693/29	.002	473/38	691/31	.018
APOE ε4 status (+ / -)	722/511	300/211	422/300	.927	0/511	722/0	
Age (years)	71.99 (4.84)	72.83 (5.10)	71.39 (4.56)	<.001	72.96 (5.08)	71.30 (4.54)	<.001
Education (years)	16.56 (2.81)	17.01 (2.93)	16.25 (2.68)	<.001	16.49 (2.95)	16.62 (2.70)	.4336
Free Recall	28.28 (5.73)	26.42 (5.64)	29.59 (5.41)	<.001	28.50 (5.70)	28.12 (5.74)	.244
TR96 (FR+TR)	75.55 (6.11)	73.58 (6.03)	76.96 (5.78)	<.001	75.78 (6.00)	75.39 (6.19)	.2666
MMSE	28.73 (1.30)	28.52 (1.39)	28.87 (1.21)	<.001	28.75 (1.30)	28.71 (1.30)	.6535
DLM	11.43 (3.33)	11.37 (3.33)	11.48 (3.33)	.5914	11.50 (3.39)	11.38 (3.28)	.5191
DSS	42.62 (8.92)	40.52 (8.24)	44.11 (9.09)	<.001	41.87 (8.54)	43.16 (9.15)	.0126
PACC	−0.40 (2.68)	−1.15 (2.64)	0.13 (2.58)	<.001	−0.40 (2.66)	−0.40 (2.69)	.9854
PET SUVr	1.33 (0.18)	1.33 (0.18)	1.33 (0.18)	.8431	1.28 (0.18)	1.36 (0.18)	<.001
HVa	6.75 (0.74)	6.59 (0.77)	6.86 (0.69)	<.001	6.73 (0.71)	6.76 (0.75)	.461

Note: Using *t*-tests or continuous variables and chi-square test for categorical variables. The cognitive exams are Free Recall, TR96, MMSE, DLM, DSS, and PACC. PET SUVr is the standardized uptake value ratio for amyloid positron emission tomography (PET) imaging and HVa is the adjusted hippocampal volume. Abbreviations: HVa, adjusted hippocampal volume; TR96, Free and Cued Selective Reminding Test; DLM, delayed logical memory; DSS, Digit Symbol substitution; MMSE, Mini-Mental State Examination; PACC, Preclinical Alzheimer's Cognitive Composite; TR, Total Recall.

shows comparative cognitive performance of subgroups with abnormal and normal HVa.

Men were slightly older ($t = 5.23$, $P < .001$) and had higher education ($t = 4.77$, $P < .001$) in comparison with women. A total of 58.5% of women and 58.7% of men were *APOE* ε4 carriers. There was no significant difference between PET SUVr levels in men and women ($t = 0.20$, $P = .585$), but men were found to have significantly larger HVa ($t = 6.44$, $P < .001$). Women were also found to have performed significantly better on all cognitive tests ($P < .001$) except for DLM ($P = .591$).

APOE ε4-negative individuals were older ($t = 6.01$, $P < .001$) in comparison with *APOE* ε4 positive individuals but did not differ in levels of education ($t = 0.78$, $P = .434$). HVa was not different between *APOE* ε4 positive and negative individuals ($t = 0.74$, $P = .461$) but *APOE* ε4 positives had higher PET SUVr values ($t = 7.51$, $P < .001$).

3.2 | Cognition and HV

Regressions were plotted along with 95% confidence intervals and (unadjusted) data points (Figure 1). In addition, box-and-whisker plots were used to visualize the distribution and spread of cognitive test scores for individuals with and without neurodegeneration (Figure S2).

Table 2 summarizes the results for various hierarchically constructed, or nested, MLR models assessing the association between cognitive scores and HV. Model 1 regresses the cognitive scores against HVa with age and education as covariates. All cognitive scores were found to be significantly associated with HVa ($P < 0.05$) except DLM ($P = 0.053$). Models 2 and 3 consist of the addition of sex and APOE ε4 status, respectively, to Model 1. Using ΔAIC as a measure of meaningful improvement between models, we observed that adding sex to Model 1 resulted in significant model improvement for all cognitive outcomes except DLM (ΔAIC ← 16 for all outcomes except ΔAIC = 2.00 for DLM). The addition of *APOE* ε4 status to Model 1 only meaningfully improved model fit (ΔAIC ← 2) for models with FR and TR96 as outcomes. Finally, in Model 4, we added both sex and *APOE* ε4 status to Model 1, which resulted in the best fitting model among all outcomes except DLM and DSS. Model 3 exhibited the lowest AIC for DLM and Model 2 for DSS.

