



Lack of neural compensatory mechanisms of BDNF val66met met carriers and APOE E4 carriers in healthy aging, mild cognitive impairment, and Alzheimer's disease

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ARTICLE INFO

Article history:

Received 17 June 2015

Received in revised form 2 November 2015

Accepted 14 December 2015

Available online 19 December 2015

Keywords:

BDNF

APOE

Aging

MCI

AD

Brain morphometry

Cognition

ABSTRACT

Compromises in compensatory neurobiologic mechanisms due to aging and/or genetic factors (i.e., APOE gene) may influence brain-derived neurotrophic factor (BDNF) val66met polymorphism effects on temporal lobe morphometry and memory performance. We studied 2 cohorts from Alzheimer's Disease Neuroimaging Initiative: 175 healthy subjects and 222 with prodromal and established Alzheimer's disease. Yearly structural magnetic resonance imaging and cognitive performance assessments were carried out over 3 years of follow-up. Both cohorts had similar BDNF Val/Val and Met allele carriers' (including both Val/Met and Met/Met individuals) distribution. In healthy subjects, a significant trend for thinner posterior cingulate and precuneus cortices was detected in Met carriers compared to Val homozygotes in APOE E4 carriers, with large and medium effect sizes, respectively. The mild cognitive impairment/Alzheimer's disease cohort showed a longitudinal decline in entorhinal thickness in BDNF Met carriers compared to Val/Val in APOE E4 carriers, with effect sizes ranging from medium to large. In addition, an effect of BDNF genotype was found in APOE E4 carriers for episodic memory (logical memory and ADAS-Cog) and semantic fluency measures, with Met carriers performing worse in all cases. These findings suggest a lack of compensatory mechanisms in BDNF Met carriers and APOE E4 carriers in healthy and pathological aging.

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1. Introduction

Brain-derived neurotrophic factor (BDNF) is a neurotrophin that facilitates episodic memory function and storage through the promotion of long term potentiation and synaptic plasticity (Egan et al., 2003), as well as neuronal survival and differentiation (Zuccato and Cattaneo, 2009). Specifically, BDNF expression is particularly high in the hippocampus (Binder and Scharfman, 2004) and is required for some forms of hippocampus-mediated plasticity (Tanaka et al., 2008). A common missense polymorphism in the human BDNF

gene produces an amino acid substitution (valine to methionine) at codon 66 (val66met). This single nucleotide polymorphism impacts intracellular trafficking of BDNF, such that val pro-BDNF is more likely to be localized in neurites, whereas met BDNF aggregates in the cell body (Egan et al., 2003). This polymorphism also impacts medial temporal lobe structural and functional integrity, as well as cognition (Goldberg et al., 2008; Hariri et al., 2003; Sambataro et al., 2010).

Nonetheless, some reports have highlighted the fact that effects of the BDNF Val66Met variant on brain structure and function are complex and may have a different impact when considering processes related to normal aging and pathological conditions. Sambataro et al. illustrated this modulatory effect of BDNF on the trajectory of age-related changes in hippocampal function; Older met carriers showed impaired activation of the hippocampus during memory encoding and memory retrieval tasks compared to val/val individuals (Sambataro et al., 2010). BDNF Met carriers have also been found to have reductions of hippocampal volumes associated with age. However, Voineskos et al. found the opposite, in that

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¹ Data used in preparation of this article were obtained from the Alzheimer's Disease Neuroimaging Initiative (ADNI) database (adni.loni.ucla.edu). As such, the investigators within the ADNI contributed to the design and implementation of ADNI and/or provided data but did not participate in analysis or writing of this report. A complete listing of ADNI investigators can be found at: http://adni.loni.ucla.edu/wp-content/uploads/how_to_apply/ADNI_Acknowledgement_List.pdf.

Val/Val individuals were more susceptible to age-related decline in late life, showing decreased thickness in the temporal lobe structures and episodic memory performance, whereas met carriers were more susceptible in early adult life (Voineskos et al., 2011). Not surprisingly then, the effect of BDNF Met and hippocampal volume has been sometimes attributed to a “winners curse” effect (Molendijk et al., 2012). More recently, Lim et al. have shown prospectively that BDNF val66met met carriage affects brain volumes in older adults only in the presence of abnormally high levels of amyloid (Lim et al., 2015). Thus, brain amyloidosis may be mediating BDNF effects on brain structure.

In the context of preclinical Alzheimer's disease (AD), it has been recently reported that healthy subjects carriers of the BDNF Met allele coupled with high PIB-PET A β uptake showed accelerated cognitive decline and atrophy of the hippocampus (Lim et al., 2014). Furthermore, the same group found an epistatic interaction of BDNF and APOE genotypes on memory decline in the context of brain amyloidosis (Lim et al., 2015). They also showed similar findings in individuals with mild cognitive impairment (MCI) who were amyloid positive (Lim et al., 2014). It is important to note that these studies used prospective designs.

However, in contrast to most studies showing deleterious effects of met carriage, it must be acknowledged that a different hypothesis regarding the neurobiological effects of BDNF genotype has arisen from the complexities associated with BDNF molecular processing. Mature BDNF has generally been associated with long-term potentiation through its interaction with the TrkB receptor. In contrast, its precursor, pro-BDNF (a form of BDNF that includes a prodomain containing val66met and the region of the mature protein) may be associated with apoptosis through interactions with p75 (Lu, 2003). Additionally, the isolated prodomain may, when it contains the met allele, be associated with various negative synaptic parameters (Anastasia et al., 2013). Goldman et al. highlighted an advantageous role of Met allele in promoting recovery of executive function (Krueger et al., 2011) and preservation of general cognitive functioning (Barbey et al., 2014) after penetrating TBI. They attributed their finding to trafficking impairments associated with met allele which ultimately reduced apoptotic effects. Therefore, Met allele may be protective in certain diseases (Zivadinov et al., 2007).

We have proposed that BDNF val66met genotypic effects may be more clearly observed in older cohorts followed longitudinally both because age effects on genotypic differences (see above) and because declines may be characterized more accurately in within-subject designs (Goldberg and Mattay, 2009; Li et al., 2010; Papenberg et al., 2015; Sambataro et al., 2010). Such a view is also consistent with increasing heritability for cognitive domains with age (Deary et al., 2012). Here, we comprehensively examine this proposal by (1) testing the effects of BDNF genotype on age-related decline in cognition and brain morphometry measures of older healthy subjects (over 3 years of follow-up) and (2) by considering MCI/AD as a pathophysiological neurodegenerative state in which to examine BDNF effects. We critically stratified our results by APOE genetic variation, considering that E4 allele is associated with an increased OR for AD in comparison to E3 homozygotes, and APOE might act as a marker for neurodegeneration and risk for AD by influencing Abeta misprocessing. Our study extends prior work by Ward et al. (2014) that found APOE \times BDNF effect on episodic memory in cross-sectional data.

2. Methods

2.1. Subjects

To test our hypotheses, we examined 2 cohorts from Alzheimer's Disease Neuroimaging Initiative (ADNI): (1) Healthy subjects (HSS)

($n = 175$) and (2) AD patients and MCI individuals who progressed to AD at follow-up, thus showing evidence of prodromal AD at baseline ($n = 222$). Details of inclusion, exclusion, and sample selection criteria can be found elsewhere (Gomar et al., 2011, 2014; Sousa et al., 2015). Briefly, healthy participants were between 55–90 (inclusive) years old, had a Clinical Dementia Rating (CDR) (Morris, 1993) score of 0, a Mini-Mental State Examination (MMSE) (Folstein et al., 1975) score between 24 and 30 (inclusive), normal memory function according to Logical Memory II subscale (delayed Paragraph Recall) from the Wechsler Memory Scaled-Revised (Wechsler, 1987), no memory complaints, absence of significant impairment in other cognitive domains, and preserved activities of daily living. MCI patients had MMSE scores between 24 and 30 (inclusive), a memory complaint, objective memory loss as indicated by 1.5 standard deviations below the education adjusted cutoff on the Logical Memory II subscale, a CDR score of 0.5, absence of significant impairment in other cognitive domains, and preserved activities of daily living. All MCI patients converted to AD at follow-up (mean time until conversion 20.44 months) (Gomar et al., 2014). AD patients had MMSE scores between 20 and 26 (inclusive), memory complaint, objective memory loss, a CDR score of 0.5 or 1, and the National Institute of Neurological and Communicative Disorders and Stroke–Alzheimer's Disease and Related Disorders (NINCDS/ADRDA) criteria for probable AD (McKhann et al., 1984). All participants signed written informed consent for participation in ADNI, as approved by the institutional board at each participating center.

