Effect of EPHA1 Genetic Variation on Cerebrospinal Fluid and Neuroimaging Biomarkers in Healthy, Mild Cognitive Impairment and Alzheimer's Disease Cohorts

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Abstract. Ephrin type-A receptor 1 (EPHA1) (11771145) was documented to be one of the most strongly associated locus with Alzheimer's disease (AD) in a recent meta-analysis of five genome wide association studies. However, its contribution to the pathogenesis of AD remains unclear to date. Here, we addressed the role of EPHA1 in AD by investigating the influence of EPHA1 on cerebrospinal fluid and neuroimaging biomarkers in three clinical stages from the Alzheimer's Disease Neuroimaging Initiative (ADNI) database. We did not detect significant association of EPHA1 with amyloid-β deposition or tau protein. However, the A-allele in the mild cognitive impairment group remarkably prevented hippocampal atrophy (partial correlation coefficient 2.812, 95% CI 0.651 to 4.973) at two-year follow-up. Additionally, AD subjects with the A-allele displayed less atrophy and greater cerebral metabolic rate for glucose (CMRgl) in the right lateral occipitotemporal gyrus (volume: partial correlation coefficient 540.10, 95% CI 247.26 to 832.95; CMRgl: partial correlation coefficient 0.056, 95% CI 0.024 to 0.087) and inferior temporal gyrus (volume: partial correlation coefficient 327.98, 95% CI 11.65 to 644.31; CMRgl: partial correlation coefficient 0.055, 95% CI 0.019 to 0.091) at baseline. This study suggests EPHA1 (rs11771145) interferes with the pathological alteration of the hippocampus and the lateral occipitotemporal and inferior temporal gyri throughout the AD process, leading to a lower risk of AD. However, the limited sample size and follow-up as well as the diversity across ethnicities precluded explanation of these findings.

Keywords: Alzheimer's disease, Alzheimer's Disease Neuroimaging Initiative (ADNI), biomarker, cerebrospinal fluid, Ephrin type-A receptor 1 (EPHA1), neuroimaging

INTRODUCTION

Recently, the National Institute on Aging-Alzheimer's Association workgroup has published the revised guidelines on diagnosis of Alzheimer's disease (AD) dementia [1]. The revised recommendations suggested that the histological pathology of AD may be found across a broad clinical spectrum (normal cognition (NC), mild cognitive impairment (MCI), and dementia due to AD), and might be detected by specific abnormality on various biomarkers. The cerebrospinal fluid (CSF) biomarkers, amyloid-β ($A\beta_{42}$) and tau protein (both total tau and phosphorylated tau), display significant lower and higher levels, respectively, in the AD process; and AD patients present with disproportionate atrophy in medial, basal, and lateral temporal lobe, and medial parietal cortex on structural magnetic resonance imaging (MRI), decreased18-fluorodeoxyglucose (FDG) uptake on position emission tomography (PET) in temporoparietal cortex, and positive amyloid imaging on Pittsburgh Compound B position emission tomography (PiB-PET) [2, 3]. In addition, the guidelines admitted the role of genetic factors in the development of AD. To date, only the APOE genetic variant was well replicated to be associated with AD worldwide [4], and strongly related to the CSF and neuroimaging biomarkers in the AD process [5–10]; however, it is estimated that variation at the APOE locus may explain 50% or less of late-onset AD risk [4], indicating that additional risk loci affecting late-onset AD exist.

Ephrin type-A receptor 1 (EPHA1) has been identified and confirmed as an AD susceptibility gene in genome wide association studies in Caucasians [11–13]. Recently, EPHA1 (11771145) was demonstrated to be the most strongly associated locus with AD at EPHA1 in a meta-analysis of five published genome wide association studies in individuals of European ancestry [14]. However, the role of EPHA1 in the pathophysiological process of AD remains unclear. This study was designed to explore the involvement of EPHA1 in the development and progression of AD by investigating the influence of EPHA1 on CSF and neuroimaging biomarkers in the three different clinical stages (NC, MCI, and dementia due to AD) from the Alzheimer's Disease Neuroimaging Initiative (ADNI) dataset.

