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Featured Articles

Temporal lobe functional activity and connectivity in young adult *APOE* ε4 carriers

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Abstract

Background: We sought to determine if the *APOE* ε4 allele influences both the functional activation and connectivity of the medial temporal lobes (MTLs) during successful memory encoding in young adults.

Methods: Twenty-four healthy young adults, i.e., 12 carriers and 12 noncarriers of the *APOE* ε4 allele, were scanned in a subsequent-memory paradigm, using event-related functional magnetic resonance imaging. The neuroanatomic correlates of successful encoding were measured as greater neural activity for subsequently remembered versus forgotten task items, or in short, encoding success activity (ESA). Group differences in ESA within the MTLs, as well as whole-brain functional connectivity with the MTLs, were assessed.

Results: In the absence of demographic or performance differences, *APOE* ε4 allele carriers exhibited greater bilateral MTL activity relative to noncarriers while accomplishing the same encoding task. Moreover, whereas ε4 carriers demonstrated a greater functional connectivity of ESA-related MTL activity with the posterior cingulate and other peri-limbic regions, reductions in overall connectivity were found across the anterior and posterior cortices.

Conclusions: These results suggest that the *APOE* ε4 allele may influence not only functional activations within the MTL, but functional connectivity of the MTLs to other regions implicated in memory encoding. Enhanced functional connectivity of the MTLs with the posterior cingulate in young adult ε4 carriers suggests that *APOE* may be expressed early in brain regions known to be involved in Alzheimer's disease, long before late-onset dementia is a practical risk or consideration. These functional connectivity differences may also reflect pleiotropic effects of *APOE* during early development.

Keywords: fMRI; Memory; Genetics; Alzheimer's disease; Functional connectivity; *APOE*

1. Introduction

The *apolipoprotein E* (*APOE*) ε4 allele has long been associated with an increased risk for mild cognitive impairment (MCI) and late-onset Alzheimer's disease (AD) [1–4]. In recent years, functional magnetic resonance imaging (fMRI) studies suggest that older healthy adult ε4 carriers, compared with noncarriers, exhibit a greater magnitude and extent of neural activity when performing memory tasks [5–7; but see 8], suggesting that the presence of the *APOE* ε4 allele is associated with alterations in neural activity. Similar enhancements in signal magnitude during memory performance have been observed in MCI [9; but see 10], suggesting that the

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APOE ε4 carrier-associated changes observed in normal elderly subjects may reflect latent compensatory neural activity [5] or increased cognitive effort [6] when performing episodic memory tasks.

Regarding genotype effects in young adults, results remain unclear regarding what neural or cognitive effects may be associated with *APOE* ε4 carrier status earlier in the lifespan. Bondi et al. [6] suggested that *APOE* ε4 plays an antagonistic pleiotropic [see also 11] role for cognition in early adulthood [see also 11], but serves as a risk factor for dementia in late life [12]. Limited support for *APOE*-related antagonistic pleiotropism comes from a recent study examining the role of *APOE* genotype in the recovery of function after traumatic brain injury in young adults (mean age, 23.91 years). Bondi and collaborators found that ε4 carriers outperformed noncarriers on a range of tests assessing attention, executive function, and episodic memory encoding [13]. These results imply a pleiotropic advantage of the ε4 genotype in developing or using neurocognitive compensatory mechanisms. Expounding on this idea, healthy older adults with the ε4 allele may be able to buffer against episodic memory declines by recruiting additional and compensatory cognitive resources to perform cognitive tasks at a level comparable to that of noncarriers [12]. Moreover, Han and Bondi [12] argued that ''buffering'' may begin early in the lifespan with ε4 carriers cognitively outperforming noncarriers, recruiting increased cognitive and neural mechanisms to accomplish said levels.

Recent work by Shaw et al. (2007) [14] may provide a neuroanatomical basis for the cognitive ''buffering'' proposed by Bondi and colleagues, as they discovered a linear decrease in anterior medial temporal lobe (MTL) grey-matter volume in healthy adolescents as a function of *APOE* allele carrier status (i.e., *APOE* $\varepsilon 2 > \varepsilon 3 > \varepsilon 4$ carriers). Shaw et al. [14] reinforced findings by other researchers that *APOE*, in concert with low-density lipoprotein receptors, may play a fundamental role in cerebral development [15,16], and that carriers of the ε4 allele may inherently have thinner entorhinal cortices, establishing a necessary condition for the cognitive ''buffering'' described by Bondi et al. [6]. In essence, any antagonistic pleiotropy of *APOE* ε4 that might be based on middle-aged neuropsychological performance outcomes [12] may arise only as a byproduct of possible negative effects on cortical development much earlier in the lifespan.