2.2. Genotyping

Genetic assessment for the functional single nucleotide polymorphism of the BDNF gene at nucleotide 196 (rs6265) was performed using the Illumina Human610-Quad BeadChip (Illumina, Inc, San Diego, CA, USA) and intensity data processed with GenomeStudio, version 2009.1. The 2 SNPs of the APOE gene (rs429358, rs7412) were genotyped from a 3-mL aliquot of blood taken in ethylenediaminetetraacetic acid (EDTA)-containing vacutainer tubes, and genomic DNA was extracted by Cogenics (now Beckman Coulter Genomics) using QIAamp DNA Blood Maxi Kit (Qiagen, Inc, Valencia, CA, USA) following manufacturer's protocol. For a more detailed description of the genotyping protocol, see Saykin et al. (2010).

2.3. Magnetic resonance imaging acquisition and extraction of brain morphometry measures

Scans were obtained from 1.5-Tesla scanners at different sites involved in ADNI with minor variations in the magnetic resonance imaging protocol based on the specific configuration of each scanner. Volumetric measures of the total brain, gray, white matter, and hippocampus, as well as cortical thickness measures of temporal lobe regions (middle temporal, inferior lateral temporal, parahippocampal, and entorhinal) were extracted. These measures were derived by Freesurfer (<http://surfer.nmr.mgh.harvard.edu/>) (Dale et al., 1999; Fischl et al., 2002). We also examined several other cortical thickness measures along the parietal and frontal lobes: posterior cingulate, precuneus, isthmus cingulate, anterior cingulate, middle frontal, lateral, and medial orbitofrontal, to extend our analyses to key brain regions associated to neurodegeneration. Detailed descriptions of magnetic resonance imaging protocol and methods are available at ADNI webpage and on request to the authors. Individuals with either partial or total failure in the Freesurfer reconstruction stream outcome were excluded from further analysis.

2.4. Cognitive assessments

Several key cognitive measures were selected. First, we focused our analyses on both immediate and delayed episodic memory measures: immediate and delayed logical memory of the Wechsler Memory Scale (Wechsler, 1987), and a composite score for ADAS-Cog memory items: word recall test, delayed word recall, and word recognition. Second, we also selected measures of working memory, language and semantic fluency, speed of processing, visuo-spatial abilities, and executive function (Goodglass and Kaplan, 1983; Wechsler, 1981, 1987). We also selected MMSE score as a measure of general cognition (Folstein et al., 1975).

2.5. Statistical analysis

Comparisons on demographic variables between BDNF Val/Val and Met carrier's (including both Val/Met and Met/Met individuals) subgroups within each group (HS and MCI/AD) at baseline were performed with chi-square and t-tests for dichotomous and quantitative variables, respectively.

The analytic approach for longitudinal data was as follows. Linear mixed models (SAS 9.3, PROC MIXED) were performed to examine the effect of BDNF genotype on temporal lobe integrity. Models included 3 factors: BDNF genotype group (Val/Val homozygotes and Met Carriers), Time (years since baseline), and a term for the interaction between BDNF \times Time. Covariates for gender, education, and year at each time point were included in all models. In these mixed models, the covariance pattern was set as heterogeneous autoregressive structure. Time was included as repeated factor and BDNF group (BDNF Val/Val vs. BDNF Met carriers) as a between-subject factor; subject was the random factor. All the analyses were performed first in the whole sample and second stratifying the sample by APOE genotype into 2 subgroups, APOE E3/E3 homozygotes, and APOE E4 carriers; APOE E2 carriers were excluded because of the association of this allele with neuroprotection (Conejero-Goldberg et al., 2014). We also note that we used APOE4 to stratify the sample because it is a risk factor for neurodegeneration through multiple molecular mechanisms. Analyses were performed in HS and MCI/AD participants independently according to the hypothesis to be tested. Results were corrected for multiple comparisons through false discovery rate (FDR) method ($p = 0.10$). The method implemented by SAS PROC MULTTEST use FDR adjustment of multiple comparisons following Benjamini and Hochberg procedure (Benjamini et al., 2001; Benjamini and Hochberg, 1995). Statistical significance was set at $p < 0.05$ level. We believe that this approach in which we conducted separate analyses based on APOE E4 positivity offered transparency and clarity in interpretation, as well as statistical rigor, due to FDR correction. Effect sizes were computed using Hedges and Olkin correction approach (Hedges and Olkin, 1985). We repeated the analyses including an APOE \times BDNF interaction effect obtaining confirmatory results.

3. Results

3.1. Participant's characteristics and BDNF genotype distribution

Within both HS and MCI/AD samples, distribution of age, gender, and education were similar between BDNF Val/Val homozygotes and Met carriers (Table 1). Distribution of BDNF Val66Met polymorphism was similar between HS and MCI/AD patients, 31% of HS and 31% of MCI/AD were Met carriers ($\chi^2 = 0.01$, $p = 0.94$). These distributions did not change when dividing MCI and AD patients, 33% of MCI and 27% of AD were Met carriers. These characteristics remained unchanged when stratifying by APOE genotype and excluding APOE E2 carriers. Among HS, 106 subjects were APOE E3 homozygotes and 45 subjects were APOE E4 carriers (1 subject APOE E2 homozygote, 21 subjects APOE E2/E3, and 2 subjects APOE E2/E4 were excluded). Among MCI/AD, 70 subjects were APOE E3 homozygotes and 138 subjects were APOE E4 carriers (6 subjects APOE E2/E3 and 8 subjects APOE E2/E4 were excluded). These results were similar when stratifying for APOE genotype.

3.2. Impact of BDNF genotype on brain integrity and cognitive markers in healthy subjects

In the HS sample, we found a marginally significant main effect of BDNF genotype in posterior cingulate thickness ($F_{1,41} = 7.99$, $p = 0.07$), in the APOE E4 carrier's subsample. BDNF Met carriers and APOE E4 carriers showed decreased thickness of the posterior cingulate area compared to APOE E3/E3 and BDNF Val/Val homozygotes (Fig. 1A). The effect size for this difference was medium ($ES = 0.42$, 95% CI 0.08–0.92). A similar pattern was also found in the precuneus area, although it was nonsignificant, with BDNF Met carriers and APOE E4 carriers showing decreased thickness compared to Val/Val homozygotes and APOE E3 homozygotes ($F_{1,41} = 5.47$, $p = 0.11$) (Fig. 1B). Nevertheless, effect size for this difference was medium ($ES = 0.46$, 95% CI 0.07–0.97). Regarding cognitive performance, all measures were above the statistical threshold set for significance. Results of the linear mixed models for brain morphometry and cognitive measures are summarized in Tables 2 and 3. No differences between BDNF Val/Val homozygotes and Met Carriers were evident at baseline for both HS and MCI/AD individuals.