MLR models were repeated in samples stratified by sex and by *APOE* ε4 status (Table 3). There was no association between any cognitive score and HVa in men ($P > .05$ for all), whereas in women all cognitive scores except MMSE were significantly associated with HVa ($P < .02$ for all; $P = .062$ for MMSE). When stratified by *APOE* ε4 status, no cognitive measure was found to be significantly associated with HVa in *APOE* ε4-negative individuals, but all cognitive variables were found to be significantly associated with HVa in positive individuals ($P < .05$ for all).

Using MDCS, in Figure 2, we illustrated the non-linear relationship between various cognitive measures and HVa for the entire population. As is confirmed in Table 4 by comparison of effect sizes between stages, during advanced neurodegeneration we see an increasing difference in all cognitive measures where the smallest change was in DSS. In addition, MMSE and DLM show little differences between scores for individuals in the intermediate stages of neurodegeneration. Figure 3 shows MDCS interpolation of relationship between PACC and HVa in sex- and *APOE* ε4-stratified subsamples. For men, PACC decreases steadily throughout the range of HVa, whereas in women there is a sharp decrease in PACC in the HVa range corresponding to advanced neurodegeneration. These observations are confirmed in Table 5, which summarizes the results for differences in

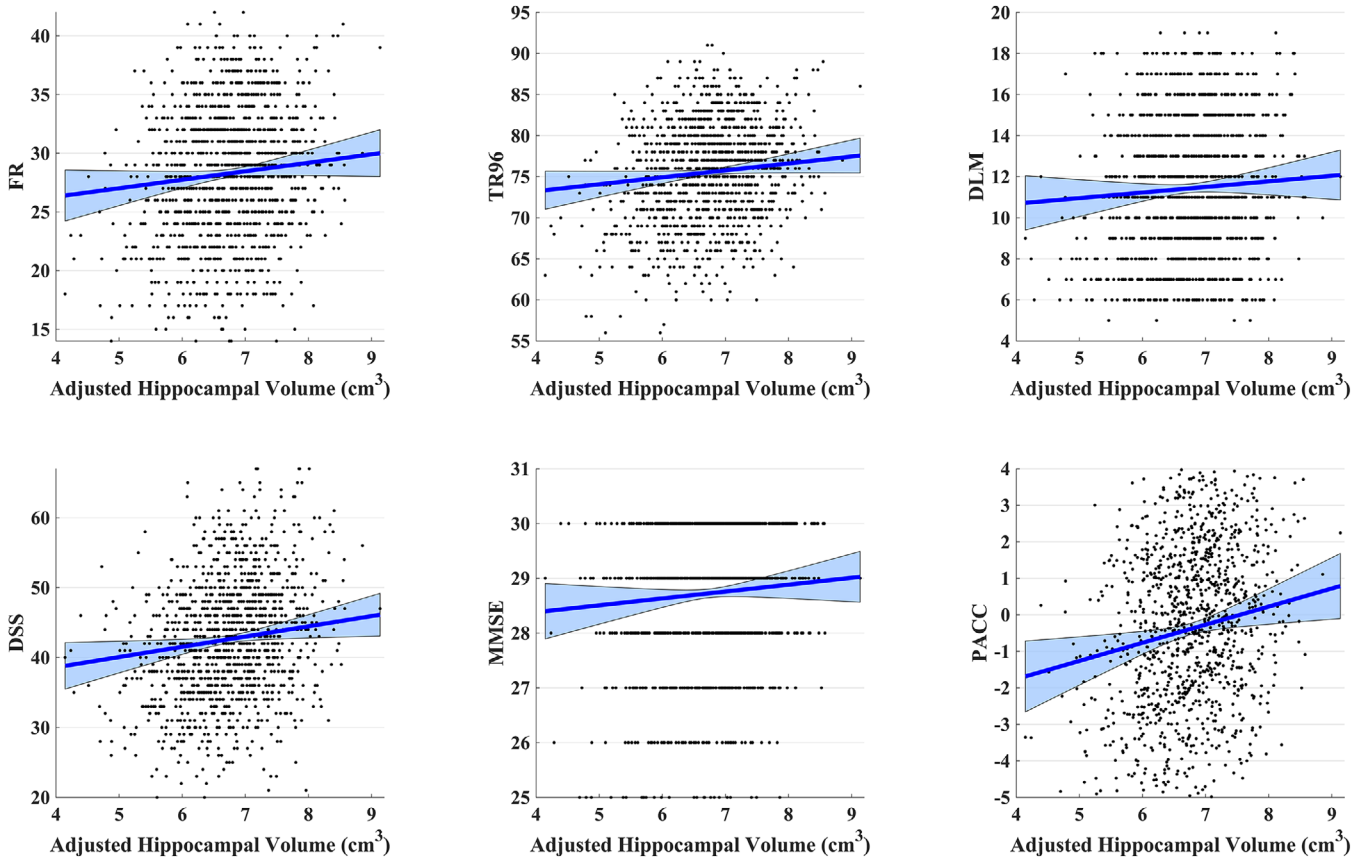

FIGURE 1 Effect of adjusted hippocampal volume (HVa) on cognitive test scores. Linear regression models adjusted for sex, age, education, and *APOE* ε4 status with 95% confidence intervals. The black markers indicate the cognitive score and HVa for each individual of the sample

TABLE 2 Results for four multiple regression models obtained by a hierarchical modification to a previous model

Variables	Model 1 HVa		Model 2 HVa+Sex			Model 3 HVa + *APOE* ε4			Model 4 HVa + Sex + *APOE* ε4		
	HVa		HVa			HVa			HVa		
Outcomes	*β*	*P*	*β*	*P*	ΔAIC	*β*	*P*	ΔAIC	*β*	*P*	ΔAIC
Free Recall	0.132	<.001	0.100	.0022	−69.49	0.125	<.001	−3.66	0.093	.0042	−72.86
TR96	0.140	<.001	0.108	<.001	−68.31	0.133	<.001	−3.72	0.102	.0017	−71.73
DLM	0.065	.0532	0.065	.0554	2.00	0.060	.0738	−0.54	0.060	.0761	1.46
DSS	0.140	<.001	0.118	<.001	−33.33	0.143	<.001	1.17	0.121	<.001	−32.37
MMSE	0.092	.0055	0.076	.0229	−16.75	0.088	.0085	−0.31	0.071	.0321	−16.90
PACC	0.168	<.001	0.142	<.001	−53.28	0.163	<.001	−1.84	0.137	<.001	−54.84

