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Apolipoprotein E ε4 and ε3 alleles associate with cerebrospinal fluid tau and cognition in the presence of amyloid-β in mild cognitive impairment but not in Alzheimer's disease

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Apolipoprotein E is the most well-established genetic risk factor for Alzheimer's disease. However, the associations of apolipoprotein E with tau pathology and cognition remain controversial. The research checks the hypothesis that the relationships between apolipoprotein E alleles and cerebrospinal fluid tau and cognition differ in persons with and without significant amyloid-β deposition. We divided 1119 subjects into cognitively normal (n = 275), mild cognitive impairment (n = 629), and Alzheimer's disease (n = 215), and these subjects were from the Alzheimer's Disease Neuroimaging Initiative database. Linear regression models were used to compare the relationships of apolipoprotein E alleles with cerebrospinal fluid tau and cognition in persons with significant amyloid-β deposition relative to individuals without significant amyloid-β deposition. The associations of apolipoprotein E ε4 and ε3 with total tau (T-tau), phosphorylated tau (P-tau), and Alzheimer's disease assessment scale was significantly substantial among participants with significant amyloid-β deposition. Stratified analyses showed that apolipoprotein E ε4 related to increased concentrations of T-tau, P-tau, and Alzheimer's disease assessment scale and apolipoprotein E ε3 associated with decreased concentrations of T-tau, P-tau, and Alzheimer's disease assessment scale in mild cognitive impairment participants with significant amyloid-β deposition, but not in Alzheimer's disease. Our study shows that the presence of apolipoprotein E ε4 and ε3 alleles is related to tau pathology and cognitive impairment in the presence of amyloid-β in mild cognitive impairment, but not in Alzheimer's disease. This work indirectly provides additional evidence that apolipoprotein E and amyloid-β may not have a role in modulating clinical Alzheimer's disease, and apolipoprotein E ε3 may be supposed to be protective to mild cognitive impairment.

Keywords

Alzheimer's disease; Amyloid-β; Apolipoprotein E; Mild cognitive impairment; Tau

1. Introduction

Alzheimer's disease (AD) is a slowly progressive disease that leads to the degeneration of brain cells. It is the major type of dementia, characterized by the decline of thinking ability and independence of daily activities [1]. On the other hand, mild cognitive impairment (MCI) is a disorder in which subjects exhibit objectively cognitive dysfunction and their ability to engage in activities of daily living is minimally affected [2, 3]. The apolipoprotein E (*APOE*) is a central regulator of cholesterol and is closely related to AD pathology due to the homeostasis of lipid and protein [4, 5]. The *APOE* gene has three alleles (ε4, ε3, and ε2) responsible for three major *APOE* subtypes (*APOE4, APOE3, and APOE2*) [6]. The *APOE* ε4 allele is the most common genetic risk factor for AD [7], and it is related to increased production of an amyloid-β (Aβ) [8] other than reduced clearance of cerebral Aβ compared to ε2 and ε3 alleles [9, 10]. Consequently, subjects with *APOE* ε4 demonstrate increased cerebral Aβ deposition [11], and *APOE* ε4 carriers have amyloid positive onset earlier than non-carriers [12]. In contrast, other subtypes of *APOE* are supposed to be protective (*APOE2*) or neutral (*APOE3*) for AD risk [13–15].

Tau pathology is a crucial aspect of AD, and the tau burden can predict cognitive decline in AD [16]. MCI individuals with high tau levels show an increased risk of cognitive decline [17]. However, the relationship between *APOE* and tau pathology is less clear and controversial. [18] has reported a significant physiological link between cerebrospinal fluid (CSF) levels of *APOE* and CSF tau in neurologically healthy, cognitively intact individuals. In contrast [19], other studies have reported no effect of *APOE* ε2 or ε4 on CSF tau in cognitively normal aging. Post-mortem evaluations suggested that

APOE ε2 and ε4 alleles were not related to paired helical filament (PHF) tau tangles in the absence of Aβ [20]. However, there was evidence that *APOE* ε4 significantly influenced tau-mediated neurodegeneration independently of Aβ in a mouse model of tauopathy [21]. Recent studies have shown that the ε4+ group has a higher rate of tau accumulation, and the enhanced effect of *APOE* ε4 on tau accumulation still exists after adjusting the Aβ load in the cortex [22]. So far, there is no study on the relationship between *APOE* ε3 and tau pathology. In addition, there were no studies that explored the effect of *APOE* alleles on tau as measured by CSF dependently or independently of Aβ in a group of individuals that spans the spectrum of cognition.

Similarly, the relationship between cognition and *APOE* allele status is also controversial. Previous researches reported a positive association [23–29]. These findings were generally interpreted to suggest that the influences of *APOE* ε4 on late-life cognitive impairment were mediated by the cascade of *APOE* that was *APOE* ε4 led Aβ deposition, then tau tangles, finally cognitive dysfunction [30]. However, other studies showed no relationship between cognition and *APOE* ε4 [31–35]. There were few studies on the relationship between cognition and *APOE* ε2 and ε3 in MCI and AD.

Is there a new pathological cascade that explains the cognitive impairment in the AD continuum? Therefore, the associations of *APOE* alleles with tau and cognition and whether Aβ mediates these associations need to be further elucidated. In this article, we test hypothesis that the associations of *APOE* alleles status with CSF tau and cognitive function differ according to the presence and absence of Aβ deposition.

2. Materials and methods

2.1 Database description and participants

Data used in this article were from the Alzheimer's Disease Neuroimaging Initiative (ADNI) database (adni.loni.usc.edu) [36].