MATERIALS AND METHODS

ADNI dataset

The ADNI is a large, multicenter, longitudinal neuroimaging study, launched in 2003 by the National Institute on Aging, the National Institute of Biomedical Imaging and Bioengineering, the Food and Drug Administration, private pharmaceutical companies, and nonprofit organizations. The initial goal of ADNI was to recruit 800 subjects but the ADNI has been followed by ADNI-GO and ADNI-2. To date, the three protocols have recruited over 1,500 adults, ages 55 to 90, to participate in the research, consisting of cognitively normal older individuals, people with early or late MCI, and people with early AD. The study was approved by the institutional review boards of all participating centers, and written informed consent was obtained from all participants or authorized representatives after extensive description of the ADNI according to the 1975 Declaration of Helsinki.

Subjects

The data used in this study were obtained from the ADNI database (http://adni.loni.usc.edu) Inclusion criteria for AD subjects included National Institute of Neurological and Communication Disorders/Alzheimer's Disease and Related Disorders Association criteria for probable AD with a Mini-Mental State Examination (MMSE) score between 20 and 26, a global Clinical Dementia Rating (CDR) of 0.5 or 1, a sum-of-boxes CDR of 1.0 to 9.0. All amnestic MCI subjects fulfilled a MMSE score of 24 to 30 and a Memory Box score of at least 0.5. On the other hand, the subjects who had any serious neurological disease other than possible AD, any history of brain lesions or head trauma, or psychoactive medication use (including antidepressants, neuroleptics, chronic anxiolytics, or sedative hypnotics) were not be recruited for this study. Other details on the ADNI cohort can be found online (http://adni.loni.usc.edu/).

The basic information of ADNI-1 subjects ($n = 665$) in the present analysis was downloaded from the ADNI web site in 2013.

Genetic data

ADNI applied the Human 610-Quad Bead Chip (Illumina, Inc., San Diego, CA) included 620,901 SNP and CNV markers to conduct genotyping [15]. Bead Studio 3.2 software and Genome Studio v2009.1 (Illumina) were used to generate SNP genotypes from bead intensity data successively. After sample verification and quality control bioinformatics, the genotype data for ADNI-1 participants was uploaded to the ADNI website (http://www.loni.usc.edu/ADNI) In addition, the widely used PLINK data format

(http://pngu.mgh.harvard.edu/∼purcell/plink/) has been provided to facilitate other groups. Here we extracted the genetic data of EPHA1 (rs11771145) ($n = 757$) from the PLINK data.

Cerebrospinal fluid data

CSF samples were collected into collection tubes, and then transferred into polypropylene transfer tubes followed by freezing on dry ice within 1 h after collection, and transported overnight to the ADNI Biomarker Core laboratory at the University of Pennsylvania Medical Center in dry ice. Preparation of aliquots (0.5 ml) from these samples was done after thawing (1 h) at room temperature and gentle mixing. The aliquots were stored in bar code–labeled polypropylene vials at $-80°$C. CSF proteins, such as $A\beta_{1-42}$, total tau (t-tau), and phosphorylated tau (p-tau$_{181p}$), were calculated in every CSF baseline aliquots on the multiplex xMAP Luminex platform (Luminex Corp, Austin, TX) with Innogenetics (INNO-BIA AlzBio3; Ghent, Belgium; for research use only reagents) immunoassay kit-based reagents. Full details of this combination of immunoassay reagents and analytical platform were described elsewhere [16]. Finally, a total of 315 participants with genetic and other information were included in CSF data analysis from the ADNI website.

Neuroimaging data

These neuroimaging data, such as regional PiB retention on PET, regional volume on MRI, and cerebral metabolic rate for glucose (CMRgl) on FDG-PET were downloaded from the ADNI dataset. The detailed description on acquiring and treating imaging data from ADNI can be found in other papers [17]. In this study we used the regions of interest (ROIs) analysis to calculate the relationship between EPHA1 and AD. Based on the revised guidelines [1], we mainly selected temporal cortex as the strongly associated regions with AD. We adopted normalized volume and CMRgl in the regional volume and metabolism analysis, and PiB retention was quantified as the ratio of uptake in cortical ROIs to the uptake in the cerebellum. There were 71 subjects (NC = 16, MCI = 47, AD = 8) with genetic and other information in PiB retention analysis, 610 (NC = 190, MCI = 288, AD = 132) in regional volume, and 306 (NC = 88, MCI = 151, AD = 67) in metabolism analysis at baseline. Furthermore, we compared the changed values on regional PiB retention ($n = 29$) and hippocampal volume ($n = 380$) in the follow-up study of two years.