Note: All models include age and education as covariates. Standardized regression coefficients (*β*), and its accompanying *P*-value, are reported for the HVa variable, but not the sex or *APOE* ε4 status variables. Additionally, ΔAIC is reported as the change in AIC (Akaike information criterion) compared to the that of Model 1, where the AIC is a relative measure of the quality of the model such that a lower AIC value is considered better. The outcomes are different cognitive tests: Free Recall, TR96, MMSE, DLM, DSS and PACC.

Abbreviations: HVa, adjusted hippocampal volume; TR96, Free and Cued Selective Reminding Test; DLM, delayed logical memory; DSS, Digit Symbol Substitution; MMSE, Mini-Mental State Examination; PACC, Preclinical Alzheimer's Cognitive Composite; TR, Total Recall.

neurodegeneration stages for PACC for the sex- and *APOE* ε4-stratified subsamples. In Table 5 it is shown that the effect size for women increases substantially between stages unlike for men. As seen in FIgure 3, the MDCS of *APOE* ε4-negative and *APOE* ε4-positive individuals see similar trajectories. A comparison of effect sizes in Table 5 found *APOE* ε4-positive individuals had a lower HVa in advanced neurodegeneration.

Table 4 shows the average test scores in the three groups based on HVa (early vs intermediate vs advanced neurodegeneration). In the entire sample, four cognitive measures—DSS ($P < .001$), PACC

TABLE 3 Regression models assessing the association between cognition and HVa where the cognitive test is the dependent variable

	Subsample							
	Men		Women		*APOE ε4+*		*APOE ε4−*	
	β	*P*	*β*	*P*	*β*	*P*	*β*	*P*
Outcomes								
Free Recall	0.060	.262	0.119	.005	0.104	.014	0.070	.168
TR96	0.072	.173	0.125	.003	0.119	.005	0.070	.172
DLM	−0.008	.884	0.108	.011	0.119	.007	−0.023	.661
DSS	0.070	.169	0.155	.000	0.128	.002	0.101	.045
MMSE	0.064	.221	0.079	.062	0.089	.041	0.039	.450
PACC	0.077	.125	0.183	<.001	0.177	<.001	0.068	.165

Note: Multiple regression models stratified based on sex and *APOE ε4* status, where the outcomes are the individual cognitive tests and HVa is the independent variable. Sex-stratified models are controlled for age, education, and *APOE ε4* status as covariates and models stratified by *APOE ε4* status control for age, sex, and education.

Abbreviations: HVa, adjusted hippocampal volume; TR96, Free and Cued Selective Reminding Test; DLM, delayed logical memory; DSS, Digit Symbol Substitution; MMSE, Mini-Mental State Examination; PACC, Preclinical Alzheimer's Cognitive Composite; TR, Total Recall.