We selected 1119 participants who had completed lumbar puncture, genotyping for *APOE* allele status, Alzheimer's disease assessment scale (ADAS)-cog, Mini-Mental State Examination (MMSE), and Clinical Dementia Rating scale (CDR). Selected participants were divided into cognitively normal (CN, n = 275), MCI (n = 629), and AD (n = 215). The criteria for CN included an MMSE score equal to or greater than 24 and a CDR score of 0 [37]. The criteria for MCI were subjects with an MMSE score equal to or greater than 24 and a CDR of 0.5, preservation of activities of daily living, and an absence of other neuropsychiatric diseases [38]. Except for the NINCDS/ADRDA standards, the MMSE score of AD patients ranged from 20 to 26, and the CDR was 0.5 or 1.0. [39].

2.2 Standard protocol approvals, registrations, and patient consents

The Institutional Review Boards approved the ADNI study of all the participating institutions. Informed written consent was obtained from all participants at every center.

2.3 APOE Genotyping

Subjects with at least one ε4 allele are called ε4 carriers [20]. Individuals who have two ε3 alleles are considered as ε3 carriers. Participants with one ε2 allele and one ε3 allele or two ε2 alleles are considered as ε2 carriers [40]. All *APOE* genotyping data used were from ADNI files "APOERES.csv" (accessed November 2020).

2.4 CSF analyses

As mentioned earlier, Aβ42, total-tau (T-tau), and phosphorylated-tau (P-tau) at threonine 181 in CSF were measured by using the Innogenetics INNO-BIA AlzBio3 immunoassay reagents and multiplex xMAP Luminex platform [41]. Subjects were classified as with significant Aβ deposition (Aβ positive or Aβ+) or without significant Aβ deposition (Aβ negative or Aβ-) using a previously established cut-off of CSF Aβ42 (192 pg/mL) [41]. All CSF data used were from the ADNI files "UPENNBIOMK5-8.csv" and "FAGAN-LAB_07_15_2015.csv" (accessed November 2020).

2.5 Statistical methods

Chi-square analyses were used to test the difference of *APOE* genotypes among the groups; all probability p values < 0.05 were reported. Differences between *APOE* ε4, ε3, and ε2 carriers and noncarriers in every diagnostic group were tested by using the chi-square analyses for gender and Aβ status (Aβ- or Aβ+), and Mann-Whitney U test for age, education, Aβ42, T-tau, P-tau, and ADAS-cog. Bonferroni correction was used for multiple comparison correction.

To analyze the differences in the association of *APOE* ε4 with T-tau, P-tau, and ADAS-cog in individuals with and without significant Aβ deposition, we fitted linear regression models with an interaction term between *APOE* ε4 and Aβ status. Then we conducted stratified analyses regressing *APOE* ε4 status on T-tau, P-tau, and ADAS-cog in individuals with and without significant Aβ deposition. Finally, we also conducted stratified analyses regressing *APOE* ε4 status on T-tau, P-tau, and ADAS-cog for CN, MCI, and AD, respectively. All models adjusted for sex, age, and education. Similar analyses were performed for *APOE* ε3 and ε2 genotypes. In these models, variables were log-transformed to fit a normal distribution. Statistical significance was defined as $p < 0.05$. Bonferroni correction was used for multiple comparison correction. All statistics were done using R (v. 3.4.2) and SPSS version 20.

3. Results

3.1 Demographic results

Demographic and clinical characteristics of subjects by diagnosis and *APOE* allele status are shown in Tables 1,2,3. There were no differences in age, sex, and education among the groups. *APOE* ε4 carriership was more common in MCI and AD than in CN ($p < 0.001$ for both) and in AD than in MCI ($p < 0.001$). *APOE* ε4 was present in 42.2% of individuals with significant Aβ deposition and only 6.0% of individuals without significant Aβ deposition in all participants ($p <$

Table 1. Demographic and clinical characteristics of *APOE* ε4 carriers and noncarriers.

Characteristics	CN		MCI		AD		All	
	ε4-	ε4+	ε4-	ε4+	ε4-	ε4+	ε4-	ε4+
N (n %)	204 (74.2%)	71 (25.8%)	318 (50.6%)	311 (49.4%)	58 (27.0%)	157 (73.0%)	580 (51.8%)	539 (48.2%)
Age (years)	74.6 (5.7)	73.5 (6.6)	73.3 (7.8)	71.5 (7.1)	76.4 (9.0)	73.9 (7.6)	74.2 (7.4)	72.5 (7.3)
Sex (F %)	103 (50.5%)	35 (49.3%)	129 (40.6%)	130 (41.8%)	23 (39.7%)	68 (43.3%)	255 (44.0%)	233 (43.1%)
Education (years)	16.3 (2.6)	16.0 (2.9)	16.2 (2.7)	16.0 (2.8)	16.0 (2.9)	15.2 (3.0)	16.2 (2.7)	15.8 (2.9)
Aβ42 (pg/mL)	210.5 (48.0)	167.5 (53.5)	194.6 (51.9)	147.5 (42.3)	137.8 (23.0)	127.5 (23.1)	194.5 (52.4)	144.3 (41.6)
T-tau (pg/mL)	66.3 (30.4)	75.2 (35.9)	73.1 (43.5)	110.0 (60.5)	134.8 (60.9)	130.3 (61.7)	76.9 (46.0)	111.2 (60.5)
P-tau (pg/mL)	28.1 (14.8)	37.0 (23.2)	32.3 (20.0)	46.5 (24.5)	53.2 (29.8)	53.5 (30.5)	32.9 (19.8)	47.3 (26.7)
ADAS-cog	6.0 (3.0)	6.4 (3.2)	9.1 (4.3)	11.0 (4.8)	19.7 (7.0)	19.5 (6.7)	9.4 (6.0)	12.8 (6.9)
Aβ- (n %)	136 (49.5%)	23 (8.4%)	173 (27.5%)	44 (7.0%)	0 (0.0%)	0 (0.0%)	309 (27.6%)	67 (6.0%)
Aβ+ (n %)	68 (24.7%)	48 (17.5%)	145 (23.1%)	267 (42.4%)	58 (27.0%)	157 (73.0%)	271 (24.2%)	472 (42.2%)

The measured data are represented by mean and standard deviation. Abbreviations: Aβ-, without significant Aβ deposition; Aβ+, with significant Aβ deposition; CN, cognitively normal; MCI, mild cognitive impairment; AD, Alzheimer's disease; ADAS-cog, Alzheimer's disease assessment scale-cog.