Although these studies suggest a rational for *APOE*-associated activation differences in young adults, the studies themselves did not test functional differences between *APOE* genotypes. In fact, only a limited number of studies have attempted to identify at what developmental stage these genotypically driven brain-activity changes are first evident [17–20]. Further, none determined whether there is an ε4 allele carrier effect in the fMRI activity of healthy young adults or if *APOE*-associated differences in neural activity are related specifically to successful memory performance. The current study used the subsequent-memory paradigm [21], which identifies brain regions showing greater study-phase activity for items that are remembered than for those that are forgotten in a subsequent memory test, to determine if the presence of the ε4 allele was associated with differences in neural correlates governing memory success in healthy young adults.

Studies of functional, structural, and behavioral differences associated with variants of the *APOE* genotype have focused on the MTL, based on abundant evidence identifying the MTL as the earliest site of AD-associated pathology [22]. The MTL is known to be the first region in the development of neurofibrillary tangles that are one of the hallmarks for the staging of AD [23], and one of the first structures to show volumetric changes in AD [e.g., 24]. These MTL-associated histopathological and volumetric changes are also associated with decreased performance on a wide range of memory tasks in older adults [e.g., 25–27]. Based on the evidence linking the MTL to the earliest site AD-associated pathology [28] and recent work suggesting that *APOE*-related structural differences in the MTL may exist as early as late childhood [14], as well as the known role of the MTL in episodic memory abilities [29], our analyses focused primarily on group differences in this region.

Based on this research, we focused our analyses of *APOE* effects on successful memory activity within the MTL. Specifically, we sought to identify genotype differences in encoding success activity (ESA) in the MTL, as well as differences in functional connectivity within this region associated with successful encoding processing. We also sought to test the compensation/pleiotropic theory of Bondi et al. [6] regarding *APOE* by examining whether ε4 carriers would exhibit increased ESA and functional connectivity in the MTL, associated with enhanced behavioral performance.

2. Materials and methods

2.1. Participants

We studied 24 right-handed healthy young adults, genotyped for *APOE* (12 *APOE* ε4 carriers and 12 noncarriers). Participants with a history of neurological difficulties or psychiatric illness, alcoholism, drug abuse, and learning disabilities were excluded from the study. Participants were matched in terms of age, years of education, and a battery of neuropsychological tests taken from the Cambridge Neuropsychological Test Automated Battery (CANTABeclipse, version 2.0; Cambridge Cognition, Ltd., Cambridge, UK; Table 1). All participants provided written, informed consent and received financial compensation for their participation. All experimental procedures were approved by the Duke University Institutional Review Board for the Ethical Treatment of Human Participants.

2.2. Genotyping

TaqMan-based allelic discrimination assay (Applied Biosystems) was used for genotype determination. Two separate genotype assays were used to establish the *APOE* status of a subject: (1) rs429358 334 T/C (Applied Biosystems assay

identification, C_3084793_20), and (2) rs7412 472 T/C (Applied Biosystems assay identification, C_904973_10). Assays were conducted according to the manufacturer's protocol, using 10 ng of DNA from each subject per assay. Fluorescence outputs were quantified in real time, using a 7900HT Fast Real Time PCR System, and the data were analyzed using SDS software version 2.2.2 (Applied Biosystems). The *APOE* genotype assignments were defined as described by Koch et al. [30].

2.3. Stimuli and procedure

Participants encoded 120 emotionally neutral pictures taken from a standardized visual object stimuli set normed for name agreement, image agreement, familiarity, and visual complexity [31]. Participants completed three runs, each of which included 5 on (task) blocks (46 seconds) and 5 off (rest) blocks (44 seconds). During each on-block, participants were presented with eight pictures serially, for 3 seconds each, during which time they were asked to make an animacy decision regarding the image (i.e., living/nonliving). Button responses and response time were recorded, using a magnetically shielded four-button box in the participant's right hand. Stimuli were separated with a jittered inter-stimulus interval (ISI) that ranged from 0 to 6 seconds, to allow the hemodynamic response to facilitate deconvolution of the hemodynamic response. Off-block stimuli consisted of a centered crosshair figure, also used for memory-task on-block ISI periods. Participants were not told that they would be asked to perform a subsequent memory test at a later time, and no corrective feedback was given during the task blocks.