Statistical analysis

Hardy–Weinberg equilibrium was tested using a chi-square test. Differences in continuous variables were examined using one-way analysis of variance (ANOVA), and categorical data were tested using Spearman's correlation analysis. These data were stratified into three groups (NC, MCI, and AD) based on diagnosis to the test the influence of EPHA1 on the three clinical stage, respectively. Furthermore, a multiple linear regression model which considered age, gender, and ApoE ε4 status as covariates was used to estimate coefficients and the 95% confidence interval (CI) for testing possible correlation between various phenotypes and EPHA1genotypes. All statistical analyses were performed by SSPS 11.5 for Windows. The statistical power of the study was calculated by PASS 11 software. The criterion for significant difference was $p < 0.05$.

RESULTS

Demographic, clinical, and cognitive characteristics

Distributions of these genotypes for EPHA1 and APOE ε4 were under the Hardy–Weinberg equilibrium in NC, MCI, and AD groups (Supplementary Table 1). Characteristics of the study sample are presented in Table 1. There was no significant difference on age ($p = 0.05$) and gender ($p = 0.7$) between the three clinical stages. As expected, the ε4 allele of APOE gene substantially increased the risk of AD with a dose effects, and the cognitive scores on various neuropsychological scales were considerably different across three groups (NC, MCI, and AD). Furthermore, AD patients had marked atrophy in hippocampus respect to MCI and NC individuals ($p < 0.01$). However, we did not detect the association of EPHA1 with AD in this study ($p = 0.37$).

CSF biomarkers and EPHA1

In this study we compared the levels of CSF proteins, such as $A\beta_{42}$, t-tau, and p-tau$_{181}$ between GG, AG, and AA groups at baseline in the three different clinical stages (NC, MCI, and AD). Finally, neither $A\beta_{42}$ nor tau protein was demonstrated to be associated with the genetic variation in EPHA1 in ANOVA in the three samples (Table 2). Moreover, we investigated the relationship between the CSF proteins and EPHA1 genotypes in multiple linear regression analysis

Table 1
Demographic characteristics and baseline neuropsychological scores of the subjects[a]

Characters		NC		MCI		AD	p^{b}
Age (years)	193	75.44 ± 4.93	293	73.95 ± 7.31	133	74.96 ± 7.49	0.05
Gender (male/female)	193	104/89	293	189/104	133	71/62	0.7
ApoE ε4 (0/1/2)	193	144/46/3	293	125/132/36	133	38/68/27	<0.01
CDRSB (scores)	193	0.03 ± 0.12	293	1.57 ± 0.83	133	4.29 ± 1.66	<0.01
ADAS11 (scores)	193	6.15 ± 2.89	293	11.65 ± 4.48	133	18.54 ± 6.04	<0.01
ADAS13 (scores)	193	9.44 ± 4.18	291	18.8 ± 6.29	130	28.86 ± 7.37	<0.01
MMSE (scores)	193	29.07 ± 1.01	293	26.97 ± 1.79	133	23.17 ± 2.06	<0.01
RAVLT total (scores)	192	43.46 ± 8.98	292	30.52 ± 9.18	132	23.41 ± 7.03	<0.01
FAQ (scores)	193	0.13 ± 0.62	291	3.82 ± 4.37	133	12.88 ± 6.84	<0.01
Hippocampus (mm^3)	193	7259.8 ± 895.4	293	6379.9 ± 1069.2	133	5602.5 ± 1059.2	<0.01
Genotype (GG/AG/AA)	193	87/80/26	293	114/144/35	133	65/59/9	0.37

NC, normal cognition; MCI, mild cognitive impairment; AD, Alzheimer's disease; CDRSB, Clinical Dementia Rating scale sum of boxes; ADAS, Alzheimer's disease Assessment Scale; MMSE, Mini-Mental State Exam; RAVLT, Rey Auditory Verbal Learning Test; FAQ, Functional Activities Questionnaire. [a]Data are given as mean $\pm$ standard deviation unless otherwise indicated. [b]p values for continuous variables are from one-way ANOVA. p values for categorical data are from Spearman's correlation analysis.