Table 2. Demographic and clinical characteristics of *APOE* ε3 carriers and noncarriers.

Characteristics	CN		MCI		AD		All	
	ε3-	ε3+	ε3-	ε3+	ε3-	ε3+	ε3-	ε3+
N (n %)	111 (40.4%)	164 (59.6%)	349 (55.5%)	280 (44.5%)	162 (75.3%)	53 (24.7%)	622 (55.6%)	497 (44.4%)
Age (years)	73.6 (6.2)	74.9 (5.8)	71.8 (7.2)	73.4 (7.9)	74.1 (7.7)	75.5 (9.2)	72.7 (7.2)	74.1 (7.4)
Sex (F %)	58 (52.3%)	84 (51.2%)	145 (41.5%)	114 (40.7%)	69 (42.6%)	22 (41.5%)	272 (43.7%)	220 (44.3%)
Education (years)	16.0 (2.9)	16.4 (2.5)	15.9 (2.8)	16.2 (2.7)	15.1 (2.9)	16.1 (3.0)	15.7 (2.9)	16.3 (2.7)
Aβ42 (pg/mL)	190.8 (59.9)	205.7 (46.9)	154.1 (46.4)	194.2 (52.5)	128.1 (23.4)	137.5 (23.1)	154.1 (49.1)	191.9 (52.1)
T-tau (pg/mL)	68.8 (32.5)	68.4 (32.0)	106.3 (60.9)	72.9 (41.9)	131.6 (62.5)	133.0 (57.8)	106.0 (60.7)	77.9 (45.2)
P-tau (pg/mL)	32.0 (20.9)	28.9 (15.0)	44.4 (23.4)	32.5 (18.3)	53.6 (30.7)	53.8 (30.6)	44.5 (26.0)	33.6 (20.3)
ADAS-cog	6.0 (3.0)	6.2 (3.1)	10.8 (4.8)	9.1 (4.3)	19.6 (6.7)	21.3 (7.2)	12.2 (6.9)	9.5 (6.1)
Aβ- (n %)	55 (20.0%)	104 (37.8%)	67 (10.7%)	150 (23.8%)	0 (0.0%)	0 (0.0%)	122 (10.9%)	254 (22.7%)
Aβ+ (n %)	56 (20.4%)	60 (21.8%)	282 (44.8%)	130 (20.7%)	162 (75.3%)	53 (24.7%)	500 (44.7%)	243 (21.7%)

The measured data are represented by mean and standard deviation. Abbreviations: Aβ-, without significant Aβ deposition; Aβ+, with significant Aβ deposition; CN, cognitively normal; MCI, mild cognitive impairment; AD, Alzheimer's disease; ADAS-cog, Alzheimer's disease assessment scale-cog.

0.001). *APOE* ε4 existed in 17.5%, 42.4%, and 73.0% of individuals with significant Aβ deposition and only 8.4%, 7.0%, and 0.0% of individuals without significant Aβ deposition in CN ($p = 0.001$), MCI ($p < 0.001$), and AD ($p < 0.001$), respectively (Table 1).

APOE ε3 carriership was more common in CN than MCI and AD ($p < 0.001$ for both), and in MCI than in AD ($p < 0.001$). *APOE* ε3 was present in 21.7% of persons with significant Aβ deposition and 22.7% of persons without significant Aβ deposition in all participants ($p = 1.782$). *APOE* ε3 existed in 21.8%, 20.7%, and 24.7% of individuals with significant Aβ deposition and 37.8%, 23.8%, and 0.0% of individuals without significant Aβ deposition in CN ($p < 0.001$), MCI ($p = 0.525$), and AD ($p < 0.001$), respectively (Table 2).

Similar to *APOE* ε3, *APOE* ε2 carriership was also more common in CN than MCI and AD ($p < 0.001$ for both), but in MCI not than in AD ($p = 0.057$). *APOE* ε2 was present in 2.7% of individuals with significant Aβ deposition and 5.0% of individuals without significant Aβ deposition in all participants ($p = 0.012$). *APOE* ε2 carriership was present in 2.9%, 2.7%, and 2.3% of individuals with significant Aβ deposition and 11.6%, 3.8%, and 0.0% of individuals without significant Aβ deposition in CN ($p < 0.001$), MCI ($p = 0.789$), and AD ($p = 0.075$), respectively (Table 3).

3.2 CSF biomarkers differ by APOE allele status

CSF Aβ42 concentrations were significantly lower in *APOE* ε4 carriers compared with those who were *APOE* ε4 noncarriers in any group ($p = 0.009$ for AD, $p < 0.001$ for others) (Table 1). CSF P-tau was higher in *APOE* ε4 carriers than *APOE* ε4 noncarriers in MCI and all participants ($p < 0.001$ for both), but there were no differences in CN ($p = 1.161$) and AD ($p = 0.474$) groups. The results of CSF T-tau were similar to that of P-tau (Table 1).

Contrary to *APOE* ε4, CSF Aβ42 concentrations were higher in *APOE* ε3 carriers compared with those who were *APOE* ε3 noncarriers in MCI ($p < 0.001$), AD ($p = 0.027$), and all participants ($p < 0.001$), but not CN ($p = 0.123$), as shown in Table 2. CSF P-tau was lower in *APOE* ε3 carriers than *APOE* ε3 noncarriers in MCI and all participants ($p < 0.001$ for both), but there were no differences in CN ($p = 1.392$) and AD ($p = 2.586$) groups. The results of CSF T-tau were also similar to that of P-tau (Table 2).

