HIPPOCAMPUS 21:980–989 (2011)

Brain-Derived Neurotrophic Factor Val66Met Polymorphism and Hippocampal Activation During Episodic Encoding and Retrieval Tasks

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ABSTRACT: Brain-derived neurotrophic factor (BDNF) is a neurotrophin which has been shown to regulate cell survival and proliferation, as well as synaptic growth and hippocampal long-term potentiation. A naturally occurring single nucleotide polymorphism in the human *BDNF* gene (val66met) has been associated with altered intercellular trafficking and regulated secretion of BDNF in met compared to val carriers. Additionally, previous studies have found a relationship between the *BDNF val66met* genotype and functional activity in the hippocampus during episodic and working memory tasks in healthy young adults. Specifically, studies have found that met carriers exhibit both poorer performance and reduced neural activity within the medial temporal lobe (MTL) when performing episodic memory tasks. However, these studies have not been well replicated and have not considered the role of behavioral differences in the interpretation of neural differences. The current study sought to control for cognitive performance in investigating the role of the *BDNF val66met* genotype on neural activity associated with episodic memory. Across item and relational memory tests, met carriers exhibited increased MTL activation during both encoding and retrieval stages, compared to noncarriers. The results suggest that met carriers are able to recruit MTL activity to support successful memory processes, and reductions in cognitive performance observed in prior studies are not a ubiquitous effect associated with variants of the *BDNF val66met* genotype. © **2010 Wiley-Liss, Inc.**

KEY WORDS: BDNF; episodic memory; fMRI; relational memory

INTRODUCTION

The medial temporal lobe (MTL), including the hippocampus, is one of the key brain regions associated with both the encoding and retrieval of episodic memories (Dobbins and Davachi, 2006). Moreover, the involvement of the hippocampus in successful memory processing increases as the complexity of the episodic memory task increases. For example, item memory, which refers to remembering individual features or items associated with what occurred in the past, involves both hippocampal and surrounding MTL activity (e.g., Wagner et al., 1998; Eldridge et al., 2000; Prince et al., in press). However, relational memory, which refers to memory for associations among the items and the context in which they were presented (for a review see Johnson et al., 1993), has been shown to necessitate greater hippocampal activity than simple item memory (e.g., Davachi, 2006; Davachi and Wagner, 2002; Dennis et al., 2008; Eichenbaum et al., 2007; Prince et al., 2005). Moreover, patients with hippocampal damage exhibit deficits on both item and relational memory tasks, with greater deficits found in relational memories (Giovanello et al., 2003; Kan et al., 2007; Kroll et al., 1996; Turriziani et al., 2004). Such evidence supports a role of the hippocampus in the binding of relational memories (e.g., item and contextual information) (e.g., Diana et al., 2007; Mitchell et al., 2006). Furthermore, it has been suggested that, during retrieval, anterior hippocampus is responsible for flexible retrieval of learned associations whereas posterior hippocampus is responsible for exact reinstatement of learned associations (Giovanello et al., 2009). Alternatively, cortical MTL regions have been shown to support memory for component parts of relational memories. For example, the perirhinal cortex has been shown to support encoding of individual items composing relational memories (e.g., Davachi et al., 2003; Kirwan and Stark, 2004; Ranganath et al., 2004), whereas the parahippocampal cortex is posited to support general contextual or spatial information associated with an object (e.g., Bar and Aminoff, 2003; Eichenbaum et al., 2007). However, evidence also exists suggesting a role of the perirhinal cortex in the associative encoding of between multiple feature of a given object (Davachi, 2006). Thus, the integrity of the hippocampus and adjacent MTL structures can have a profound impact on both item and relational memory processing and functional activation within this region.

Recently, research has focused on genetic factors that may contribute to individual differences in memory performance and neural functioning. One candidate gene for understanding individual difference in memory processing is brain-derived neurotrophic factor (*BDNF*). BDNF is a neurotrophin, which has been shown to regulate cell survival and proliferation, as well as synaptic growth and hippocampal long-term potentiation (LTP) in nonhumans (Lu and Gottschalk, 2000; Poo, 2001; Tyler et al., 2002). A natu-

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Grant sponsor: Duke University Provost Common Fund; Grant sponsor: National Institute on Aging grants; Grant numbers: R01 AG019731, R01 AG23770, T32 AG000029.