3.3. Impact of BDNF on brain integrity and cognitive markers in MCI/AD patients

In the MCI/AD sample, a BDNF \times Time interaction effect was evident for entorhinal thickness ($F_{3,298} = 5.16$, $p = 0.004$), that resulted in a marginally significant effect in the APOE E4 carriers subgroup ($F_{3,182} = 3.26$, $p = 0.06$), that is, BDNF Met carriers and APOE E4 carriers showed greater atrophy compared to Val/Val homozygotes and APOE E3 homozygotes over 3 years in the entorhinal cortex (Fig. 2A). Effect sizes went from 0.48 to 1.03 (medium to large) between baseline and third year of follow-up in BDNF Met

Table 1
Sociodemographic and clinical status characteristics

Sociodemographic and clinical variables	HS (N = 175)			MCI/AD (N = 222)		
	BDNF Val/Val, N = 120	BDNF met carriers, N = 55	Statistical test	BDNF Val/Val, N = 153	BDNF met carriers, N = 69	Statistical test
Age, mean (SD)	76 (5), range: 60–88	76 (5), range: 63–90	$t_{173} = -0.12$, $p = 0.91$	75 (7), range: 55–91	75 (7), range: 55–88	$t_{220} = -0.24$, $p = 0.80$
Gender M/F	62/58	31/24	$\chi^2 = 0.33$, $p = 0.56$	59/94	34/35	$\chi^2 = 2.24$, $p = 0.13$
Education, mean (SD)	16 (2), range: 10–20	16 (3), range: 6–20	$t_{173} = 0.43$, $p = 0.67$	15 (3), range: 6–20	15 (3), range: 8–20	$t_{220} = -0.60$, $p = 0.55$
CDR-SB, Mean (SD)	0 (0), range: 0–1.5	0 (0), range: 0–1.5	$t_{167} = -0.31$, $p = 0.75$	2.5 (1.6), range: 0.5–9	2.7 (1.6), range: 0.5–8	$t_{220} = -0.39$, $p = 0.70$

Key: AD, Alzheimer's disease; BDNF, brain-derived neurotrophic factor; CDR, Clinical Dementia Rating; HS, Healthy subjects; MCI, mild cognitive impairment; SD, standard deviation.

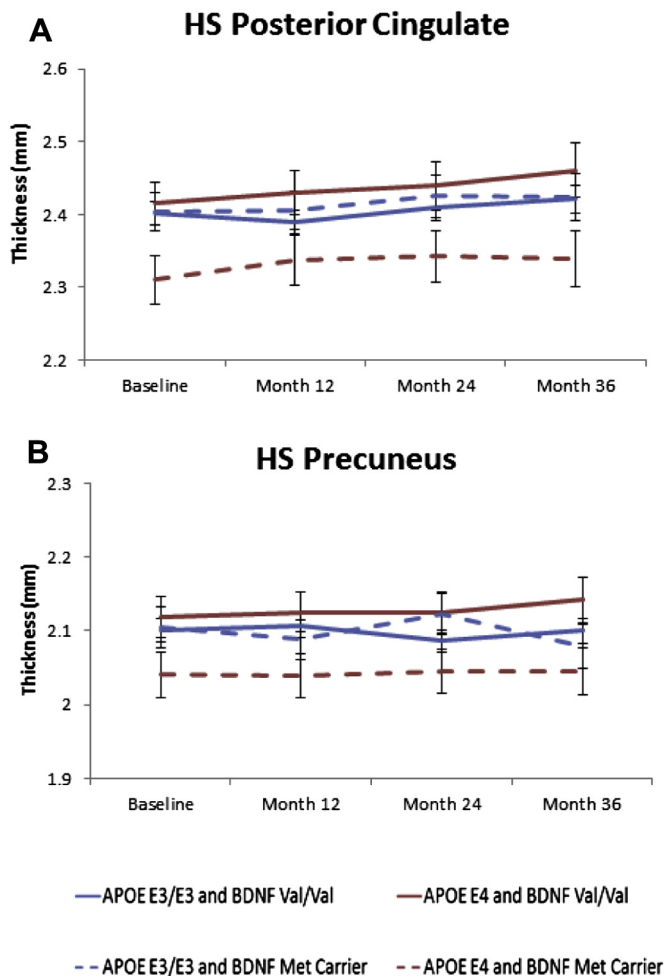


Fig. 1. Posterior cingulate and precuneus thickness by APOE and BDNF genotype in HS. (A) Least square means from longitudinal mixed models of posterior cingulate thickness in HS according to APOE (E3/E3 and E4 carriers) and BDNF (Val/Val and Met carriers) interaction subgroups across 3 years of follow-up (BDNF main effect: $F_{1, 41} = 7.99$, $p = 0.07$, ES = 0.42); (B) Least square means from longitudinal mixed models of longitudinal precuneus thickness in HS according to APOE (E3/E3 and E4 carriers) and BDNF (Val/Val and Met carriers) interaction subgroups across 3 years of follow-up (BDNF main effect: $F_{1, 41} = 5.47$, $p = 0.11$, ES = 0.46). All p values have been FDR corrected. Error bars represent standard errors of the mean. Abbreviations: APOE, apolipoprotein E; BDNF, brain-derived neurotrophic factor; ES, effect size; FDR, false discovery rate; HS, healthy subject.

and APOE E4 carriers, as compared to 0.31 to 0.77 in Val/Val homozygotes. As expected in the context of MCI/AD neuropathology, several brain areas showed an effect of time, that is, decreasing volume or thickness through 3 years: cortical volume, hippocampus, and other key regions of the temporal and parietal cortex. This atrophy pattern was evident independent of APOE genotype (i.e., both APOE subgroups showed significant atrophy over time). However, this was not the case for frontal cortex thickness measures, where only APOE E4 carriers showed significant atrophy, specifically in regions such as the rostral part of the anterior cingulate cortex, middle frontal area, lateral and medial orbito-frontal cortex (see [Supplement](#)).

Regarding cognition, we found several measures that showed a main effect of BDNF genotype in the APOE E4 subgroup: Logical memory immediate ($F_{1, 134} = 5.63$, $p = 0.05$) ([Fig. 2B](#)), digit span ($F_{1, 134} = 4.95$, $p = 0.06$) ([Fig. 2C](#)), ADAS memory ($F_{1, 134} = 5.72$, $p = 0.05$) ([Fig. 2D](#)), and semantic fluency ($F_{1, 134} = 5.53$, $p = 0.05$) ([Fig. 2E](#)). In all cases, BDNF Met carriers showed poorer performance compared to Val/Val homozygotes. Differences in effect sizes

were medium for all 3 measures: 0.38 (95% CI 0.02–0.74) for logical memory immediate, 0.36 (95% CI 0.00–0.72) for digit span, 0.26 (95% CI 0.10–0.62) for ADAS memory, and 0.38 (95% CI 0.02–0.74) for semantic fluency. Practically, all cognitive measures showed an effect of Time irrespective of APOE status, suggesting, as expected, a lack of the ability to benefit from practice of repeated cognitive testing in established AD as well as in MCI progressors, and/or frank decline. Results of the linear mixed models for brain morphometry and cognitive measures are displayed in [Tables 2 and 3](#).

4. Discussion

Taken together, our findings suggest lack of compensatory mechanisms in carriers of the BDNF Met allele and carriers of the APOE E4 allele. In the context of healthy aging, posterior cingulate and precuneus thickness showed a trend for significantly decreases in BDNF Met carriers compared to Val/Val homozygotes in the APOE E4 carriers subgroup. In MCI/AD carriers of the BDNF Met allele and APOE E4 allele, a significantly steeper rate of atrophy of the entorhinal cortex over 3 years was evident compared to Val/Val homozygotes. This was accompanied by a significant impairment in several cognitive functions (specifically memory and semantic fluency).