Twenty-four hours after the scanning session, participants were brought back to the laboratory and performed a surprise recognition test. Participants were presented with 200 words serially, and asked to make an old/new decision about each word. One hundred and twenty words represented the set of pictures that were presented during the scanning session, and 80 words represented new (not previously viewed) items. After making each memory decision, participants rated their response confidence on a 6-point scale. The retention delay in the present study (24 hours) was longer than in most studies examining subsequent-memory effects. This was done to equate memory performance between young, healthy older adults and patients with MCI (results to be presented in a - subsequent publication).

2.4. Image acquisition and processing

Images were collected using a 4-T GE scanner (GE Healthcare, UK). Stimuli were presented using liquid crystal display goggles (Resonance Technology, Northridge, CA), and behavioral responses were recorded using a four-button fiberoptic response box (Resonance Technology). Scanner noise was reduced with earplugs, and head motion was minimized using foam pads and a headband. Anatomical scanning began with a T_2-weighted sagittal localizer series. The anterior commissure (AC) and posterior commissure (PC) were identified in a midsagittal slice, and 34 contiguous oblique slices were prescribed parallel to the AC-PC plane. High-resolution T_1-weighted structural images were collected with a 5.3-millisecond echo time (TE), a 12-millisecond repetition time (TR), a 24-cm field of view (FOV), a 256^2 matrix, 68 slices, and a slice thickness of 1.9 mm. Functional images were acquired using an inverse spiral sequence with a 1500-millisecond TR, a 6-millisecond TE, a 24-cm FOV, a 64^2 matrix, and a 60° flip angle. Thirty-four contiguous slices were acquired with the same slice prescription as the anatomical images. The slice thickness was 3.75 mm, resulting in cubic 3.75 mm^3 isotropic voxels. Head motion was assessed before preprocessing. No individual moved more than 3 mm in any direction, in any run.

Data were preprocessed and analyzed using Statistical Parameter Mapping (SPM2; Wellcome Trust Centre for Neuroimaging, London, UK), implemented in MATLAB (MathWorks, Inc., Natick, MA.). Time-series data were corrected for differences in slice acquisition time, motion, and field inhomogeneity. After realigning the full-brain anatomical images and individually checking subject-wise error, we coregistered the functional images to the target structurals. Structural images were then spatially normalized to a standard stereotaxic space, using the Montreal Neurological Institute (MNI; Montreal Neurological Institute, Montreal, Quebec, Canada) templates implemented in SPM2, after which the

Table 1
Demographic and behavioral characteristics

	ε4 carriers (n = 12)	Noncarriers (n = 12)	*P* value*
APOE genotype distribution	11 (3/4)	4 (2/3)	
	1 (4/4)	8 (3/3)	
Gender (female)	7	9	
Mean age (y) (SD)	21.8 (3.3)	20.8 (2.9)	0.44
Neuropsychological Assessment Battery performances†			
Spatial span length (SD)	7.2 (1.5)	7.3 (1.3)	0.89
Intra-extra dimensional set shifting errors (SD)	3.3 (3.2)	5.2 (8.5)	0.49
Paired associate learning errors (SD)	2.7 (3.0)	3.7 (4.8)	0.51
Pattern recognition memory % correct (SD)	91.5 (15.3)	89.2 (10.0)	0.67
Choice reaction time (ms) (SD)	340.7 (37.0)	316.9 (45.9)	0.18
Spatial recognition memory % correct (SD)	87.9 (10.5)	84.6 (10.3)	0.44
Rapid visual information processing hit probability (SD)	0.8 (0.2)	0.8 (0.2)	0.93
fMRI procedure performances			
Animacy decision accuracy (SD)	97 (0.1)	100 (0.0)	0.20
Animacy decision reaction time (ms) (SD)	1010 (210.0)	1090 (250.0)	0.40
Subsequent item memory hit rate (SD)	0.5 (0.2)	0.4 (0.2)	0.44
Subsequent item memory false-alarm rate (SD)	0.3 (0.2)	0.3 (0.1)	0.48

*Independent, two-sample *t*-test comparisons.
†Cambridge Neuropsychological Test Automated Battery (CANTA-Beclipse, version 2.0, Cambridge Cognition, Ltd.) subtests.

resulting transformation matrices were applied to the functional data to warp them to MNI space. Functional volumes were then spatially smoothed using a 10-mm isotropic Gaussian kernel.