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Accepted for publication 23 March 2010
DOI 10.1002/hipo.20809
Published online 20 May 2010 in Wiley Online Library (wileyonlinelibrary.com).

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rally occurring polymorphism in the human *BDNF* gene leads to a valine to methionine substitution at position 66 in the prodomain (val66met). This substitution results in differences in cellular distribution of the BDNF protein (Egan et al., 2003). Previous research has suggested that carriers of the 66met variant (vs. val homozygotes) exhibit poorer memory function (e.g., Egan et al., 2003; Hariri et al., 2003; Goldberg et al., 2008), smaller hippocampal volume (e.g., Bueller et al., 2006), and reduced MTL activity in association with both working memory (Egan et al., 2003; Cerasa et al., 2010) and episodic memory tasks (Hariri et al., 2003; Hashimoto et al., 2008). However, associations with cognitive measures have not been supported by genome-wide association studies (Papassotiropoulos et al., 2006; Need et al., 2009), and the individual imaging findings remain to be replicated.

An important goal of neurogenomics as an emerging field is to establish reliability across the relatively small sample sizes typically used in fMRI studies (de Geus et al., 2008). Regarding the role of *BDNF* on episodic memory activity in the MTL, Hariri et al. (2003) observed that, during both encoding and retrieval, met carriers exhibited reduced MTL activation, which was, in turn, associated with reduced memory performance on the episodic memory task. Recently, Hashimoto et al. (2008) replicated Hariri et al.'s finding of reduced hippocampal activity in met carriers during episodic encoding, but failed to find any effect of the polymorphism on retrieval-related activity in an Asian sample. Although this finding strengthens the argument for a genetic effect of this BNDF polymorphism on hippocampal function, it also strengthens the need for more replication studies to sort out the exact nature and mechanism of this effect, as results were somewhat contradictory with the previous literature.

The current study sought to both replicate the original findings by Hariri et al. and expand upon the foregoing research by addressing some of the limitations in the foregoing studies. First, because performance was not equated across the experimental groups (Hariri et al., 2003), it is unclear if the hippocampal effects were independent of performance differences. Performance confounds can cloud interpretation of memory neuroimaging studies when between-groups comparisons are made (e.g., Schlaggar et al., 2002; Daselaar et al., 2003; Dennis et al., 2009). Although Hariri's finding is one of reduced MTL activity associated impaired memory performance, Hashimoto found equivalent memory performance across groups. There are several examples in the literature where reduced neural activity is associated with enhanced or null effects in performance (Dickerson et al., 2004; Duverne et al., 2008), leading to the interpretation that increases in overall neural activations represent inefficient processing. Thus, we sought to clarify the nature of differences in hippocampal activation between groups by controlling for group differences in performance.

Second, as a blocked-design study, the previous studies (Hariri et al., 2003; Hashimoto et al., 2008) were unable to assess effects of the *BDNF* genotype on successful memory activation, which can only be examined with event-related designs that compare activity for remembered vs. forgotten items during encoding and retrieval (e.g., Prince et al., 2005). To address this issue, this study also tested participants' memory using an event-related memory paradigm. To further expand upon previous results, the event-related memory task used a relational memory paradigm in which participants were asked to study and remember the association between two distinct items (i.e., faces and scenes). Compared to item memory, relational memory has been shown to be associated with even greater hippocampal processing (e.g., Davachi and Wagner, 2002; Dennis et al., 2008). Thus, with its use, we can not only investigate the effect of the *BDNF* val66met genotype on successful memory performance, but also its effect on hippocampal activation given a critical need for hippocampal involvement.