Posterior cingulate and precuneus are considered to be important brain regions associated specifically with preclinical AD. Additionally, these are key regions forming part of the default mode network that has been found to be disrupted in pathological aging ([Greicius et al., 2004](#); [Rami et al., 2012](#); [Sperling et al., 2014](#)). The BDNF effects in APOE E4 carriers that we have found in our sample, that is, Met carriers having thinner posterior cingulate and precuneus cortex, suggest compromises in this neurotrophic factor in one of the brain regions undergoing primary manifestations of pathological aging associated to AD. Indeed, APOE E4 increases amyloid and tau misprocessing plus brain atrophy in healthy older adults and in MCI. This is broadly consistent with Lim et al. findings in the Australian Imaging Biomarkers & Lifestyle Study of Aging that concluded that BDNF effects were conditioned by the presence and/or absence of amyloid ([Lim et al., 2015](#)).

Entorhinal cortex is a key region in both memory formation ([Squire et al., 2004](#)) and development of AD pathophysiology ([Desikan et al., 2009](#); [Small et al., 2011](#)). It seems to be also a crucial area for BDNF expression, as Nagahara et al. demonstrated that increasing BDNF expression in the entorhinal cortex ameliorates neurodegeneration in animal models of AD and aging ([Nagahara et al., 2009](#)). Furthermore, BDNF protein has been found to be reduced in entorhinal cortex of AD patients (at postmortem) ([Connor et al., 1997](#)). Our findings suggest that improper trafficking and/or reduction of secretion of BDNF in Met carriers may contribute to the progressive neuronal atrophy of the entorhinal cortex in prodromal and established AD.

These findings might reflect a mechanism by which BDNF and APOE contribute to AD-related localized brain atrophy, or alternatively cannot compensate for, or ameliorate AD-related neurodegeneration. It has been suggested that BDNF Met allele may accelerate the progression of AD; BDNF Met effect on longitudinal cognition and hippocampal volume is only present if coupled with high AB amyloid levels ([Lim et al., 2013](#)). Furthermore, epistatic interaction between BDNF and APOE E4 on disease progression in preclinical AD has also been reported ([Adamczuk et al., 2013](#); [Hashimoto et al., 2009](#)). As stated in the Introduction section, this is in contrast to the findings that Met allele may have advantageous effects in the context of traumatic brain injury, although it remains possible that these 2 neuropathological states, one consisting in a one-time trauma and the other consisting of progressive brain changes might result in BDNF Val/Met differential effects on brain morphometry and cognition.

Table 2

Linear mixed models results on brain morphometric measures for HS and MCI/AD groups stratified by APOE