CSF Aβ42 concentrations were significantly higher in *APOE* ε2 carriers compared with those who were *APOE* ε2

Table 3. Demographic and clinical characteristics of *APOE* ε2 carriers and noncarriers.

Characteristics	CN		MCI		AD		All	
	ε2-	ε2+	ε2-	ε2+	ε2-	ε2+	ε2-	ε2+
N (n %)	235 (85.5%)	40 (14.5%)	588 (93.5%)	41 (6.5%)	210 (97.7%)	5 (2.3%)	1033 (92.3%)	86 (7.7%)
Age (years)	74.5 (6.1)	73.5 (5.4)	72.4 (7.5)	72.9 (7.8)	74.5 (8.2)	77.8 (7.8)	73.3 (7.4)	73.6 (7.0)
Sex (F %)	114 (48.9%)	28 (66.7%)	342 (58.9%)	28 (58.3%)	126 (60.1%)	4 (50.5%)	582 (57.0%)	60 (61.2%)
Education (years)	16.3 (2.6)	15.8 (3.1)	16.1 (2.8)	16.0 (2.9)	15.5 (3.0)	15.4 (1.9)	16.0 (2.8)	15.9 (2.9)
Aβ42 (pg/mL)	194.0 (51.7)	229.5 (49.5)	170.1 (52.8)	185.7 (54.0)	130.0 (23.4)	136.6 (24.7)	167.4 (52.5)	200.5 (57.4)
T-tau (pg/mL)	70.1 (32.8)	60.3 (26.4)	92.6 (55.8)	74.7 (51.6)	129.5 (59.1)	182.5 (93.8)	94.8 (55.7)	77.3 (57.1)
P-tau (pg/mL)	31.0 (16.4)	27.4(24.1)	40.0 (22.9)	32.0 (16.1)	53.6 (30.6)	50.2 (29.1)	40.7 (24.6)	31.5 (20.8)
ADAS-cog	6.3 (3.1)	5.3 (2.4)	10.0 (4.6)	9.6 (4.8)	19.9 (6.9)	20.0 (6.0)	11.2 (6.7)	8.6 (5.7)
Aβ- (n %)	127 (46.2%)	32 (11.6%)	193 (30.7%)	24 (3.8%)	0 (0.0%)	0 (0.0%)	320 (28.6%)	56(5.0%)
Aβ+ (n %)	108 (39.3%)	8 (2.9%)	395 (62.8%)	17 (2.7%)	210 (97.7%)	5 (2.3%)	713 (63.7%)	30 (2.7%)

The measured data are represented by mean and standard deviation. Abbreviations: Aβ-, without significant Aβ deposition; Aβ+, with significant Aβ deposition; CN, cognitively normal; MCI, mild cognitive impairment; AD, Alzheimer's disease; ADAS-cog, Alzheimer's disease assessment scale-cog.

noncarriers in CN and all participants ($p < 0.001$ for both), but not in MCI ($p = 0.162$) and AD ($p = 1.596$), as shown in Table 3. CSF P-tau was lower in *APOE* ε2 carriers than *APOE* ε2 noncarriers in CN ($p = 0.036$), MCI ($p = 0.042$), and all participants ($p < 0.001$), but there were no differences in AD ($p = 2.055$) group. CSF T-tau was lower in *APOE* ε2 carriers than *APOE* ε2 noncarriers in MCI ($p = 0.015$) and all participants ($p < 0.001$), but there were no differences in CN ($p = 0.270$) and AD ($p = 0.282$) groups (Table 3).

3.3 ADAS-cog scores differ by APOE allele status

ADAS-cog scores were higher in *APOE* ε4 carriers compared with *APOE* ε4 noncarriers in MCI and all participants ($p < 0.001$ for both), but there were no significant differences in CN ($p = 1.161$) and AD ($p = 0.474$) groups (Table 1).

Contrary to *APOE* ε4, ADAS-cog scores were lower in *APOE* ε3 carriers than *APOE* ε3 noncarriers in MCI and all participants ($p < 0.001$ for both), but there were also no significant differences between *APOE* ε3 carriers and *APOE* ε3 noncarriers in CN ($p = 2.208$) and AD ($p = 0.318$) groups (Table 2).

Though ADAS-cog scores were lower in *APOE* ε2 carriers than *APOE* ε2 noncarriers in all participants ($p < 0.001$), there were no significant differences between *APOE* ε2 carriers and *APOE* ε2 noncarriers in CN ($p = 0.252$), MCI ($p = 1.455$), and AD ($p = 2.556$) groups (Table 3).

3.4 The associations of APOE with T-tau, P-tau, and ADAS-cog in all participants with and without significant Aβ deposition

The associations of *APOE* with T-tau, P-tau, and ADAS-cog were first tested in linear regression models with an interaction term between *APOE* ε4, ε3, and 2ε status and the presence of Aβ, adjusting for age, sex, and education. The interaction was significant between *APOE* ε4 and ε3 allele status and the presence of Aβ for T-tau, P-tau, and ADAS-cog (Tables 4,5). However, the ε2 by Aβ interaction was not significant, as shown in Table 6.

Next, we carried out separate regression analyses for persons with (n = 743) and without (n = 376) significant Aβ deposition. In individuals with significant Aβ deposition, the *APOE* ε4 allele is associated with increased T-tau, P-tau, and ADAS-cog (Table 7). We did not observe an association among individuals without significant Aβ deposition (Table 7). *APOE* ε3 was related to decreased T-tau, P-tau, and ADAS-cog levels in individuals with significant Aβ deposition but not individuals without significant Aβ deposition (Table 7). However, in this model, *APOE* ε2 was not associated with levels of T-tau, P-tau, and ADAS-cog levels in individuals with or without significant Aβ deposition, as shown in Table 7.