2.5. Statistical analysis

Individual trial activity was assessed within the context of a mixed model, to control for error variance and blood oxygen level dependent (BOLD) signal drift in the MTL. The onset of each trial (i.e., subsequently remembered and subsequently forgotten items, as defined below) was modeled with a stick function at each stimulus onset, convolving the neural response with two hemodynamic response functions (HRFs): the first was the canonical HRF, and the second was an HRF shifted forward 1 TR, with the latter orthogonalized to the first so as to attribute any shared variance to the earlier function [32]. In accordance with the typical mixed model, task blocks were also modeled and added with head motion and scanner drift as control factors, all of which were treated as regressors of no interest.

Encoding success activity (ESA) is neural activity modulated by memory performance. ESA is operationalized as activity associated with subsequently remembered > subsequently forgotten items. Subsequent hits are categorized as encoding trials that correctly lead to an ''old'' response, combined with a confidence response of 4 to 6. Subsequent misses are defined as encoding trials, classified as ''new'' or ''old,'' but with low confidence. This approach was used in other subsequent-memory fMRI studies to evaluate the neural correlates of successful encoding under high recollective strength [33,34].

Based on our a priori hypotheses focusing on group differences in the MTL, we constructed and applied an anatomically defined mask of the region of interest (ROI) to the group analyses, using a standard anatomical library [35] available within SPM2. Within each group, ESA was assessed at $P < 0.05$, with a minimum cluster extent threshold of 10 contiguous voxels within the focal MTL ROI. These results were subsequently used as an inclusive mask for identifying group effects at $P < 0.05$ and a minimum cluster size of 10 contiguous voxels. Thus, significant activations were required to pass a two-threshold process: (1) a significant ESA effect (subsequent hit > subsequent miss) within one of the groups ($P < 0.05$ and ≥ 10 voxels); and (2) a group difference in ESA ($P < 0.05$ and ≥ 10 voxels). The conjoint probability after inclusive masking approached $P < 0.0025$ [36–38], but this estimate should be treated with caution, given that the contrasts were not completely independent. As an added methodological check on the ESA paradigm, we also conducted an omnibus, whole-brain ESA analysis, including all participants at $P < 0.005$ and a minimum cluster size of 10 voxels. Results are shown in the Supplementary data (Supplementary Fig. 3 and Supplementary Table 4), and reveal the expected pattern of ESA activity (i.e., left prefrontal cortex, left MTL, parietal cortex, and bilateral occipitotemporal cortex), reinforcing that any differences observed in the a priori MTL ROIs are not attributable to any systematic bias in ESA task engagement between groups. Finally, whole-brain differences in ESA were also examined at a threshold of $P < 0.001$ and an extent of 10 contiguous voxels.

To follow up the foregoing ESA within the MTL, we also investigated group differences in functional connectivity of the MTL associated with successful encoding task activity. We used the MTL regions that exhibited group differences in ESA as seed regions, and examined correlations of these MTL activations with whole-brain activity.* A new analytical model was created in which each task trial was uniquely coded as a separate event. This allows for the activity in the MTL seed region to be correlated with activity in all other voxels in the brain. The validity and utility of this technique to interrogate functional brain networks was confirmed elsewhere [34,39,40]. Because the goal of this analysis was to assess group differences in connectivity for ESA, the analysis was constrained to subsequently remembered trials. Moreover, as a second step, group averages of MTL connectivity on remembered trials were calculated by employing a one-sample *t*-test on the resulting correlation maps across all group members. Group differences were again calculated, using a multiple-contrast approach. A between-group two-sample *t*-test was conducted at $P < 0.05$ with a minimum cluster size of 20 voxels, inclusively masking for effects of interest within each group ($P < 0.05$ and a minimal cluster size of 20 voxels). Thus, as in previous analyses, the resulting activity showing group differences in the functional connectivity with the MTL also had to be confirmed by differences observed in individual groups.

Finally, voxel-based morphometry analyses were conducted in SPM2, in accordance with established methodologies [41,42], to examine for grey-matter and white-matter volume differences between *APOE* $\varepsilon 4$ carrier groups. Modulated grey-matter and white-matter images were smoothed with a 9-mm kernel, and individually assessed for regional grey-matter and white-matter volume differences between groups at $P < 0.001$ and 10 voxels.

3. Results

3.1. Behavioral results

Neuropsychological assessment performances were equivalent between groups on a range of tasks tapping memory skills, information-processing speed, attention, and

*A similar connectivity analysis was conducted by choosing peak voxels from the common ESA regions within the MTL as seeds; see coordinates identified in Supplementary Table 4 and Supplementary Fig. 4. The pattern of connectivity differences between groups closely matched that which was found in the main connectivity analysis [Supplementary Fig. 4]. Results indicate that group differences in MTL functional connectivity are not isolated to regions exhibiting overall differences in activation between groups, but are indicative of overall MTL function within the ESA network.