Brain morphometry measures	HS		MCI/AD	
	APOE E3/E3	APOE E4 carriers	APOE E3/E3	APOE E4 carriers
Cerebral cortex volume	BDNF ($F_{1,93} = 0.07/p = 0.79$), Time ($F_{3,173} = 4.01/p = 0.08$), BDNF \times Time ($F_{3,173} = 1.10/p = 0.52$), $N = 97$	BDNF ($F_{1,38} = 0.14/p = 0.94$), Time ($F_{3,71} = 0.16/p = 0.94$), BDNF \times Time ($F_{3,71} = 0.13/p = 0.94$), $N = 42$	BDNF ($F_{1,53} = 0.44/p = 0.72$), Time ($F_{3,56} = 3.82/p = 0.04$), BDNF \times Time ($F_{3,56} = 0.52/p = 0.75$), $N = 57$	BDNF ($F_{1,111} = 0.12/p = 0.78$), Time ($F_{3,135} = 12.42/p < 0.0001$), BDNF \times Time ($F_{3,135} = 1.15/p = 0.55$), $N = 115$
Cerebral white matter volume	BDNF ($F_{1,93} = 3.52/p = 0.19$), Time ($F_{3,173} = 1.79/p = 0.34$), BDNF \times Time ($F_{3,173} = 0.58/p = 0.77$), $N = 97$	BDNF ($F_{1,38} = 0.79/p = 0.94$), Time ($F_{3,71} = 0.73/p = 0.94$), BDNF \times Time ($F_{3,71} = 3.49/p = 0.18$), $N = 42$	BDNF ($F_{1,53} = 0.06/p = 0.80$), Time ($F_{3,56} = 4.40/p = 0.034$), BDNF \times Time ($F_{3,56} = 2.15/p = 0.24$), $N = 57$	BDNF ($F_{1,111} = 0.83/p = 0.55$), Time ($F_{3,135} = 6.75/p < 0.0001$), BDNF \times Time ($F_{3,135} = 0.36/p = 0.78$), $N = 115$
Hippocampus	BDNF ($F_{1,93} = 1.13/p = 0.52$), Time ($F_{3,173} = 3.18/p = 0.11$), BDNF \times Time ($F_{3,173} = 0.49/p = 0.77$), $N = 97$	BDNF ($F_{1,38} = 0.45/p = 0.94$), Time ($F_{3,71} = 0.50/p = 0.94$), BDNF \times Time ($F_{3,71} = 0.19/p = 0.94$), $N = 42$	BDNF ($F_{1,53} = 0.34/p = 0.72$), Time ($F_{3,56} = 5.61/p = 0.02$), BDNF \times Time ($F_{3,56} = 0.71/p = 0.72$), $N = 57$	BDNF ($F_{1,111} = 0.83/p = 0.55$), Time ($F_{3,135} = 35.01/p < 0.0001$), BDNF \times Time ($F_{3,135} = 0.36/p = 0.78$), $N = 115$
Middle temporal thickness	BDNF ($F_{1,99} = 0.00/p = 0.96$), Time ($F_{3,214} = 0.65/p = 0.85$), BDNF \times Time ($F_{3,214} = 0.15/p = 0.96$), $N = 103$	BDNF ($F_{1,39} = 6.08/p = 0.27$), Time ($F_{3,80} = 0.25/p = 0.99$), BDNF \times Time ($F_{3,80} = 0.03/p = 0.99$), $N = 42$	BDNF ($F_{1,57} = 0.14/p = 0.82$), Time ($F_{3,83} = 7.18/p < 0.01$), BDNF \times Time ($F_{3,83} = 0.83/p = 0.65$), $N = 61$	BDNF ($F_{1,121} = 0.85/p = 0.60$), Time ($F_{3,182} = 30.20/p < 0.0001$), BDNF \times Time ($F_{3,182} = 0.96/p = 0.62$), $N = 125$
Inferior temporal thickness	BDNF ($F_{1,99} = 0.24/p = 0.85$), Time ($F_{3,214} = 1.27/p = 0.85$), BDNF \times Time ($F_{3,214} = 1.04/p = 0.85$), $N = 103$	BDNF ($F_{1,39} = 2.23/p = 0.63$), Time ($F_{3,80} = 0.39/p = 0.99$), BDNF \times Time ($F_{3,80} = 0.15/p = 0.99$), $N = 42$	BDNF ($F_{1,57} = 1.38/p = 0.59$), Time ($F_{3,83} = 3.79/p = 0.04$), BDNF \times Time ($F_{3,83} = 1.32/p = 0.59$), $N = 61$	BDNF ($F_{1,121} = 0.31/p = 0.66$), Time ($F_{3,182} = 15.96/p < 0.0001$), BDNF \times Time ($F_{3,182} = 1.65/p = 0.38$), $N = 125$
Parahippocampal	BDNF ($F_{1,99} = 0.04/p = 0.96$), Time ($F_{3,214} = 2.21/p = 0.66$), BDNF \times Time ($F_{3,214} = 0.27/p = 0.96$), $N = 103$	BDNF ($F_{1,39} = 2.23/p = 0.63$), Time ($F_{3,80} = 0.39/p = 0.99$), BDNF \times Time ($F_{3,80} = 0.15/p = 0.99$), $N = 42$	BDNF ($F_{1,57} = 0.02/p = 0.89$), Time ($F_{3,83} = 5.78/p < 0.01$), BDNF \times Time ($F_{3,83} = 0.88/p = 0.65$), $N = 61$	BDNF ($F_{1,121} = 0.02/p = 0.88$), Time ($F_{3,182} = 26.09/p < 0.0001$), BDNF \times Time ($F_{3,182} = 1.50/p = 0.41$), $N = 125$
Entorhinal	BDNF ($F_{1,99} = 1.77/p = 0.80$), Time ($F_{3,214} = 2.58/p = 0.66$), BDNF \times Time ($F_{3,214} = 1.04/p = 0.81$), $N = 103$	BDNF ($F_{1,39} = 1.62/p = 0.63$), Time ($F_{3,80} = 0.01/p = 0.99$), BDNF \times Time ($F_{3,80} = 0.30/p = 0.99$), $N = 42$	BDNF ($F_{1,57} = 0.05/p = 0.89$), Time ($F_{3,83} = 3.74/p = 0.04$), BDNF \times Time ($F_{3,83} = 1.20/p = 0.59$), $N = 61$	BDNF ($F_{1,121} = 0.02/p = 0.88$), Time ($F_{3,182} = 43.17/p < 0.0001$), BDNF \times Time ($F_{3,182} = 3.26/p = 0.06$), $N = 125$
Fusiform	BDNF ($F_{1,99} = 1.31/p = 0.80$), Time ($F_{3,214} = 0.88/p = 0.85$), BDNF \times Time ($F_{3,214} = 0.74/p = 0.85$), $N = 103$	BDNF ($F_{1,39} = 1.62/p = 0.63$), Time ($F_{3,80} = 0.10/p = 0.99$), BDNF \times Time ($F_{3,80} = 1.17/p = 0.82$), $N = 42$	BDNF ($F_{1,57} = 0.55/p = 0.65$), Time ($F_{3,83} = 4.61/p = 0.02$), BDNF \times Time ($F_{3,83} = 0.75/p = 0.66$), $N = 61$	BDNF ($F_{1,121} = 0.37/p = 0.66$), Time ($F_{3,182} = 26.85/p < 0.0001$), BDNF \times Time ($F_{3,182} = 0.70/p = 0.66$), $N = 125$
Posterior cingulate	BDNF ($F_{1,100} = 0.11/p = 0.83$), Time ($F_{3,220} = 1.98/p = 0.53$), BDNF \times Time ($F_{3,220} = 1.22/p = 0.68$), $N = 104$	BDNF ($F_{1,41} = 7.99/p = 0.07$) , Time ($F_{3,99} = 0.39/p = 0.87$), BDNF \times Time ($F_{3,99} = 0.25/p = 0.87$), $N = 45$	BDNF ($F_{1,62} = 0.01/p = 0.92$), Time ($F_{3,104} = 4.43/p = 0.02$), BDNF \times Time ($F_{3,104} = 0.55/p = 0.92$), $N = 66$	BDNF ($F_{1,127} = 0.08/p = 0.84$), Time ($F_{3,215} = 6.84/p < 0.0001$), BDNF \times Time ($F_{3,215} = 0.28/p = 0.84$), $N = 131$
Precuneus	BDNF ($F_{1,100} = 0.00/p = 0.97$), Time ($F_{3,220} = 0.86/p = 0.76$), BDNF \times Time ($F_{3,220} = 5.42/p = 0.01$) , $N = 104$	BDNF ($F_{1,41} = 5.47/p = 0.11$), Time ($F_{3,99} = 0.28/p = 0.87$), BDNF \times Time ($F_{3,99} = 0.24/p = 0.87$), $N = 45$	BDNF ($F_{1,62} = 0.05/p = 0.92$), Time ($F_{3,104} = 2.47/p = 0.15$), BDNF \times Time ($F_{3,104} = 1.41/p = 0.44$), $N = 66$	BDNF ($F_{1,127} = 0.61/p = 0.65$), Time ($F_{3,215} = 16.00/p < 0.0001$), BDNF \times Time ($F_{3,215} = 1.13/p = 0.61$), $N = 131$
Isthmus-cingulate	BDNF ($F_{1,100} = 0.29/p = 0.76$), Time ($F_{3,220} = 0.71/p = 0.76$), BDNF \times Time ($F_{3,220} = 1.58/p = 0.58$), $N = 104$	BDNF ($F_{1,41} = 2.37/p = 0.39$), Time ($F_{3,99} = 0.32/p = 0.87$), BDNF \times Time ($F_{3,99} = 0.99/p = 0.87$), $N = 45$	BDNF ($F_{1,62} = 0.12/p = 0.92$), Time ($F_{3,104} = 5.64/p = 0.01$), BDNF \times Time ($F_{3,104} = 2.45/p = 0.15$), $N = 66$	BDNF ($F_{1,127} = 1.79/p = 0.41$), Time ($F_{3,215} = 17.27/p < 0.001$), BDNF \times Time ($F_{3,215} = 0.33/p = 0.84$), $N = 131$
Rostral anterior cingulate	BDNF ($F_{1,100} = 5.28/p = 0.21$), Time ($F_{3,220} = 1.08/p = 0.82$), BDNF \times Time ($F_{3,220} = 0.09/p = 0.99$), $N = 104$	BDNF ($F_{1,41} = 0.15/p = 0.83$), Time ($F_{3,99} = 0.88/p = 0.82$), BDNF \times Time ($F_{3,99} = 0.99/p = 0.81$), $N = 45$	BDNF ($F_{1,62} = 4.14/p = 0.42$), Time ($F_{3,105} = 2.05/p = 0.45$), BDNF \times Time ($F_{3,105} = 1.96/p = 0.45$), $N = 66$	BDNF ($F_{1,127} = 0.16/p = 0.78$), Time ($F_{3,215} = 4.53/p = 0.01$), BDNF \times Time ($F_{3,215} = 0.53/p = 0.78$), $N = 131$
Caudal anterior cingulate	BDNF ($F_{1,100} = 1.31/p = 0.82$), Time ($F_{3,220} = 0.47/p = 0.99$), BDNF \times Time ($F_{3,220} = 1.69/p = 0.77$), $N = 104$	BDNF ($F_{1,41} = 5.86/p = 0.18$), Time ($F_{3,99} = 0.30/p = 0.83$), BDNF \times Time ($F_{3,99} = 2.09/p = 0.48$), $N = 45$	BDNF ($F_{1,62} = 0.31/p = 0.74$), Time ($F_{3,105} = 1.06/p = 0.60$), BDNF \times Time ($F_{3,105} = 1.17/p = 0.58$), $N = 66$	BDNF ($F_{1,127} = 4.38/p = 0.11$), Time ($F_{3,215} = 1.56/p = 0.45$), BDNF \times Time ($F_{3,215} = 1.17/p = 0.58$), $N = 131$
Rostral middle frontal	BDNF ($F_{1,100} = 0.88/p = 0.82$), Time ($F_{3,220} = 0.19/p = 0.99$), BDNF \times Time ($F_{3,220} = 0.44/p = 0.99$), $N = 104$	BDNF ($F_{1,41} = 4.10/p = 0.30$), Time ($F_{3,99} = 1.15/p = 0.81$), BDNF \times Time ($F_{3,99} = 0.39/p = 0.83$), $N = 45$	BDNF ($F_{1,62} = 0.02/p = 0.89$), Time ($F_{3,105} = 2.95/p = 0.42$), BDNF \times Time ($F_{3,105} = 1.52/p = 0.55$), $N = 66$	BDNF ($F_{1,127} = 0.01/p = 0.95$), Time ($F_{3,215} = 12.56/p < 0.0001$), BDNF \times Time ($F_{3,215} = 0.99/p = 0.65$), $N = 131$
Caudal middle frontal	BDNF ($F_{1,100} = 0.83/p = 0.82$), Time ($F_{3,220} = 1.68/p = 0.77$), BDNF \times Time ($F_{3,220} = 3.66/p = 0.21$), $N = 104$	BDNF ($F_{1,41} = 6.42/p = 0.18$), Time ($F_{3,99} = 0.38/p = 0.83$), BDNF \times Time ($F_{3,99} = 0.99/p = 0.81$), $N = 45$	BDNF ($F_{1,62} = 0.05/p = 0.88$), Time ($F_{3,105} = 2.42/p = 0.42$), BDNF \times Time ($F_{3,105} = 1.68/p = 0.53$), $N = 66$	BDNF ($F_{1,127} = 1.24/p = 0.53$), Time ($F_{3,215} = 11.69/p < 0.0001$), BDNF \times Time ($F_{3,215} = 0.66/p = 0.78$), $N = 131$
Lateral orbitofrontal	BDNF ($F_{1,100} = 0.00/p = 1.00$), Time ($F_{3,220} = 0.85/p = 0.94$), BDNF \times Time ($F_{3,220} = 0.48/p = 0.99$), $N = 104$	BDNF ($F_{1,41} = 1.02/p = 0.81$), Time ($F_{3,99} = 0.67/p = 0.83$), BDNF \times Time ($F_{3,99} = 0.40/p = 0.81$), $N = 45$	BDNF ($F_{1,62} = 0.05/p = 0.88$), Time ($F_{3,105} = 1.39/p = 0.56$), BDNF \times Time ($F_{3,105} = 0.53/p = 0.80$), $N = 66$	BDNF ($F_{1,127} = 2.33/p = 0.33$), Time ($F_{3,215} = 7.87/p < 0.0001$), BDNF \times Time ($F_{3,215} = 0.60/p = 0.78$), $N = 131$
Medial orbitofrontal	BDNF ($F_{1,100} = 0.08/p = 1.00$), Time ($F_{3,220} = 0.32/p = 0.99$), BDNF \times Time ($F_{3,220} = 0.18/p = 1.00$), $N = 104$	BDNF ($F_{1,41} = 0.05/p = 0.83$), Time ($F_{3,99} = 0.39/p = 0.83$), BDNF \times Time ($F_{3,99} = 1.29/p = 0.81$), $N = 45$	BDNF ($F_{1,62} = 0.34/p = 0.74$), Time ($F_{3,105} = 1.25/p = 0.58$), BDNF \times Time ($F_{3,105} = 0.99/p = 0.60$), $N = 66$	BDNF ($F_{1,127} = 0.00/p = 0.95$), Time ($F_{3,215} = 9.76/p < 0.0001$), BDNF \times Time ($F_{3,215} = 0.57/p = 0.78$), $N = 131$