Table 2
Cerebrospinal fluid (CSF) biomarkers concentrations in subjects

Biomarker	GG		AG		AA		ANOVA	Linear
	n	Mean $\pm$ SD	n	Mean $\pm$ SD	n	Mean $\pm$ SD	p value	p value
NC group								
Aβ_{42} (pg/ml)	46	209.39 ± 52.27	35	209.06 ± 60.24	15	204.27 ± 58.28	0.95	ns
t-tau (pg/ml)	46	68.07 ± 30.51	35	65.77 ± 27.55	15	76.4 ± 29.62	0.5	ns
p-tau$_{181P}$ (pg/ml)	46	23.63 ± 13.07	35	22.66 ± 12.1	15	28.27 ± 16.78	0.39	ns
MCI group								
Aβ_{42} (pg/ml)	53	172.55 ± 56.71	79	156.85 ± 52.45	14	154.71 ± 48.48	0.22	ns
t-tau (pg/ml)	52	92.69 ± 49.12	77	110.94 ± 71.38	14	111.93 ± 47.68	0.24	ns
p-tau$_{181P}$ (pg/ml)	54	31.81 ± 15.26	79	38.08 ± 19.62	14	37.71 ± 15.57	0.13	ns
AD group								
Aβ_{42} (pg/ml)	35	142.69 ± 36.98	32	143.47 ± 36	5	160.6 ± 71.31	0.63	ns
t-tau (pg/ml)	34	110.26 ± 47.1	32	138.84 ± 66.43	5	115.2 ± 68.22	0.14	ns
p-tau$_{181P}$ (pg/ml)	35	38.29 ± 15.09	32	47.31 ± 23.63	5	37 ± 27.18	0.16	ns

AD, Alzheimer's disease; MCI, mild cognitive impairment; NC, normal cognition; N, number; SD, standard deviation; ANOVA, one-way analysis of variance; Linear, multiple linear regression analysis that considered age, gender, and ApoE ε4 allele as covariates; ns, no significance.

considering age, gender, clinical ratings, and ApoE ε4 allele as concomitant variables; likewise, these CSF biomarkers were not indicated to be linearly correlated with EPHA1 genotypes in the three different stages.

Neuroimaging markers and EPHA1

Based on the recent guidelines, temporal cortex was suggested as the most common brain region associated with AD, and was treated as our ROI in our study. Finally, we did not observe significant differences on PiB retention in the NC or AD groups at baseline, but in the MCI cohort, medial temporal (GG: 1.673 ± 0.322, AG: 1.819 ± 0.357, AA: 1.085 ± 0.021, $p = 0.014$) and lateral temporal cortexes (GG: 1.811 ± 0.361, AG: 1.907 ± 0.371, AA: 1.14 ± 0.071, $p = 0.023$) displayed marked differences on retention values between the three genotypes in ANOVA; however, the difference disappeared in multiple linear regression after control-

ling for age, gender, and ApoE ε4 allele (Table 3a). At the two-year follow-up, both brain regions did not show statistically significant differences in PiB uptake values in one-way ANOVA, nor in multiple linear regression analysis, which considered age, gender, and ApoE ε4 allele as covariates (Table 3b).

In regional volume analysis, AD patients with AA and AG genotypes had significantly greater volume than those with GG genotypes in right lateral occipitotemporal (AA: 8478.8 ± 894.9 mm^3, AG: 8374.6 ± 1231.1 mm^3, GG: 7658.4 ± 1337.7 mm^3, $p = 0.002$) and inferior temporal gyrus (AA: 7189.1 ± 661.6 mm^3, AG: 6261.4 ± 1109.1 mm^3, GG: 6068.5 ± 1320.2 mm^3, $p = 0.033$) in ANOVA at baseline (Fig. 1a). Based on the regional volume of the three genotypes, our sample size had a more than 90% power to detect a significant difference of 0.05 for right lateral occipitotemporal and inferior temporal gyrus, respectively. Furthermore, the volume of

Table 3a
Brain regions and regional changed PiB retention values on PET at baseline