3.5 APOE status on levels of T-tau, P-tau, and ADAS-cog in CN, MCI, and AD groups with and without significant Aβ deposition

Finally, we performed stratified analyses regressing *APOE* ε4 status on levels of T-tau, P-tau, and ADAS-cog in CN, MCI, and AD groups with and without significant Aβ deposition. We found that *APOE* ε4 strongly associated with increased levels of T-tau, P-tau, and ADAS-cog in MCI group with significant Aβ deposition ($\beta = 0.27$, $p < 0.001$; $\beta = 0.20$, $p < 0.001$; $\beta = 0.17$, $p < 0.001$, respectively) (Fig. 1A–C), and increased levels of P-tau in CN group with significant Aβ deposition ($\beta = 0.22$, $p = 0.049$) (Fig. 1B). However, we did not observe the same associations among persons without significant Aβ deposition, as shown in Fig. 1A–C.

Contrary to *APOE* ε4, *APOE* ε3 was strongly related to decreased levels of T-tau, P-tau, and ADAS-cog in MCI group with significant Aβ deposition ($\beta = -0.25$, $p < 0.001$; $\beta = -0.18$, $p < 0.001$; $\beta = -0.18$, $p < 0.001$, respectively) (Fig. 2A–C). As shown in Fig. 2A–C, we did not observe the same relationships among persons without significant Aβ deposition.

We repeated the analysis for the *APOE* ε2 allele. Again, we found a significant association of the *APOE* ε2 allele with decreased T-tau levels only in the MCI group with significant Aβ deposition ($\beta = -0.27$, $p = 0.036$) (Fig. 3A).

Table 4. Linear regression results of *APOE* ε4 status and the presence of Aβ.

Parameters	Models	Aβ β (SE), pc	*APOE* ε4 β (SE), *p*	Aβ + *APOE* ε4 (Interaction) β (SE), *p*
T-tau	Model 1	0.54 (0.03), <0.001	-	-
	Model 2	-	0.4 (0.03), <0.001	-
	Model 3	0.4 (0.04), <0.001	0.1 (0.06), 0.360	0.24 (0.07), 0.018
P-tau	Model 1	0.58 (0.03), <0.001	-	-
	Model 2	-	0.38 (0.03), <0.001	-
	Model 3	0.47 (0.04), <0.001	0.07 (0.06), 0.870	0.11 (0.07), 0.036
ADAS-cog	Model 1	0.48 (0.04), <0.001	-	-
	Model 2	-	0.38 (0.04), <0.001	-
	Model 3	0.31 (0.05), <0.001	0.02 (0.08), 2.310	0.25 (0.09), 0.015

Table 4 indicated β coefficient, Standard error (SE), and p value from the models. Model 1 = age + sex + education + Aβ; Model 2 = age + sex + education + *APOE* ε4; Model 3 = age + sex + education + Aβ + *APOE* ε4 + interaction of *APOE* ε4 and Aβ. Abbreviations: ADAS-cog, Alzheimer's disease assessment scale-cog; *APOE*, apolipoprotein E.

Table 5. Linear regression results of *APOE* ε3 status and the presence of Aβ.

Parameters	Models	Aβ β (SE), *p*	*APOE* ε3β (SE), *p*	Aβ + *APOE* ε3 (Interaction) β (SE), *p*
T-tau	Model 1	0.54 (0.03), <0.001	-	-
	Model 2	-	–0.31 (0.03), <0.001	-
	Model 3	0.61 (0.05), <0.001	–0.02 (0.05), 2.160	–0.24 (0.06), 0.009
P-tau	Model 1	0.58 (0.03), <0.001	-	-
	Model 2	-	–0.29 (0.03), <0.001	-
	Model 3	0.64 (0.05), <0.001	0.00 (0.05), 2.910	–0.16 (0.07), 0.036
ADAS-cog	Model 1	0.48 (0.04), <0.001	-	-
	Model 2	-	–0.30 (0.04), <0.001	-
	Model 3	0.65 (0.06), <0.001	0.07 (0.06), 0.870	–0.32 (0.08), <0.001

Table 5 indicated β coefficient, Standard error (SE), and p value from the models. Model 1 = age + sex + education + Aβ; Model 2 = age + sex + education + *APOE* ε3; Model 3 = age + sex + education + Aβ + *APOE* ε3 + interaction of *APOE* ε3 and Aβ. Abbreviations: ADAS-cog, Alzheimer's disease assessment scale-cog; *APOE*, apolipoprotein E.

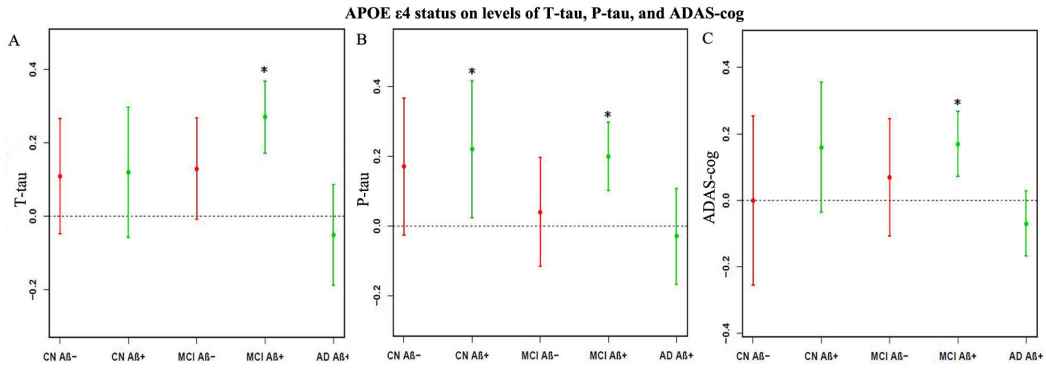

Fig. 1. *APOE* ε4 status on levels of T-tau, P-tau, and ADAS-cog in CN, MCI, and AD with or without significant Aβ deposition. (A–C) The data are estimates (β-coefficients) from stratified analyses, and the confidence interval of regression is 95%. All values are Log transformed. Effects were significant (*), for T-tau (A) In MCI with significant Aβdeposition ($\beta = 0.27$, $p < 0.001$); for P-tau. (B) In CN and MCI with significant Aβdeposition ($\beta = 0.22$, $p = 0.049$; $\beta = 0.20$, $p < 0.001$, respectively); for ADAS-cog. (C) In MCI significant with Aβdeposition ($\beta = 0.17$, $p < 0.001$).