Table 2
Medial temporal lobe ESA contrasts

Difference ESA contrasts*	Activation locus (Brodmann areas)†	$SPM_{(t)}$	k_E	Local maxima‡		
				x	y	z
(ε4 carriers > noncarriers)						
Right hemisphere	PHG (BA 28/35)/hippocampus	3.11	221	26	−40	2
Left hemisphere	PHG (BA 28/35)/hippocampus	2.42	98	−22	−26	−8
(ε4 carriers < noncarriers)						
	None					

Abbreviations: BA, Brodmann area; MTL, medial temporal lobe; PHG, parahippocampal gyrus.
*Statistical parametric mapping (SPM), independent samples *t*-test contrast of MTL ESA.
†Anatomical and Brodmann area labels based upon coordinate nearest grey matter search of the Talairach Daemon Database.
‡Talairach and Tourneau coordinates.

executive skills. Accuracy and response time to making the in-scanner animacy decision were equivalent between groups. No differences were evident in the hit and false-alarm rates for the subsequent memory task (Table 1).

3.2. Neuroimaging results

3.2.1. Medial temporal lobe differences in ESA

Carriers of the ε4 allele exhibited greater ESA in the bilateral hippocampus and parahippocampal cortex compared with noncarriers (Fig. 1, Table 2). No MTL region exhibited greater ESA for noncarriers.

Outside of the MTL, carriers of the ε4 allele did not exhibit greater ESA in any region, whereas noncarriers exhibited greater ESA only in the right middle temporal gyrus (48x, −19y, −9z voxels = 18).

3.2.2. Group differences in medial temporal lobe connectivity

Carriers of the ε4 allele also exhibited greater functional connectivity with the MTL only within a very limited set of brain regions, but notably regions that were implicated in positron emission tomography studies of *APOE* effects in normal adults across the age span [43,44]. For left MTL connectivity, these included the right hippocampus and left fusiform gyrus/middle temporal gyrus, and for right MTL connectivity, the retrosplenial cortex and bilateral caudate. Noncarriers, on the other hand, exhibited much greater functional connectivity with both the left and right MTL, including regions in the bilateral prefrontal cortex, visual cortex, dorsal posterior cingulate cortex, and anterior cingulate cortex (Fig. 2 and Table 3).

3.2.3. Structural imaging comparisons

No group differences in proportionally scaled, modulated regional grey-matter or white-matter volumes were detected in the MTL or remaining cortex, suggesting that both groups had fairly equitable measures of brain parenchyma in regions showing functional activity and connectivity differences.

4. Discussion

The present study yielded two main findings. Despite exhibiting no significant differences in memory performance or grey-matter volume, young healthy adult *APOE* ε4 carriers exhibited significantly greater ESA in the bilateral hippocampus and parahippocampal gyrus compared with noncarriers.