Regions	GG		AG		AA		ANOVA	Linear
	n	Mean ± SD	n	Mean ± SD	N	Mean ± SD	p	p
NC group								
Medial temporal cortex	6	1.162 ± 0.064	9	1.126 ± 0.076	1	1.14	0.644	ns
Lateral temporal cortex	6	1.515 ± 0.392	9	1.406 ± 0.278	1	1.57	0.77	ns
MCI group								
Medial temporal cortex	25	1.673 ± 0.322	20	1.819 ± 0.357	2	1.085 ± 0.021	0.014	0.919
Lateral temporal cortex	25	1.811 ± 0.361	20	1.907 ± 0.371	2	1.14 ± 0.071	0.023	0.630
AD group								
Medial temporal cortex	3	1.023 ± 0.18	5	1.07 ± 0.103	–	–	0.649	ns
Lateral temporal cortex	3	1.817 ± 0.131	5	1.696 ± 0.41	–	–	0.647	ns

Table 3b
Brain regions and regional changed PiB retention values on PET over two years

Regions	GG		AG		AA		ANOVA	Linear
	n	Mean ± SD	n	Mean ± SD	n	Mean ± SD	p	p
NC group								
Medial temporal cortex	4	−0.06 ± 0.039	6	−0.03 ± 0.057	–	–	0.387	ns
Lateral temporal cortex	4	−0.073 ± 0.12	6	0.02 ± 0.04	–	–	0.112	ns
MCI group								
Mesial temporal cortex	8	−0.02 ± 0.083	9	0.061 ± 0.115	2	−0.015 ± 0.078	0.248	ns
Lateral temporal cortex	8	0.01 ± 0.131	9	0.126 ± 0.109	2	0.03 ± 0	0.141	ns

NC, normal cognition; MCI, mild cognitive impairment; AD, Alzheimer's disease; *N*, number; SD, standard deviation; ANOVA, one-way analysis of variance; Linear, multiple linear regression analysis that considered age, gender, and ApoE ε4 allele as covariates; ns, no significance.
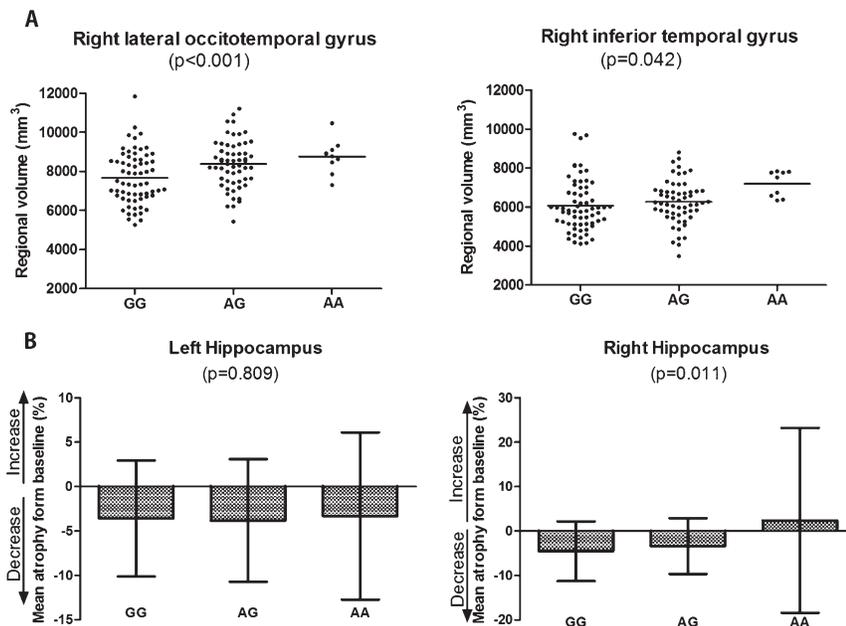
Fig. 1. Regional volume analyses on MRI in subjects. A) Significant brain locations and volume values on MRI at baseline in AD group. B) Hippocampus and percentage of atrophy on MRI over two years in MCI group. MRI, magnetic resonance imaging; AD, Alzheimer's disease; MCI, mild cognitive impairment; *p* values were from multiple linear regression analysis that considered age, gender, and ApoE ε4 allele as covariates.
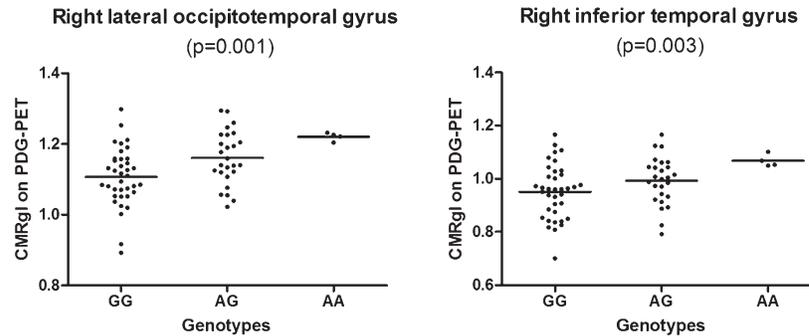
Fig. 2. Significant brain locations and CMRgl values on PDG-PET at baseline in AD group. CMRgl, cerebral metabolic rate for glucose; PDG-PET, fluorodeoxyglucose on position emission tomography; AD, Alzheimer's disease; *p* values were from multiple linear regression analysis that considered age, gender, and ApoE ε4 allele as covariates.