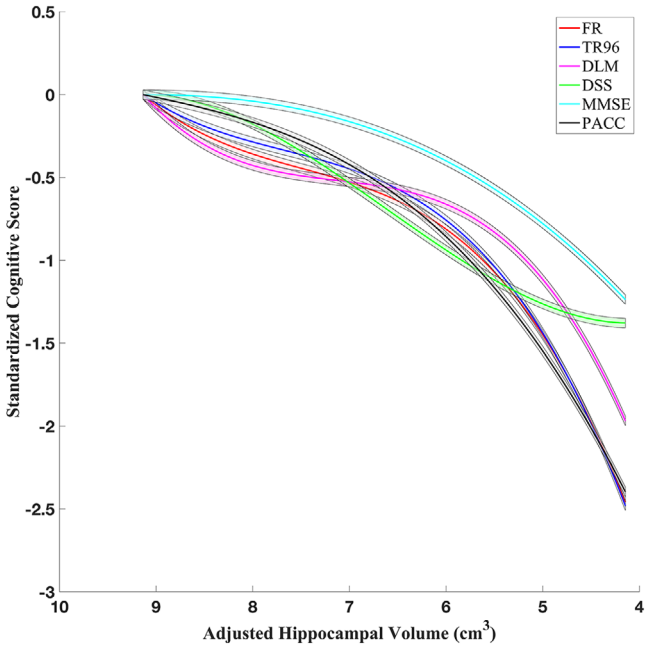

FIGURE 2 Plot of monotonically decreasing cubic spline interpolations for the z-scores for all cognitive measures versus adjusted hippocampal volume (HVa), with confidence intervals corresponding to the standard error of mean

(*P* = .001), FR (*P* = .049), and TR96 (*P* = .044)—showed significant differences in early-to-intermediate stage neurodegeneration. All cognitive measures had significant differences between intermediate and advanced neurodegeneration stages (*P* < .001 for all). The cognitive measures with the largest effect sizes (ES) for detecting early-to-intermediate neurodegeneration were DSS (ES = −0.32) and PACC (ES = −0.27). For detecting intermediate-to-advanced neurodegeneration, the largest effect sizes were observed in PACC (ES = −0.90) and TR96 (ES = −0.67).

Tables S1-S4 summarizes the results for the parallel analysis performed for the sex- and *APOE ε4*-stratified subsamples. From sex-stratified analysis, in men, differences in MMSE and PACC were most pronounced in early-to-intermediate neurodegeneration (MMSE: *P* = .041, ES = −0.32; PACC: *P* = .021, ES = −0.31) and differences in DSS and TR96 were observed in intermediate-to-advanced neurodegeneration (DSS: <0.001, ES = −0.42; TR96: *P* = .001, ES = −0.41). For women, DSS and PACC were found to have significant differences in early-to-intermediate neurodegeneration (DSS: *P* < .001, ES = −0.34; PACC: *P* = .031, ES = −0.22) and all cognitive measures had significant differences during intermediate-to-advanced neurodegeneration stages (*P* < .001 for all) with PACC having the largest effect size (ES = −1.18).

From *APOE ε4*-stratified analysis, differences in PACC and DSS in *APOE ε4*-negative individuals were most pronounced in early-to-intermediate neurodegeneration (PACC: *P* < .001, ES = −0.34; DSS: *P* = .002, ES = −0.30) and the largest differences in intermediate-to-advanced neurodegeneration were observed in PACC (PACC: *P* < .001, ES = −1.07). For *APOE ε4*-positive individuals, DSS was found to have the largest effect size in early-to-intermediate neurodegeneration (DSS: *P* = .005, ES = −0.35), whereas PACC exhibited the largest effect size in intermediate-to-advanced neurodegeneration (PACC: *P* < .001, ES = −0.67).

4 | DISCUSSION

At cross-section, HVa, a proxy for neurodegeneration, was associated with cognitive performance on the PACC and each of its components except the DLM. After stratifications based on sex and *APOE ε4*, associations remained significant only in women and *APOE ε4*-positive individuals. Average scores on DSS and TR96 decreased early with respect to neurodegeneration stage, whereas performance on all cognitive tests was worse when neurodegeneration was more advanced, with TR96 displaying the largest effects.

Using multiple linear regression and nested models, we found the inclusion of sex to improve the models for all cognitive measures except DLM. Similarly, the inclusion of *APOE ε4* status to Model 1 improved all models except DSS. PACC, which showed a decreased AIC with the inclusion of each additional variable, is a composite measure of four scores that assess different key domains. The separate analysis for the individual scores allows for more clarity in understanding the relationship between neurodegeneration, sex, *APOE ε4*, and cognition in preclinical AD. As such, the inclusion of sex to Model 1 resulted in a worse model for DLM, whereas the inclusion of the *APOE ε4* status to Model 1 resulted in a worse model for DSS.