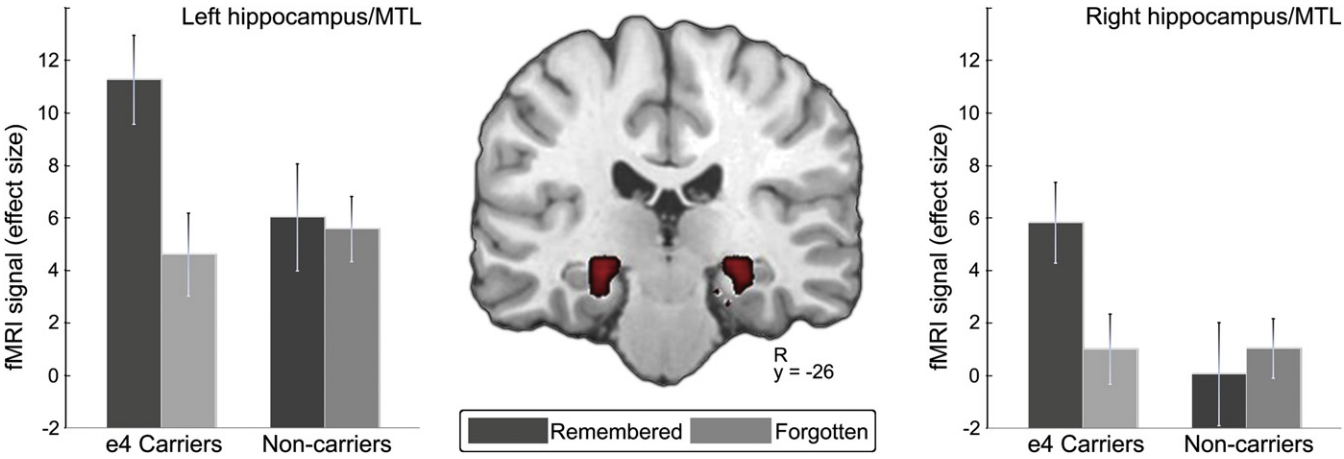

Fig. 1. Group differences in MTL activity predictive of successful subsequent memory (ε4 carriers > noncarriers). Activation bars represent effect sizes extracted from peak voxels within activated region.

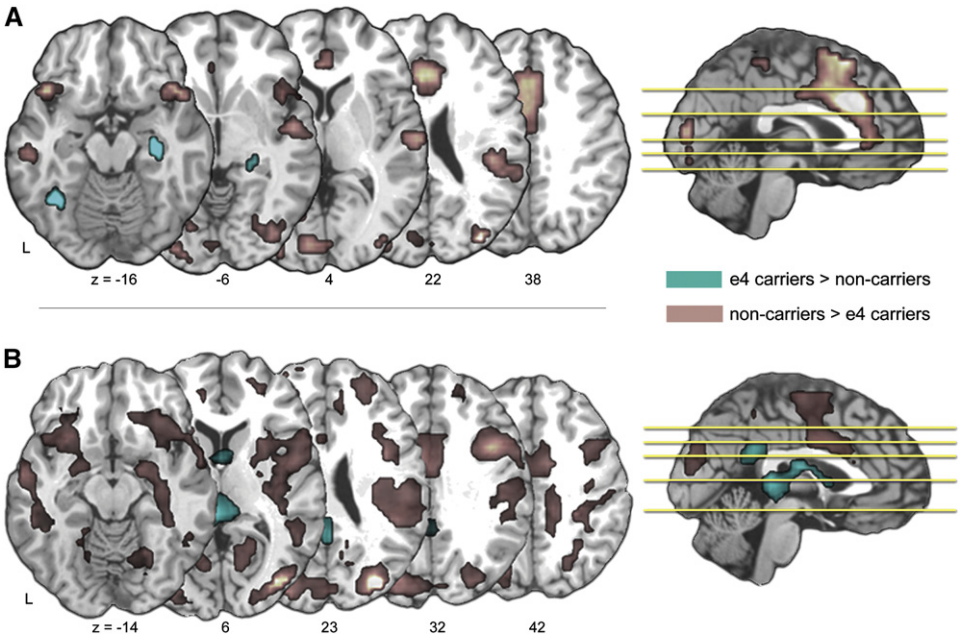

Fig. 2. Whole-brain differences in MTL functional connectivity (seed regions based on regions of interest showing group differences in ESA). (**A**) Functional connectivity to left MTL seed region (Table 2; −22x, −26y, −8z locus). (**B**) Functional connectivity to right MTL seed region (Table 2; 26x, −40y, 2z locus).

In addition, although exhibiting greater overall ESA in these MTL regions, ε4 carriers exhibited globally reduced connectivity to other task-associated cortical regions relative to noncarriers (Table 3). Notably, exceptions to the reduced functional connectivity in ε4 carriers were in regions known to evince some of the earliest functional-activation changes in ε4 adults, the posterior cingulate cortex [43–45]. These results argue against a positive (or antagonistic) pleiotropic role of the *APOE* ε4 allele in successful memory encoding in young adulthood. The ε4 carriers may still derive an early beneficial effect, but results suggest that any window for benefit may be much earlier in the developmental process.

Focusing on our first finding, young adult *APOE* ε4 carriers, compared with noncarriers, exhibited greater ESA activation within the MTL (Fig. 1), specifically within the hippocampus and parahippocampal gyrus. However, both groups exhibited equitable memory performance, and were found to have comparable volumes of grey matter within the MTL, indicating that these functional differences cannot be explained by either differences in memory performance or MTL atrophy. These results are consistent with previous studies in older adults, which also found increased MTL activity for *APOE* ε4 carriers compared with noncarriers [6,46–48]. Those studies concluded that increased activations (in the absence of behavioral and volumetric differences) reflect a compensatory mechanism in *APOE* ε4 carriers. Specifically, ε4 carriers required additional activation, perhaps reflecting additional cognitive effort in memory-related tasks to maintain performances similar to those of noncarriers.