4. Discussion

This work evaluated the effects of different *APOE* allele statuses on T-tau, P-tau, and cognition in relation to Aβ deposition in a large cohort of subjects. We have the following main findings: Firstly, there were significant differences between *APOE* allele carriers and noncarriers in the measures of T-tau, P-tau, and ADAS-cog scores in MCI, but not in CN and AD. Secondly, there was an interaction between *APOE* ε4 and ε3 and the presence of Aβ. Finally, *APOE* ε4 and *APOE* ε3 were associated with CSF tau and cognition in MCI participants with Aβ deposition, but not in AD participants with Aβ deposition.

Table 6. Linear regression results of *APOE* ε2 status and the presence of Aβ.

Parameters	Models	Aβ β (SE), *p*	*APOE* ε2 β (SE), *p*	Aβ + *APOE* ε2 (Interaction) β (SE), *p*
T-tau	Model 1	0.54 (0.03), <0.001	-	-
	Model 2	-	–0.23 (0.06), <0.001	-
	Model 3	0.54 (0.03), <0.001	–0.04 (0.07), 1.770	–0.11 (0.1), 0.870
P-tau	Model 1	0.58 (0.03), <0.001	-	-
	Model 2	-	–0.24 (0.06), <0.001	-
	Model 3	0.59 (0.03), <0.001	–0.04 (0.07), 1.560	–0.09 (0.1), 1.140
ADAS-cog	Model 1	0.48 (0.04), <0.001	-	-
	Model 2	-	–0.27 (0.07), <0.001	-
	Model 3	0.47 (0.04), <0.001	–0.13 (0.08), 0.330	–0.03 (0.12), 2.490

Table 6 indicated β coefficient, Standard error (SE), and p value from the models Model 1 = age + sex + education + Aβ; Model 2 = age + sex + education + *APOE* ε2; Model 3 = age + sex + education + Aβ + *APOE* ε2 + interaction of *APOE* ε2 and Aβ. Abbreviations: ADAS-cog, Alzheimer's disease assessment scale-cog; *APOE*, apolipoprotein E.

Table 7. Correlation of *APOE* ε4, *APOE* ε3, and *APOE* ε2 status with T-tau, P-tau, and ADAS-cog.

Aβ status	Model	*APOE* ε4 β (SE), *p*	*APOE* ε3 β (SE), *p*	*APOE* ε2 β (SE), *p*
Aβ+	T-tau	0.23 (0.04), <0.001	–0.21(0.04), <0.001	–0.14 (0.08), 0.279
	P-tau	0.19 (0.04), <0.001	–0.16(0.04), <0.001	–0.13 (0.08), 0.300
	ADAS-cog	0.27 (0.05), <0.001	–0.25(0.05), <0.001	–0.15 (0.1), 0.330
Aβ-	T-tau	0.12 (0.05), 0.195	–0.04 (0.04), 1.180	–0.04 (0.06), 1.560
	P-tau	0.08 (0.06), 0.510	–0.01 (0.05), 2.430	–0.04 (0.06), 1.560
	ADAS-cog	0.05 (0.08), 1.440	0.06 (0.05), 1.110	–0.13 (0.08), 0.261

Table 7 presented β coefficient, Standard error (SE), and p value from the models considering all subjects as a whole. All models were adjusted for age, sex, and education. Abbreviations: ADAS-cog, Alzheimer's disease assessment scale-cog; *APOE*, apolipoprotein E; Aβ-, without significant Aβ deposition; Aβ+, with significant Aβ deposition.

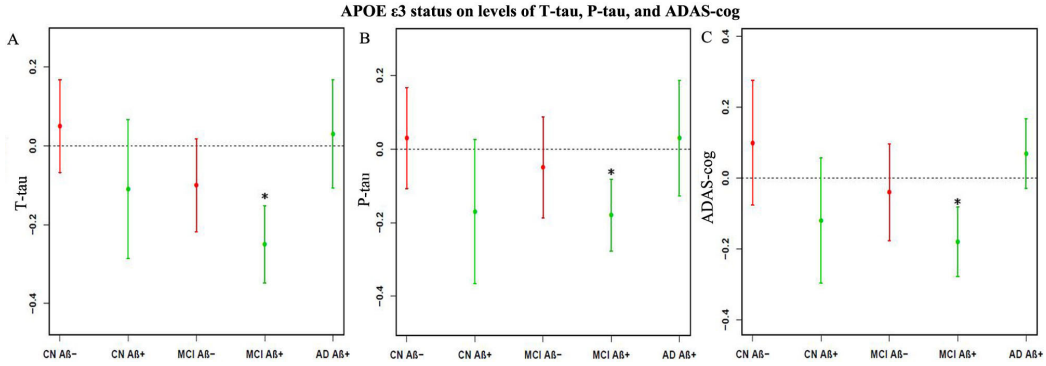

Fig. 2. *APOE* ε3 status on levels of T-tau, P-tau, and ADAS-cog in CN, MCI, and AD with or without significant Aβ deposition. (A–C) The data are estimates (β-coefficients) from stratified analyses, and the confidence interval of regression is 95%. All values are Log transformed. Effects were significant (*), for T-tau (A) In MCI with significant Aβ deposition ($\beta = -0.25$, $p < 0.001$); for P-tau. (B) In MCI with significant Aβ deposition ($\beta = -0.18$, $p < 0.001$); for ADAS-cog. (C) In MCI with significant Aβ deposition ($\beta = -0.18$, $p < 0.001$).