Sex-specific differences contribute to the risk of cognitive decline in prodromal AD.[14] In a large study of cognitively normal individuals across the adult lifespan, A*β* burden on PET increased similarly with age in both men and women; however, men showed worse

TABLE 4 Mean cognitive scores, standard deviations, and effect sizes

	Neurodegeneration Stage											
	Early (large HVa) (N = 193)		Intermediate (intermediate HVa) (N = 914)		Advanced (Small HVa) (N = 126)		Early versus intermediate			Intermediate versus advanced		
	Mean	SD	Mean	SD	Mean	SD	*P*-value	Δ	Effect Size ($\Delta_{1\text{-}2}$/SD)	*P*-value	Δ	Effect Size ($\Delta_{2\text{-}3}$/SD)
Free Recall	29.37	5.11	28.50	5.67	24.98	5.92	.049	−0.87	−0.17	<.001	−3.53	−0.62
TR96	76.76	5.32	75.82	5.99	71.82	6.78	.044	−0.94	−0.18	<.001	−4.00	−0.67
DLM	11.71	3.38	11.57	3.30	10.05	3.16	.580	−0.15	−0.04	<.001	−1.52	−0.46
DSS	45.66	9.28	42.69	8.63	37.48	8.16	<.001	−2.97	−0.32	<.001	−5.22	−0.60
MMSE	28.93	1.12	28.77	1.27	28.10	1.58	.110	−0.16	−0.14	<.001	−0.68	−0.53
PACC	0.39	2.46	−0.27	2.54	−2.54	2.96	.001	−0.66	−0.27	<.001	−2.27	−0.90

Note: HVa range for each group: HVa of the early group ($N_1 = 193$) is between 7.4717 and 9.1350 cm^3, HVa of intermediate group ($N_2 = 914$) is between 5.8084 and 7.4717cm^3, and HVa of advanced group ($N_3 = 126$) is between 4.1452 and 5. 8084 cm^3. The *P*-values in the table corresponds to the *P*-values from a two-sample *t*-test between two groups. Δ is the difference between means of each group and the next one. Tables stratified by sex and *APOE* ε4 status are included in Table 5 and the Supplementary Material.

Abbreviations: HVa, adjusted hippocampal volume; TR96, Free and Cued Selective Reminding Test; DLM, delayed logical memory; DSS, Digit Symbol Substitution; MMSE, Mini-Mental State Examination; PACC, Preclinical Alzheimer's Cognitive Composite; TR, Total Recall.