BDNF significant and trend effects are in bold ($p < 0.10$); All p values have been FDR corrected.

Key: AD, Alzheimer's disease; APOE, apolipoprotein E; BDNF, brain-derived neurotrophic factor; FDR, false discovery rate; HS, healthy subject; MCI, mild cognitive impairment.

Table 3

Linear mixed models results on cognitive measures for HS and MCI/AD groups stratified by APOE

Cognitive measures	HS		MCI/AD	
	APOE E3/E3, N = 106	APOE E4 carriers, N = 45	APOE E3/E3, N = 70	APOE E4 carriers, N = 138
MMSE	BDNF ($F_{1,102} = 0.00/p = 1.00$), Time ($F_{3,291} = 0.18/p = 0.99$), BDNF \times Time ($F_{3,291} = 1.37/p = 0.99$)	BDNF ($F_{1,41} = 0.24/p = 0.86$), Time ($F_{3,119} = 0.46/p = 0.87$), BDNF \times Time ($F_{3,119} = 0.05/p = 0.98$)	BDNF ($F_{1,66} = 0.20/p = 0.77$), Time ($F_{3,155} = 19.74/p < 0.001$), BDNF \times Time ($F_{3,155} = 0.42/p = 0.77$)	BDNF ($F_{1,134} = 1.92/p = 0.30$), Time ($F_{3,315} = 45.68/p < 0.0001$), BDNF \times Time ($F_{3,315} = 0.94/p = 0.55$)
Logical memory immediate	BDNF ($F_{1,102} = 2.61/p = 0.65$), Time ($F_{3,291} = 5.45/p = 0.04$), BDNF \times Time ($F_{3,291} = 0.43/p = 0.99$)	BDNF ($F_{1,41} = 1.38/p = 0.63$), Time ($F_{3,119} = 2.28/p = 0.50$), BDNF \times Time ($F_{3,119} = 1.15/p = 0.63$)	BDNF ($F_{1,66} = 3.81/p = 0.15$), Time ($F_{3,154} = 6.59/p = 0.001$), BDNF \times Time ($F_{3,154} = 0.65/p = 0.77$)	BDNF ($F_{1,134} = 5.63/p = 0.05$) , Time ($F_{3,313} = 19.16/p < 0.0001$), BDNF \times Time ($F_{3,313} = 0.10/p = 0.96$)
ADAS memory	BDNF ($F_{1,102} = 0.47/p = 0.99$), Time ($F_{3,289} = 0.57/p = 0.99$), BDNF \times Time ($F_{3,289} = 1.28/p = 0.99$)	BDNF ($F_{1,41} = 0.03/p = 0.90$), Time ($F_{3,119} = 2.64/p = 0.40$), BDNF \times Time ($F_{3,119} = 1.82/p = 0.63$)	BDNF ($F_{1,66} = 0.37/p = 0.77$), Time ($F_{3,152} = 16.78/p < 0.001$), BDNF \times Time ($F_{3,152} = 0.37/p = 0.77$)	BDNF ($F_{1,134} = 5.72/p = 0.05$) , Time ($F_{3,313} = 28.45/p < 0.0001$), BDNF \times Time ($F_{3,313} = 0.67/p = 0.71$)
Logical memory delayed	BDNF ($F_{1,102} = 3.50/p = 0.48$), Time ($F_{3,290} = 0.81/p = 0.99$), BDNF \times Time ($F_{3,290} = 0.24/p = 0.99$)	BDNF ($F_{1,41} = 2.32/p = 0.63$), Time ($F_{3,119} = 0.77/p = 0.77$), BDNF \times Time ($F_{3,119} = 1.53/p = 0.63$)	BDNF ($F_{1,66} = 2.35/p = 0.28$), Time ($F_{3,149} = 5.95/p < 0.01$), BDNF \times Time ($F_{3,149} = 1.87/p = 0.28$)	BDNF ($F_{1,134} = 3.27/p = 0.15$), Time ($F_{3,312} = 7.74/p < 0.0001$), BDNF \times Time ($F_{3,312} = 1.64/p = 0.30$)
ADAS no memory	BDNF ($F_{1,102} = 0.09/p = 0.99$), Time ($F_{3,286} = 0.98/p = 0.99$), BDNF \times Time ($F_{3,286} = 1.06/p = 0.99$)	BDNF ($F_{1,41} = 0.95/p = 0.63$), Time ($F_{3,117} = 2.72/p = 0.40$), BDNF \times Time ($F_{3,117} = 0.45/p = 0.87$)	BDNF ($F_{1,66} = 0.53/p = 0.77$), Time ($F_{3,149} = 21.20/p < 0.001$), BDNF \times Time ($F_{3,149} = 1.04/p = 0.67$)	BDNF ($F_{1,134} = 0.99/p = 0.46$), Time ($F_{3,306} = 30.63/p < 0.0001$), BDNF \times Time ($F_{3,306} = 0.11/p = 0.96$)
Digit span	BDNF ($F_{1,102} = 0.02/p = 0.99$), Time ($F_{3,291} = 0.50/p = 0.99$), BDNF \times Time ($F_{3,291} = 0.40/p = 0.99$)	BDNF ($F_{1,41} = 0.13/p = 0.87$), Time ($F_{3,119} = 1.13/p = 0.63$), BDNF \times Time ($F_{3,119} = 0.91/p = 0.74$)	BDNF ($F_{1,66} = 0.13/p = 0.77$), Time ($F_{3,152} = 12.37/p < 0.001$), BDNF \times Time ($F_{3,152} = 2.25/p = 0.21$)	BDNF ($F_{1,134} = 4.95/p = 0.06$) , Time ($F_{3,310} = 15.43/p < 0.0001$), BDNF \times Time ($F_{3,310} = 0.17/p = 0.96$)
Digit symbol	BDNF ($F_{1,102} = 0.11/p = 0.99$), Time ($F_{3,288} = 2.99/p = 0.32$), BDNF \times Time ($F_{3,288} = 0.05/p = 0.99$)	BDNF ($F_{1,41} = 0.60/p = 0.74$), Time ($F_{3,119} = 1.26/p = 0.63$), BDNF \times Time ($F_{3,119} = 0.38/p = 0.88$)	BDNF ($F_{1,66} = 0.15/p = 0.77$), Time ($F_{3,148} = 14.63/p < 0.001$), BDNF \times Time ($F_{3,148} = 0.73/p = 0.77$)	BDNF ($F_{1,134} = 1.02/p = 0.46$), Time ($F_{3,308} = 20.45/p < 0.0001$), BDNF \times Time ($F_{3,308} = 0.34/p = 0.89$)
Semantic fluency	BDNF ($F_{1,102} = 0.54/p = 0.99$), Time ($F_{3,291} = 0.71/p = 0.99$), BDNF \times Time ($F_{3,291} = 1.16/p = 0.99$)	BDNF ($F_{1,41} = 1.19/p = 0.63$), Time ($F_{3,119} = 0.84/p = 0.75$), BDNF \times Time ($F_{3,119} = 0.24/p = 0.90$)	BDNF ($F_{1,66} = 0.14/p = 0.77$), Time ($F_{3,154} = 9.06/p < 0.001$), BDNF \times Time ($F_{3,154} = 0.38/p = 0.77$)	BDNF ($F_{1,134} = 5.53/p = 0.05$) , Time ($F_{3,311} = 38.99/p < 0.0001$), BDNF \times Time ($F_{3,311} = 2.00/p = 0.21$)
Trail A	BDNF ($F_{1,102} = 0.00/p = 0.99$), Time ($F_{3,290} = 3.72/p = 0.18$), BDNF \times Time ($F_{3,290} = 0.22/p = 0.99$)	BDNF ($F_{1,41} = 0.17/p = 0.87$), Time ($F_{3,119} = 3.76/p = 0.19$), BDNF \times Time ($F_{3,119} = 0.26/p = 0.90$)	BDNF ($F_{1,66} = 1.06/p = 0.58$), Time ($F_{3,148} = 7.60/p < 0.001$), BDNF \times Time ($F_{3,148} = 1.70/p = 0.34$)	BDNF ($F_{1,134} = 0.65/p = 0.55$), Time ($F_{3,308} = 15.55/p < 0.0001$), BDNF \times Time ($F_{3,308} = 0.34/p = 0.89$)
Trail B	BDNF ($F_{1,102} = 0.19/p = 0.99$), Time ($F_{3,290} = 1.59/p = 0.95$), BDNF \times Time ($F_{3,290} = 0.51/p = 0.99$)	BDNF ($F_{1,41} = 1.31/p = 0.63$), Time ($F_{3,117} = 6.33/p = 0.01$), BDNF \times Time ($F_{3,117} = 1.36/p = 0.63$)	BDNF ($F_{1,66} = 0.13/p = 0.77$), Time ($F_{3,132} = 7.72/p < 0.001$), BDNF \times Time ($F_{3,132} = 0.46/p = 0.77$)	BDNF ($F_{1,134} = 1.28/p = 0.41$), Time ($F_{3,274} = 16.12/p < 0.0001$), BDNF \times Time ($F_{3,274} = 0.52/p = 0.80$)