right lateral occipitotemporal (partial correlation coefficient 540.10, 95% CI 247.26 to 832.95, *p* < 0.001) and inferior temporal gyrus (partial correlation coefficient 327.98, 95% CI 11.65 to 644.31, *p* = 0.042) in the AD group showed positive linear association with the dose of A-allele in multiple linear regression model after adjusting for age, gender, and ApoE ε4 allele. In addition, we detected that right hippocampus of A-allele carriers in the MCI subjects showed significant improvement on the percentage of atrophy in ANOVA (*p* = 0.014) and multiple linear regression (partial correlation coefficient 2.812, 95% CI 0.651 to 4.973, *p* = 0.011) controlling for age, gender, and ApoE ε4 allele (Fig. 1b).

With respect to regional CMRgl calculation on FDG-PET, we observed that AD subjects carrying the A-allele had significantly higher CMRgl in the right lateral occipitotemporal (AA: 1.22 ± 0.012, AG: 1.16 ± 0.076, GG: 1.106 ± 0.082, *p* = 0.003) and inferior temporal gyrus (AA: 1.068 ± 0.023, AG: 0.993 ± 0.089, GG: 0.952 ± 0.102, *p* = 0.035) in one-way ANOVA, and the statistical power of the test was at least 90%. In multivariate variance analyses, CMRgl in the right lateral occipitotemporal (partial correlation coefficient 0.056, 95% CI 0.024 to 0.087, *p* = 0.001) and inferior temporal gyrus (partial correlation coefficient 0.055, 95% CI 0.019 to 0.091, *p* = 0.003) were also positively associated with the A-allele of EPHA1 genotypes (Fig. 2). However, we did not observe significant difference on other regional CMRgl in the other two groups (NC or MCI) at baseline.

DISCUSSION

This present study investigated the effects of EPHA1 genotypes (GG, AG, and AA) on CSF and neuroimag-

ing biomarkers in three clinical stages (NC, MCI, and AD). We observed that the EPHA1 gene mutation did not affect the levels of Aβ_{42} in CSF nor the Aβ_{42} deposition in PiB-PET in the three periods. On the other hand, the A-allele did not increase or decrease the concentration of t-tau or p-tau$_{181p}$ in CSF analysis. However, AD patients with the A-allele displayed both less loss and higher cerebral metabolic rate for glucose (CMRgl) in the right lateral occipitotemporal and inferior temporal gyri in neuroimaging examination at baseline. Furthermore, the A-allele in MCI subjects remarkably prevented right hippocampal atrophy at the two-year follow-up. To our knowledge, our investigation was the first study to explore the role of EPHA1 rs11771145 in CSF and neuroimaging phenotypes and detected that EPHA1 was associated with the pathological and metabolic alteration of right hippocampus and occipitotemporal and inferior temporal gyri.