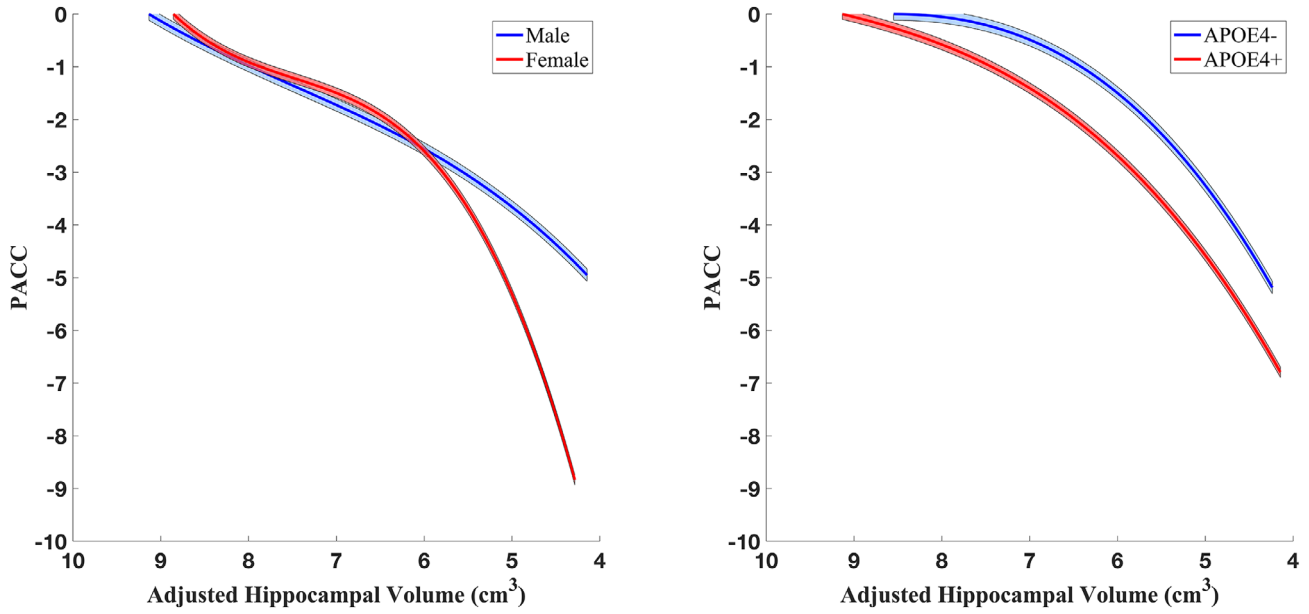

FIGURE 3 Plot of monotonically decreasing cubic spline interpolations for the z-scores for sex-stratified (left) and *APOE* ε4-stratified (right) subsamples of the Preclinical Alzheimer's Cognitive Composite (PACC) score versus adjusted hippocampal volume (HVa) with confidence intervals corresponding to the standard error of mean

memory and lower HVs over time, compared to women.[17] In this study, after stratification based on sex, we found that all cognitive measures, except MMSE, were significantly associated with HVa in women, but not in men. In women, the effect of neurodegeneration on cognition in early stages of neurodegeneration appears to be minimal; however, this effect seems to accelerate in more advanced stages of neurodegeneration (Figure 3 and Tables S1 and S2). This suggests that the effect of neurodegeneration on women may appear more pronounced later and this would have implications for both the cognitive assessment of women as well as determination of preclinical AD in these individuals.

APOE ε4 is considered the greatest genetic risk factor for AD.[20,21] In human studies, some but not all imaging biomarker studies have shown early AD-like findings in healthy older *APOE* ε4 carriers.[27–29] Longitudinal biomarker studies suggest that increased rates of hippocampal loss are associated with presence of the *APOE* ε4 alleles in Aβ+ individuals.[29] In the current study, only in the *APOE* ε4-positive group were cognitive scores associated with HV (except DSS in the *APOE* ε4-negative group, $P = .45$). This might be due to more advanced neurodegeneration in *APOE* ε4-positive individuals, even in prodromal stages of AD. There were no *APOE* ε4 by sex interactions on cognition and

TABLE 5 Mean PACC, standard deviations, and effect sizes for sex- and *APOE* ε4-stratified subsamples

		Neurodegeneration stage											
		Early (large HVa)		Intermediate (intermediate HVa)		Advanced (small HVa)		Early versus intermediate			Intermediate versus advanced		
	Subsample	Mean	SD	Mean	SD	Mean	SD	*P*-value	Δ	Effect Size ($\Delta_{1\text{-}2}$/SD)	*P*-value	Δ	Effect Size ($\Delta_{2\text{-}3}$/SD)
PACC	Male	−0.25	2.47	−1.01	2.50	−2.48	2.90	.02	−0.76	−0.31	<.001	−1.47	−0.59
	Female	0.74	2.39	0.22	2.44	−2.66	3.10	.03	−0.52	−0.22	<.001	−2.88	−1.18
	APOE ε4−-	0.10	2.37	−0.28	2.64	−2.05	2.70	.25	−0.38	−0.16	<.001	−1.77	−0.67
	APOE ε4+	0.58	2.50	−0.26	2.46	−2.88	3.09	<.001	−0.84	−0.34	<.001	−2.62	−1.07

Note: The ranges for the stage of neurodegeneration were the same as were used in Table 4. The *P*-values in the table corresponds to the *P*-values from a two-sample *t*-test between two groups. Δ is the difference between means of each group and the next one.
Abbreviations: HVa, adjusted hippocampal volume; PACC, Preclinical Alzheimer's Cognitive Composite

neurodegeneration found in this sample. Once the longitudinal data from the A4 Study are available, we can assess whether the *APOE* ε4 association with cognitive decline (and incident AD) is more pronounced in women, as suggested by other studies.[11,30]

PACC was also found to decline earlier and exhibit large effects in the advanced neurodegeneration stage. The observed changes in DSS in early to intermediate neurodegeneration is different from the pattern observed related to increasing amyloid burden.[8,31] In other studies, FR alone or combined with total recall (TR96) have been reported as the only tests in the PACC composite to show differences between the Aβ+ group who progressed from CDR of 0 to CDR 0.5 versus those who remained stable.[31] Thus early neurodegenerative changes may affect cognitive domains that are different from those affected by amyloid deposition. Our results indicate that the sensitivity of different cognitive tests in detecting neurodegenerative changes may vary depending on the stage of neurodegeneration in clinically unimpaired older adults.