BDNF significant and trend effects are in bold ($p < 0.10$); All p values have been FDR corrected.

Key: AD, Alzheimer's disease; APOE, apolipoprotein E; BDNF, brain-derived neurotrophic factor; FDR, false discovery rate; HS, healthy subject; MCI, mild cognitive impairment; MMSE, Mini-Mental State Examination.

In addition, our findings also highlight the important role of BDNF genotype in episodic memory performance (Goldberg et al., 2008; Hariri et al., 2003), especially in the context of neurodegeneration in APOE E4 individuals. In our MCI/AD sample, we found episodic memory (plus semantic fluency) to be impaired in Met carriers compared to Val/Val homozygotes, as others have also found (Lim et al., 2014), but interestingly in our study only in APOE E4 carriers. Consistent with prior research, logical memory is probably one of the more sensitive measures for identifying BDNF genotypic modulatory effects on memory. However, we did not find BDNF/APOE effects on episodic memory in our older healthy sample, as others have recently reported using cross-sectional data; It should be noted that in this study, APOE E2 carriers seem to drive the difference found in episodic memory performance (Ward et al., 2014). Semantic fluency performance is thought to engage temporal lobe areas (Henry and Crawford, 2004; Troyer et al., 1998) that are known to be brain regions in which BDNF expression is high.

One of the main conclusions that might be drawn from this set of data is that in the examination of BDNF genotype effects on temporal lobe morphometry and memory performance, APOE status should be considered along with BDNF genotype. Recently, taking an analytic approach in which HS, MCI, and AD individuals were

combined, Honea et al. in ADNI context found a BDNF interaction with age in whole-brain volume (Older Val/Val had smaller volume), and greater rate of hippocampal and whole-brain atrophy over 2 years (Honea et al., 2013), such that Val/Met declined more quickly. Their analytic strategy was to pool HS, MCI, and AD individuals. Alternatively, we took a different approach, first by examining diagnostic groups separately, and second by stratifying by APOE genotype. This strategy allowed us to observe APOE modulatory effects on BDNF that otherwise would have been masked by pooling the diagnostic groups. Our findings are also broadly consistent with a similar set of conclusions drawn from another recent study for the Australian Imaging Biomarkers & Lifestyle Study of Aging that found faster episodic memory decline in healthy subjects with both BDNF Met and APOE E4 alleles who were amyloid positive (Lim et al., 2015). As our APOE stratification analysis indicated, positive BDNF findings were uniquely evident in APOE E4 carriers. Therefore, this signal points toward a greater deleterious effect of Met BDNF only with an APOE E4 background, suggesting that both genetic factors interact to impact cortical morphology of important brain regions associated with pathological aging. In this respect, it is important to note that E4 produces robust decreases in CSF AB and increases in tau in both HS and MCI groups in ADNI (Conejero-Goldberg et al., 2014; Schuff et al., 2009; Shaw et al., 2009).