Compared with noncarriers, previous studies reported *APOE* ε4 carriers had higher deposition of Aβ in the cerebral cortex in late-onset AD [42, 43]. A low CSF Aβ level is considered a marker of Aβ deposition in AD patients's brains [44]. Consistent with the report by Vemuri *et al.* [45], within CN, MCI, and AD group, *APOE* ε4 carriers had lower CSF Aβ42 than noncarriers. In addition, in CN and MCI groups, results demonstrated that *APOE* ε4 was more common in individuals with significant Aβ deposition than in subjects without significant Aβ deposition. There were no individuals without significant Aβ deposition in the AD group, suggesting that *APOE* ε4 may relate strongly to CSF Aβ in the different phases of cognitive damage. On the contrary, *APOE* ε3 carriers had higher CSF Aβ42 than noncarriers in any group. However, *APOE* ε2 carriers had higher CSF Aβ42 than noncarriers only in the CN group. *APOE* ε3 and ε2 were widespread in individuals with significant Aβ deposition in the AD group, and they were prevalent in par-

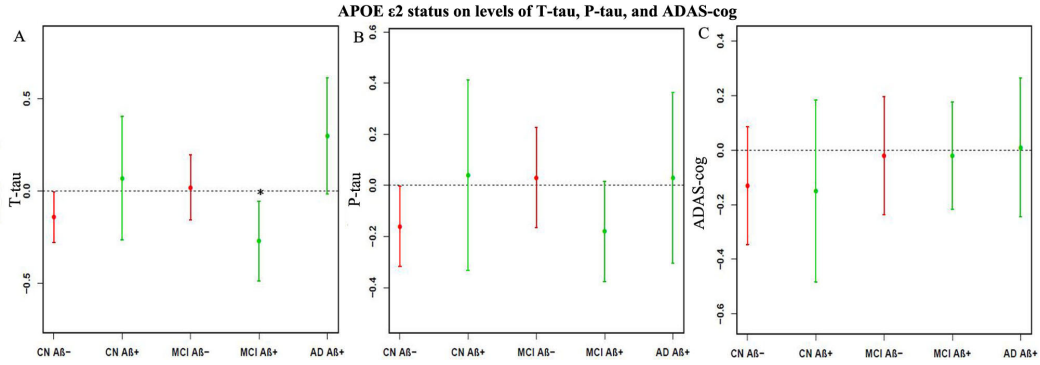

Fig. 3. *APOE* ε2 status on levels of T-tau, P-tau, and ADAS-cog in CN, MCI, and AD with or without significant Aβ deposition. (A–C) The data are estimates (β-coefficients) from stratified analyses, and the confidence interval of regression is 95%. All values are Log transformed. Effects were significant (*), for T-tau (A) In MCI with significant Aβ deposition (β = -0.27, p = 0.036).

ticipants without significant Aβ deposition in the CN group. This phenomenon of *APOE* ε3 and ε2 in the AD group may be related to Aβ deposition in all AD patients. Relative to *APOE* ε4, we speculate that *APOE* ε3 and ε2 may have opposite effects in CN subjects.

There was no significant difference in T-tau and ADAS-cog scores between *APOE* allele carriers and noncarriers among CN. Among MCI, T-tau, P-tau, and ADAS-cog scores were significantly different between *APOE* allele carriers and noncarriers. Interestingly, there was not a single difference between *APOE* allele carriers and noncarriers in the measures of T-tau, P-tau, and ADAS-cog scores in AD subjects. Our data show significant differences in CSF Aβ42 levels between *APOE* allele carriers and noncarriers in all clinical groups. Still, there are no significant differences in T-tau values between *APOE* allele carriers and noncarriers in CN and AD individuals. In patients with clinically diagnosed cognitive impairment, the effect of *APOE* genotype on cognitive decline is the most consistent in MCI patients but not in AD patients. This is not to say that *APOE* genotypes are not associated with neuropathological parameters. When all individuals are combined, *APOE* ε4 significantly increases the risk of more severe clinical damage and has higher levels of P-tau and T-tau. However, *APOE* ε3 and ε2 have opposite effects. *APOE* genotype is not deterministic because of many ε4 carriers without dementia and many ε4 noncarriers with dementia [45]. In contrast, there are many ε3 and ε2 carriers with dementia and many ε3 and ε2 noncarriers without dementia.