This study has several limitations. The cross-sectional design precludes establishing a direct causal link between HV and cognition. Although the sample used in this study was large, the A4 Study was restricted to Aβ+ individuals over the age of 65 years who were considered cognitively normal (CDR = 0). Thus our results apply only to cognitively normal, Aβ+ individuals. Furthermore, this inclusion criteria can introduce a selection bias in our sample, limiting analyses to the effect of early neurodegenerative changes on cognition within late-onset preclinical AD. Although we focused on hippocampal atrophy as a proxy for neurodegeneration, there are several other brain regions affected by AD pathology, early in the course of disease.[4] In addition, although stratified analysis was performed to better understand the association between neurodegeneration and cognition and the effect of sex and *APOE* ε4 status, the interactions between them were not explicitly studied. Finally, the participants in the A4 Study are large non-Hispanic and White. Because AD biomarkers have a possible race-dependent biological mechanism,[32,33] the findings here are limited to the racial demographics of this study and should be further studied in more diverse populations.

Despite these limitations, this work did have several strengths. First, the data used in this study were from a large sample of cognitively normal, Aβ+ individuals. Furthermore, scores from several cognitive tests assessing different key domains were available and able to be utilized in the study. In addition, several different analytical approaches were used to investigate the stated hypotheses. As such, we were able to demonstrate that the effect of neurodegeneration in detecting cognitive changes may vary depending on the stage of neurodegeneration and may be influenced by the sex and *APOE* ε4 status of the individual.

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CONFLICTS OF INTEREST

The authors have no conflicts of interest to report except Ellen Grober, PhD, who receives a small royalty for commercial use of the Free and Cued Selective Reminding Test with Immediate Recall. The test is available at no cost for research or clinical activities from the Albert Einstein College of Medicine.