The increased MTL activation in ε4 carriers in the present study may reflect an innate difference in functionality of this region compared with noncarriers, which, in turn, may be indicative of premorbid functional weakness or generally noncontributory hyperactive processing in regions known to be sensitive to late-onset AD pathology [49,50]. The prospect of *APOE*-driven innate differences support the results of Shaw et al. [14] who found a linear decrease in anterior MTL grey-matter volume in healthy adolescents as a function of *APOE* allele carrier status (i.e., *APOE* $\varepsilon 2 > \varepsilon 3 > \varepsilon 4$ carriers). Differences between our results and those of Shaw et al. [14] in regard to MTL volume may be based on differences in methodology (e.g., separation of genotyped groups).

In addition to these group differences in ESA, our second finding focused on evident group differences in functional connectivity with the MTL. Despite exhibiting greater ESA in the bilateral hippocampus, *APOE* ε4 carriers exhibited reduced functional connectivity with these regions, compared with noncarriers. Taken together, the results suggest that *APOE* ε4 noncarriers may be better able to integrate processing mediated by the MTL with that of other brain regions, i.e., regions also involved in ESA. The reduced functional connectivity exhibited by *APOE* ε4 carriers may indicate that activation in this region is modulated relatively independently of other brain regions. The *APOE* ε4 carriers did exhibit increased connectivity with the ventral posterior cingulate cortex, a region exhibiting reduced metabolism and altered activations in those genetically at risk for late-onset AD [44]. In accordance with the hyperactivation of the MTL by *APOE* ε4 carriers, the results may reflect a change in the underlying physiology of this region that is present at an early age. Whether these changes in MTL activation and functional connectivity reflect early changes associated with the disease state or are a causal factor in developing these impairments is a research question that needs to be answered.

Table 3
Whole-brain functional connectivity to medial temporal regions showing increased ε4 carrier effect

Connectivity contrasts*	Activation locus (Brodmann area)†	SPM	k_E	Local maxima		
				x	y	z
Right MTL seed connectivity‡						
ε4 carriers > noncarriers						
Left hemisphere	Retrosplenial/PHG (BA 30)	3.49	1368	−7	−36	9
Left/right hemisphere	Ventral PCC (BA 23)	3.58	266	0	−46	23
ε4 carriers < noncarriers						
Left hemisphere	Middle temporal gyrus (BA 21)	4.39	1702	−59	−8	−9
	Cuneus (BA 18)	4.13	1500	−15	−68	17
	ACC (BA 24)	2.87	446	−4	1	28
	Occipital gyrus (BA 19)	2.42	116	−41	−80	4
Right hemisphere	Middle frontal gyrus (BA 11)	4.61	285	26	25	−17
	Dorsal PCC (BA 31)	4.03	3499	7	−42	44
	Superior temporal gyrus (BA 38)	3.67	431	59	7	−10
	Insula (BA 13)	3.46	712	41	−32	19
	Inferior frontal gyrus (BA 9)	3.30	218	37	9	28
	Superior frontal gyrus (BA 10)	3.11	334	30	48	22
Left MTL seed connectivity§						
ε4 carriers > noncarriers						
Left hemisphere	Inferior temporal gyrus (BA 37)	3.96	1102	−48	−69	0
	Superior parietal lobule (BA 7)	3.23	158	−22	−52	59
Right hemisphere	Fusiform gyrus (BA 37)	3.10	206	48	−59	−10
	Retrosplenial/PHG (BA 30)	2.89	116	19	−40	−4
ε4 carriers < noncarriers						
Left hemisphere	Fusiform gyrus (BA 20)	3.88	945	−45	−23	−18
	Middle frontal gyrus (BA 6)	3.63	446	−30	14	52
	Superior frontal gyrus (BA 10)	3.60	375	−33	55	15
	Superior temporal gyrus (BA 38)	3.41	124	−45	13	−23
	Angular gyrus (BA39)	2.62	146	−48	−64	35
	Superior temporal gyrus (BA 38)	2.48	248	−45	11	−7
Right hemisphere	Superior frontal gyrus (BA9)	3.76	3390	19	52	29
	Inferior frontal gyrus (BA 45)	3.56	319	52	19	6
	Middle temporal gyrus (BA 21)	3.52	930	52	−33	−5
	Dorsal PCC (BA 31)	3.25	116	26	−58	13
	Dorsal entorhinal cortex (BA 34)	2.78	562	19	6	−19

Abbreviations: ACC, anterior cingulate cortex; BA, Brodmann area; PCC, posterior cingulate cortex; PHG, parahippocampal gyrus.
*Statistical parametric mapping (SPM) *t*-test contrast of ESA.
†Anatomical and Brodmann-area labels are based on Talairach-coordinate nearest grey-matter search of Talairach Daemon Database.
‡Connectivity to ε4 carrier > noncarrier ESA right PHG locus (Table 2; coordinate 26x, −40y, 2z).
§Connectivity to ε4 carrier > noncarrier ESA left PHG locus (Table 2; coordinate −22x, −26y, −8z).