EPHA1, EPH receptor A1 (ephrin type-A receptor 1), is one member of the ephrin receptor subfamily from the protein-tyrosine kinase family [18]. EPH and EPH-related receptors participate in mediating developmental events, particularly in the nervous system [19]. Receptors in the EPH subfamily typically have a single kinase domain and an extracellular region containing a Cys-rich domain and 2 fibronectin type III repeats [18]. It has been documented that EPHA1 is implicated in immune system function [20], synaptic dysfunction [21, 22], and cell membrane processes [23], which are all related to the occurrence of AD [24]. Thus, we investigated the mechanism underlying the interaction between EPHA1 and AD in this study, and found that right inferior temporal and right lateral occipitotemporal gyri displayed both less loss and higher CMRgl in A-allele carriers in the AD group at baseline, and A-allele significantly inhibited right

hippocampal atrophy in the MCI group at follow-up, suggesting that EHPHA1 plays an important role in AD-related neurodegenerative processes.

Abnormal accumulation of $A\beta_{42}$ in neurotic fibrillar plaques occurs firstly throughout the AD pathophysiological process, which can be detected by the decreased levels of $A\beta_{42}$ in CSF and positive amyloid imaging in PiB-PET examination. In the present study, we did not observe a significant relationship between $A\beta_{42}$ levels and EPHA1 in CSF, nor the correlation between PiB uptake values and EPHA1 in PiB-PET analysis. The following hallmark was the presence of tau-mediated neuronal injury and dysfunction after the pathological $A\beta_{42}$ deposition, which manifests as increased tau levels (both t-tau and p-tau$_{181p}$) in CSF examination. Likewise, these findings did not support the relationship between EPHA1 and tau protein. However, the lower power from the small sample size limited the final interpretation of the relationship between $A\beta_{42}$ deposition, tau, and EPHA1.

Next, AD-related pathologic degeneration arises stealthily, which could be determined by the disproportionate atrophy in temporal cortex on MRI, and decreased CMRgl on PET in temporo-parietal cortex. Here, we selected temporal lobe as the brain region most affected by AD, and detected that lateral occipitotemporal and inferior temporal gyrus were significantly associated with the A-allele at EPHA1 gene in AD subjects. It has been demonstrated the lateral occipitomporal gyrus has a higher number of neurofibrillary tangles in AD patients than that in normal cognitive individuals [25]. Besides, inferior temporal lobe is one of the most strongly associated brain regions with metabolic and cognitive decline indicative of AD dementia [26], which displays $A\beta_{42}$ deposition, neurofibrillary tangles, and synaptic loss in AD pathology [27, 28], and has been demonstrated to be associated with AD in PET examination [28]. Furthermore, we tested the influence of EPHA1 on the volume of hippocampus (the most strongly associated structure marker with AD) in the two-year follow-up study, and the A-allele in MCI subjects markedly prevented the atrophy in right hippocampus. Therefore, it is possible that EPHA1 (rs11771145) could affect the histological pathology and glucose metabolism of hippocampus and lateral occipitomporal and inferior temporal gyri from participating in the AD pathological process, leading to influence the development and progression of AD.

There are several potential limitations in this study. First, the ADNI-1 sample was limited in sample size when considering longitudinal imaging data of specific genotypes and diagnosis subgroups, and it will still be necessary to replicate these findings in a larger dataset. Second, a follow-up of two years may be too short to detect the significant influence of EPHA1 on the AD process, and an increase of follow-up is useful to detect a stronger influence of EPHA1 genetic variation. Moreover, except hippocampus and temporal lobe, other regions, such as anterior cingulate, precuneus cortex, and frontal, parietal and occipital cortexes were indicated to be associated with AD in recent studies. Finally, our sample was restricted to Caucasians to avoid genetics stratification across ethnicities. EPHA1 (rs11771145), however, has different frequencies and polymorphisms in different population; thus, our results cannot represent other ethnicities, and replications in other populations are imperative.

Overall, our study demonstrated that the EPHA1 (rs11771145) genetic variant is involved in the structural and functional modification of hippocampus and lateral occipitotemporal and inferior temporal gyri throughout the AD physiopathological process, and decreases the risk of AD by inhibiting the AD-related pathologic formation. However, several limitations precluded the explanation of these findings, and it is necessary to explore the effect of EPHA1 on AD in a larger sample, with longer follow-up and other ethnicities.

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DISCLOSURE STATEMENT

Authors' disclosures available online (http://www.j-alz.com/disclosures/view.php?id=2495)

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