In 2012, there was a change in the diagnostic criteria for AD neuropathology [46], requiring the presence of Aβ deposition for the neuropathological diagnosis of AD. However, the previous view shows that even in the absence of Aβ, the appearance of neurofibrillary tangles (NFT) is the earliest neuropathological manifestation of AD [47].Therefore, it has been argued that tau tangles are a pathophysiological process different from AD in the absence of Aβ [20, 48]. Several studies revealed a relationship between *APOE* and Aβ pathology and tau pathology, indicating that the association between *APOE* and tau pathology may be mediated by Aβ [20, 49]. We found an interaction between *APOE* ε4 and the presence of Aβ such that the associations of *APOE* ε4 with T-tau and P-tau were much more robust in persons with Aβ. When we considered all subjects as a whole, there was a significant association between *APOE* ε4 and increased CSF T-tau and P-tau concentrations in individuals with significant Aβ deposition. There is no similar phenomenon in individuals without significant Aβ deposition. In the stratified analyses regressing within CN, MCI, and AD groups, we found that *APOE* ε4 was significantly related to increased CSF T-tau and P-tau concentrations in MCI but not in AD to Aβ status. Few studies have tested the relationship between *APOE* ε3 and tau pathology. However, there was also an interaction between *APOE* ε3 and the presence of Aβ such that the associations of *APOE* ε3 with T-tau and P-tau were much more robust in persons with Aβ, and it revealed that *APOE* ε3 was associated with decreased concentrations of CSF T-tau and P-tau in individuals with Aβ deposition. In the stratified analyses regression within CN, MCI, and AD groups, the *APOE* ε3 allele was significantly associated with decreased CSF T-tau and CSF P-tau levels in the MCI with significant Aβ deposition. These results were not observed in individuals without significant Aβ deposition. Some studies reported that *APOE* ε2 carriers had reduced NFT [50, 51], though inconsistent findings exist [52, 53]. We did not find an interaction between *APOE* ε2 and the presence of Aβ related to tau. *APOE* ε2 was only associated with decreased levels of CSF T-tau in MCI individuals with significant Aβ deposition. Our results show that *APOE* ε4 and ε3 may only affect tau pathology in MCI patients, and Aβ mediates this effect. This work indirectly supports the concept that *APOE* alleles influence tau pathology dependently on Aβ, and tau pathology without Aβ may reflect a different pathological process from MCI.

A longitudinal study has reported that the relationship between *APOE* and global cognitive decline was mediated by Aβ and tau [54]. It was also found that the effects of *APOE* on a decline in episodic memory and non-episodic cognition were mediated by Aβ [30]. However, these findings did not divide the subjects according to the severity of cognitive im-

pairment. We found an interaction between *APOE* ε4 and ε3 and the presence of Aβ such that the associations of *APOE* ε4 and *APOE* ε3 with ADAS-cog were much more robust in persons with Aβ. When we considered all participants as a whole, there was a significant correlation between *APOE* ε4 and increased ADAS-cog scores and between *APOE* ε3 and decreased ADAS-cog scores in persons with significant Aβ deposition but not in persons without significant Aβ deposition. However, *APOE* ε2 was not associated with ADAS-cog in individuals with and without significant Aβ deposition. In the stratified analyses regressing within CN, MCI, and AD groups, we revealed that *APOE* ε4 was only significantly associated with increased ADAS-cog scores in the MCI individuals with significant Aβ deposition, and *APOE* ε3 was only significantly associated with decreased ADAS-cog scores in the MCI individuals with significant Aβ deposition. *APOE* ε2 was not associated with ADAS-cog in the MCI and AD individuals with or without significant Aβ deposition. Our work suggests that the effect of *APOE* ε4 and *APOE* ε3 on cognitive decline is only observed in MCI, and Aβ also mediates this effect. In addition, it demonstrates that *APOE* ε3 has a protective effect on MCI but not AD, and *APOE* ε2 has no protective effect on MCI and AD. These seem to differ from previous conclusions that *APOE* ε3 is considered neutral and *APOE* ε2 is protective of AD risk. We do not know what the reason is, but we believe it is an interesting question for further research.

Our data suggest that the *APOE* genotype may only influence CSF tau and cognition in MCI participants. Just as we know, *APOE* ε4 likely predates the onset of Aβ deposition [45], then Aβ deposition initiates the cascade. Once Aβ triggers the downstream process is, other factors will lead to the AD's complete pathologic/clinical manifestations [55]. Therefore, we speculate that tau pathology and cognition in AD may be more affected by other factors, such as inflammatory factors, loss of cells, synapses, and dendrites and so on. The other possibility is that the groups are defined by being in a specific cognitive range, and the effect may not be noticed. However, future work is needed to determine why *APOE* genotype is only related to tau pathology and cognition in MCI patients. In addition, *APOE* ε3 was associated with lower amyloid (higher CSF Aβ42). Thus, it perhaps slows the trajectory of conversion from MCI to AD. However, its downstream signaling mechanism is still unknown, which may be an exciting topic in future research.

There are a few limitations. First of all, it lacks longitudinal data, so it cannot observe the dynamic impact of *APOE* on CSF tau and cognition. Secondly, it did not contain non-AD neurodegenerative disorders. Finally, the ADNI database consists of self-selected, highly educated volunteers interested in participating in AD research, which may concern their cognition. As such, our findings will benefit from replication in another population-based cohort.

5. Conclusion

We found that *APOE* ε4 and ε3 were associated with CSF tau and ADAS-cog. However, *APOE* ε4 and ε3 only affect tau pathology and cognitive function in MCI patients, and Aβ mediates these effects. Thus, in addition to positron emission tomography (PET) data for Aβ and tau, our findings highlight the need for future longitudinal studies examining the effects of *APOE* on tau and ADAS-cog.

Abbreviations

Aβ, amyloid-β; AD, Alzheimer's disease; ADAS-cog, Alzheimer's disease assessment scale-cog; ADNI, Alzheimer's disease Neuroimaging Initiative; *APOE*, Apolipoprotein E; CDR, Clinical Dementia Rating scale; CN, cognitively normal; CSF, cerebrospinal fluid; MCI, mild cognitive impairment; MMSE, Mini-mental State Examination; NFT, neurofibrillary tangles; PET, positron emission tomography; PHF, paired helical filament.

Author contributions

FX: manuscript drafting and composition of figures. TM: analysis of data. JT: collection of data. JL: interpretation of data. HZ: concept and supervision of the research.

Ethics approval and consent to participate

The Institutional Review Boards approved the ADNI study of all the participating institutions. In addition, informed written consent was obtained from all participants at every center.

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Conflict of interest

The authors declare an interest in the Alzheimer's Disease Neuroimaging Initiative.

Data availability statement

The datasets used and/or analyzed in this study may be obtained from the corresponding author on reasonable request.

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