ORCID

Kellen K. Petersen https://orcid.org/0000-0003-3195-3456

REFERENCES

1. Sperling RA, Aisen PS, Beckett LA, et al. Toward defining the preclinical stages of Alzheimer's disease: recommendations from the National Institute on Aging-Alzheimer's Association workgroups on diagnostic guidelines for Alzheimer's disease. *Alzheimers Dement*. 2011; 7(3): 280–292.
2. Jack CR Jr, Knopman DS, Weigand SD, et al. An operational approach to National Institute on Aging-Alzheimer's Association criteria for preclinical Alzheimer disease. *Ann Neurol*. 2012; 71(6): 765–775.
3. Jack CR Jr, Wiste HJ, Weigand SD, et al. Different definitions of neurodegeneration produce similar amyloid/neurodegeneration biomarker group findings. *Brain*. 2015; 138(Pt 12): 3747–3759.
4. Jack CR Jr, Bennett DA, Blennow K, et al. NIA-AA Research Framework: toward a biological definition of Alzheimer's disease. *Alzheimers Dement*. 2018; 14(4): 535–562.
5. Jack CR, Knopman DS, Jagust WJ, et al. Tracking pathophysiological processes in Alzheimer's disease: an updated hypothetical model of dynamic biomarkers. *Lancet Neurol*. 2013; 12(2): 207–216.
6. Sperling RA, Donohue MC, Raman R, et al. Association of factors with elevated amyloid burden in clinically normal older individuals. *JAMA Neurol*. 2020; 77(6): 735–745.
7. Donohue MC, Sperling RA, Salmon DP, et al. The preclinical Alzheimer cognitive composite: measuring amyloid-related decline. *JAMA Neurol*. 2014; 71(8): 961–970.
8. Insel PS, Donohue MC, Sperling R, Hansson O, Mattsson-Carlgren N. The A4 study: beta-amyloid and cognition in 4432 cognitively unimpaired adults. *Ann Clin Transl Neurol*. 2020; 7(5): 776–785.
9. Buckley RF, Mormino EC, Rabin JS, et al. Sex differences in the association of global amyloid and regional tau deposition measured by positron emission tomography in clinically normal older adults. *JAMA Neurol*. 2019; 76(5): 542–551.
10. Buckley RF, Scott MR, Jacobs HIL, et al. Sex mediates relationships between regional tau pathology and cognitive decline. *Ann Neurol*. 2020; 88(5): 921–932.
11. Altmann A, Tian L, Henderson VW, Greicius MD. Sex modifies the APOE-related risk of developing Alzheimer disease. *Ann Neurol*. 2014; 75(4): 563–573.
12. Wisch J, Hudson D, Coble DW, et al. Socioeconomic status mediating sex and racial differences using the AT(N) framework. *Alzheimers Dement*. 2020; 16(S10): e041229.
13. Mosconi L, Berti V, Quinn C, et al. Sex differences in Alzheimer risk: brain imaging of endocrine vs chronologic aging. *Neurology*. 2017; 89(13): 1382–1390.
14. Laws KR, Irvine K, Gale TM. Sex differences in cognitive impairment in Alzheimer's disease. *World J Psychiatry*. 2016; 6(1): 54–65.
15. Mielke MM, Wiste HJ, Weigand SD, et al. Indicators of amyloid burden in a population-based study of cognitively normal elderly. *Neurology*. 2012; 79(15): 1570–1577.
16. Morris JC, Roe CM, Xiong C, et al. APOE predicts amyloid-beta but not tau Alzheimer pathology in cognitively normal aging. *Ann Neurol*. 2010; 67(1): 122–131.
17. Jack CR Jr, Wiste HJ, Weigand SD, et al. Age, sex, and APOE ε4 effects on memory, brain structure, and β-amyloid across the adult life span. *JAMA Neurol*. 2015; 72(5): 511–519.
18. Buckley RF, Mormino EC, Amariglio RE, et al. Sex, amyloid, and APOE ε4 and risk of cognitive decline in preclinical Alzheimer's disease: findings from three well-characterized cohorts. *Alzheimers Dement*. 2018; 14(9): 1193–1203.
19. Koran MEI, Wagener M, Hohman TJ. Sex differences in the association between AD biomarkers and cognitive decline. *Brain Imaging Behav*. 2017; 11(1): 205–213.
20. Apostolova LG, Risacher SL, Duran T, et al. Associations of the top 20 Alzheimer disease risk variants with brain amyloidosis. *JAMA Neurol*. 2018; 75(3): 328–341.
21. Di Battista AM, Heinsinger NM, Rebeck GW. Alzheimer's disease genetic risk factor APOE-ε4 also affects normal brain function. *Curr Alzheimer Res*. 2016; 13(11): 1200–1207.
22. Hohman TJ, Dumitrescu L, Barnes LL, et al. Sex-specific association of apolipoprotein E With cerebrospinal fluid levels of tau. *JAMA Neurol*. 2018; 75(8): 989–998.
23. Farrer LA, Cupples LA, Haines JL, et al. Effects of age, sex, and ethnicity on the association between apolipoprotein E genotype and Alzheimer disease. A meta-analysis. APOE and Alzheimer Disease Meta Analysis Consortium. *JAMA*. 1997; 278(16): 1349–1356.
24. Anti-amyloid treatment in asymptomatic Alzheimer's. https://ida.loni.usc.edu
25. Burnham KP, Anderson DR. Multimodel inference: understanding AIC and BIC in model selection. *Sociol Methods Res*. 2004; 33(2): 261–304.
26. Ramsay JO. Monotone regression splines in action. *Statistical Science*. 1988; 3(4): 425–441.
27. Sunderland T, Mirza N, Putnam KT, et al. Cerebrospinal fluid beta-amyloid1-42 and tau in control subjects at risk for Alzheimer's disease: the effect of APOE epsilon4 allele. *Biol Psychiatry*. 2004; 56(9): 670–676.
28. Shaw LM, Vanderstichele H, Knapik-Czajka M, et al. Cerebrospinal fluid biomarker signature in Alzheimer's disease neuroimaging initiative subjects. *Ann Neurol*. 2009; 65(4): 403–413.
29. Schuff N, Woerner N, Boreta L, et al. MRI of hippocampal volume loss in early Alzheimer's disease in relation to ApoE genotype and biomarkers. *Brain*. 2009; 132(Pt 4): 1067–1077.
30. Dubal DB. Sex difference in Alzheimer's disease: an updated, balanced and emerging perspective on differing vulnerabilities. *Handbook Clin Neurol*. 2020; 175: 261–273.
31. Mormino EC, Papp KV, Rentz DM, et al. Early and late change on the preclinical Alzheimer's cognitive composite in clinically normal older individuals with elevated amyloid β. *Alzheimers Dement*. 2017; 13(9): 1004–1012.
32. Morris JC, Schindler SE, McCue LM, et al. Assessment of racial disparities in biomarkers for Alzheimer disease. *JAMA Neurol*. 2019; 76(3): 264–273.
33. Beydoun MA, Weiss J, Beydoun HA, et al. Race, APOE genotypes, and cognitive decline among middle-aged urban adults. *Alzheimers Res Ther*. 2021; 13(1): 120.

SUPPORTING INFORMATION

Additional supporting information may be found in the online version of the article at the publisher's website.

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