Again, whether these functional changes represent a compensatory mechanism of the MTL network, or a more fundamental difference in functional brain organization associated with functional variants of the *APOE* genotype, has yet to be determined. Han and Bondi [12] have argued that the *APOE* gene may have pleiotropic (beneficial) effects in young adults, serving to enhance cognitive performance. Likewise, Mondadori et al. [19] posit that their young adult cohort exhibited beneficially increased neural activity. The present results do not appear to support the notion of pleiotropic effects in memory function by young adulthood, because increased MTL activity and connectivity to the posterior cingulate cortex in our ε4 carriers did not result in enhanced but equitable memory performance compared with noncarriers. Furthermore, our results support a recent finding by Filippini and colleagues [20], who concluded that the presence of the *APOE* ε4 allele was associated with greater MTL activity during encoding in healthy young adults (25 to 30 years old; 18 carriers versus 18 noncarriers) in the absence of behavioral or structural differences between the two groups.

Rather, the present findings fit better with either an inefficiency or a compensatory account of cognitive function. Based on a lack of behavioral differences between genotyped groups, increased MTL activity in ε4 carriers may be interpreted as either (1) inefficient processing associated with dysfunction in the MTL, or (2) compensatory processing necessary to achieve similar cognitive output to that seen in noncarriers who are able to rely on more efficient MTL functioning. These two ideas are not incompatible. The fact that, in ε4 carriers, these task-induced increases in MTL activation are, in turn, associated with reductions in functional connectivity with this region suggest that this enhanced processing is not reflected in other ESA regions. Rather, the observed changes are limited to the MTL and posterior cingulate cortex, another region affected early in AD pathology [51–53], indicating that they may reflect region-specific neural

alterations associated with the ε4 allele. Although the basis for these differences requires future research, the results suggest that investigations should not be limited to individuals in older cohorts.

Furthermore, our compensatory theory (defined as increased processing necessary to achieve similar cognitive function) differs from that of Han and Bondi [12] in that their account is, in actuality, one of enhancement, rather than compensation, in young ε4 carriers. Han and Bondi [12] proposed, ''Young ε4 participants perform better on memory and other neurocognitive tasks.'' They further suggest that increased neural recruitment in older ε4 carriers would help compensate for cognitive declines. Their theory does not outline whether enhanced (not comparable) performance in young carriers is accompanied by increased or comparable neural recruitment compared with noncarriers. The present results involved increased MTL recruitment and *equal* performance in ε4 carriers compared with noncarriers, suggesting either that increased activation is inefficient in some way, or that young ε4 carriers require increased neural processing to accomplish performances similar to those of noncarriers. Further research investigating other cognitive processing associated with MTL function (e.g., retrieval, novelty, or source memory) will be necessary to elucidate fully the validity of the compensation theory. In doing so, it will be crucial to consider not only group differences in activation levels and performance, but also whether cognitive processes may differ between groups. Moreover, the present study used a sample size of 24 participants. Future studies should focus on larger samples to validate the present findings.

Taken together, our findings suggest that the *APOE* ε4 allele influences brain activity earlier in the lifespan than previously reported. Moreover, the presence of the *APOE* ε4 allele appears to alter not only functional activations within the MTL, but also the functional connectivity of the MTL, during the memory-encoding process. Brain regions implicated in this investigation are known to incur structural and functional changes in MCI and AD, and were observed to be altered in healthy middle-aged adults and seniors with the ε4 allele. Although these commonalities existed in our participant sample, it is unclear whether the brain-activation differences evident in the young adult *APOE* ε4 carriers reflected an adaptive response to an underlying weakness in the episodic memory network or a more fundamental, *APOE*-driven pleiotropism in functional brain organization. The results of our functional connectivity analyses suggest the former, because the *APOE* ε4 carriers appeared less able to integrate processing mediated by the MTL with other ESA regions.

Supplementary data

Note: to access the supplementary materials accompanying this article, visit the online version of *Alzheimer's & Dementia* at. www.alzheimersanddemetia.org.

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