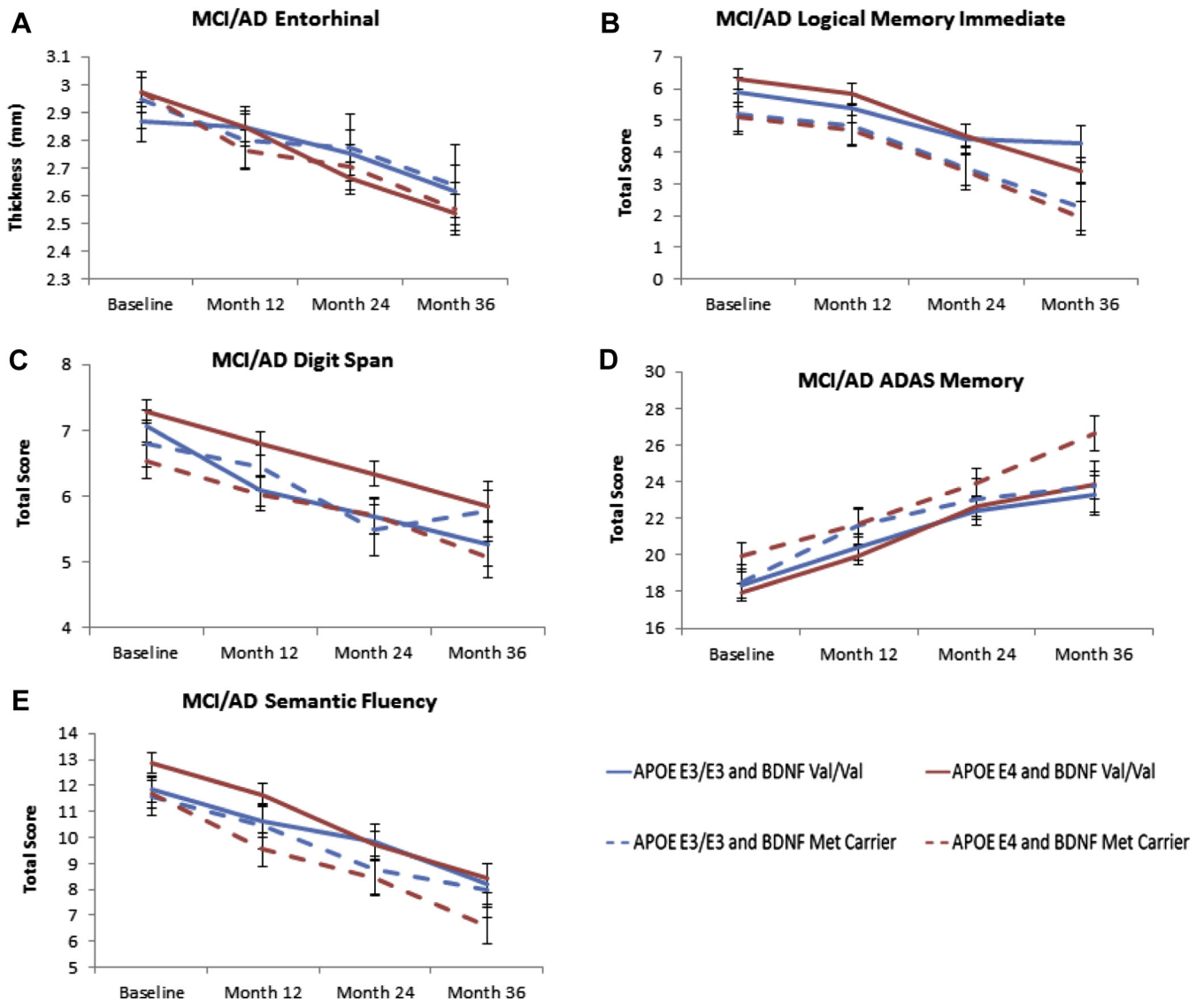


Fig. 2. APOE and BDNF interaction effects in entorhinal thickness and cognitive measures in MCI/AD patients. (A) Least square means from longitudinal mixed models of entorhinal atrophy over 3 years in MCI/AD according to APOE (E3/E3 and E4 carriers) and BDNF (Val/Val and Met carriers) interaction subgroups (BDNF \times Time effect: $F_{3,182} = 3.26$, $p = 0.06$); (B) Least square means from longitudinal mixed models of logical memory immediate scores in MCI/AD according to APOE (E3/E3 and E4 carriers) and BDNF (Val/Val and Met carriers) interaction subgroups (BDNF main effect: $F_{1,134} = 5.63$, $p = 0.05$); (C) Least square means from longitudinal mixed models of working memory (digit span) showed that APOE E4 and BDNF Met carriers were also more impaired compared to APOE E3/E3 and BDNF Val/Val (BDNF main effect: $F_{1,134} = 4.95$, $p = 0.06$); (D) Least square means from longitudinal mixed models of ADAS memory score showed that BDNF Met carriers had higher scores (higher scores correspond to worse performance) compared to Val homozygotes in the APOE E4 subgroup compared to APOE E3/E3 (BDNF main effect: $F_{1,134} = 5.72$, $p = 0.05$); (E) Similar result for least square means from longitudinal mixed models of semantic fluency, where BDNF Met carriers showed lower score compared to Val/Val in the APOE E4 subgroup compared to APOE E3/E3 (BDNF main effect: $F_{1,134} = 5.53$, $p = 0.05$). All p values have been FDR corrected. Error bars represent standard errors of the mean. Abbreviations: APOE, apolipoprotein E; BDNF, brain-derived neurotrophic factor; FDR, false discovery rate; MCI, mild cognitive impairment.

Several caveats in our study must be acknowledged. First, longitudinal follow-up may also be too short to have sensitively detected more generalized BDNF patterns of brain atrophy and cognitive decline. Second, cell size at the 2 levels of stratification of genotype association (BDNF and APOE genotype) could have been too small for detecting subtle effects. We acknowledge the possibility of false positive errors and the moderate effect size d value. Finally, we were unable to directly study the effect of brain amyloid burden; however, given that APOE E4 decreases CSF AB, we believe APOE might act as marker for AB-related neurodegeneration.

In conclusion, our set of findings suggests lack of neural compensatory mechanisms in BDNF Met carriers and APOE E4 carriers. This was less robust in the brain cortical integrity of healthy aging, but nevertheless in key characteristic areas for

pathological aging (posterior-cingulate/precuneus). In MCI/AD, BDNF/APOE effects were stronger, especially in the form of both entorhinal atrophy and cognitive performance.

Disclosure statement

Dr. Goldberg receives royalties for the use of the Brief Assessment of Cognition in Schizophrenia (BACS) in clinical trials.

Acknowledgements

Data collection and sharing for this project was funded by the Alzheimer's Disease Neuroimaging Initiative (ADNI) (National Institutes of Health Grant U01 AG024904). ADNI is funded by the

National Institute on Aging, the National Institute of Biomedical Imaging and Bioengineering, and through generous contributions from the following: Alzheimer's Association; Alzheimer's Drug Discovery Foundation; BioClinica, Inc.; Biogen Idec; Bristol-Myers Squibb Company; Eisai; Elan Pharmaceuticals, Inc.; Eli Lilly and Company; F. Hoffmann-La Roche Ltd. and its affiliated company Genentech, Inc.; GE Healthcare; Innogenetics, N.V.; IXICO Ltd.; Janssen Alzheimer Immunotherapy Research & Development, LLC.; Johnson & Johnson Pharmaceutical Research & Development LLC.; Medpace; Merck & Co., Inc.; Meso Scale Diagnostics, LLC.; NeuroRx Research; Novartis Pharmaceuticals Corporation; Pfizer Inc.; Piramal Imaging; Servier; Synarc; and Takeda Pharmaceutical Company. The Canadian Institutes of Health Research is providing funds to support ADNI clinical sites in Canada. Private sector contributions are facilitated by the Foundation for the National Institutes of Health (www.fnih.org). The grantee organization is the Northern California Institute for Research and Education, and the study is coordinated by the Alzheimer's disease Cooperative Study at the University of California San Diego. ADNI data are disseminated by the Laboratory for Neuro-Imaging at the University of California, Los Angeles. This research was also supported by the Litwin-Zucker Research Center and by grant RO1 AG038734 (Goldberg TE, PI) from the National Institutes of Health.

Appendix A. Supplementary data

Supplementary data related to this article can be found at the online version at <http://dx.doi.org/10.1016/j.neurobiolaging.2015.12.004>.

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