Thus, to clarify these issues surrounding the effect of the *BDNF* val66met genotype on MTL, and specifically hippocampal activation and memory performance, the current study focused on three main goals. The first goal was to eliminate behavioral confounds associated with the *BDNF* genotype on memory performance to directly compare genotype differences in MTL activity. The second goal was to attempt to replicate the previous hippocampal finding by Hariri et al. (2003) for item memory using a blocked design. Specifically, we aimed to investigate whether group differences in hippocampal activity during this task were independent of previously reported memory differences between 66met allele carriers and noncarriers. The third goal aimed to examine *BDNF* val66met genotype differences to successful encoding- and retrieval-related hippocampal activations during an event-related relational memory task. With the use of a relational memory task, a memory paradigm which has shown to be more hippocampal-dependent than item memory, we aimed to also expand previous findings of genotype differences by investigating whether the *BDNF* val66met genotype continues to affect hippocampal function as demands on hippocampal processing increases.

MATERIALS AND METHODS

Participants

Twenty-two healthy young adults were chosen from a pool of 175 individuals recruited from the Duke and North Carolina State University communities. They were genotyped for *BDNF* status, and 11 met allele carriers (6 met/met, 5 met/val), and 11 val homozygotes were selected for inclusion in the fMRI study (see Table 1 for participant demographics and neuropsychological test results). Because one goal of the study was to match participants on demographics and cognitive function, the participants were chosen to be as close as possible in age, gender, and ethnicity (We repeated all analyses removing the three individuals of Asian decent. This analysis did not change the pattern of results. Thus all results reported here include all 22 participants.), as well as North American Adult Reading Test (NAART) score and tests of spatial span and spatial working memory taken from the Cambridge Neuropsychological

TABLE 1.

Demographics and Behavioral Results

	Met carriers	Val homozygotes
Demographics		
N	11	11
Male/female	4/7	7/4
Age	22.8	22.3
Neuropsych tasks		
NAART	17.45 (8.4)	16.7 (9.9)
CANTAB		
Spatial span	7.91 (0.39)	8.45 (0.14)
Spatial span errors	10.63 (9.42)	9.55 (21.79)
SWM (errors)	3.91 (6.48)	8.18 (40.15)
SWM (strategy)	25.64 (9.18)	23.36 (6.10)
Item memory task		
Hit rate	0.92(0.11)	0.86(0.10)
False alarm rate	0.08(0.07)	0.05(0.04)
d′	3.70(0.01)	3.62(0.01)
Relational memory task		
High confidence hit rate	41.51(13.40)	42.44(20.12)
High confidence false alarm rate	9.24(6.39)	6.26(6.03)
Low confidence hit rate	33.20(11.19)	29.59(17.35)
Low confidence false alarm rate	25.88(7.94)	17.71(9.18)
Overall d′	1.09(0.51)	1.32(0.38)

NAART, North American Adult Reading Test; CANTAB, Cambridge Neuropsychological Test Automatic Battery; means and standard deviation reported

Test Automated Battery (CANTABeclipse, version 2.0; Cambridge Cognition Ltd). The participants were also balanced according to catechol-O-methlytransferase (*COMT*) val108/158met gene polymorphism (*BDNF* met carriers: six *COMT* val homozgygotes and five *COMT* met homozygotes; *BDNF* val homozygotes: five *COMT* val homozgygotes and six *COMT* met homozygotes). All participants gave written informed consent and received financial compensation. All experimental procedures were approved by the Duke University Institutional Review Board for the ethical treatment of human participants.

Genotyping

Genotyping of the *BDNF* val66met (rs6265) and *COMT* val108/158met (rs4680) SNPs was carried out using the Illumina HumanHap550 or 610quad whole-genome chips (Illumina, San Diego, CA).

Materials

Neuropsychological test

Neuropsychological test materials were comprised of the Cambridge Neuropsychological Test Automated Battery (CANTABeclipse, version 2.0; Cambridge Cognition Ltd), a computerized battery of neuropsychological tests designed to assess verbal and visual episodic and working memory, executive functions, attention, and language using a PC equipped with a touch screen display.

Item memory task

The item memory paradigm was designed to replicate that of Hariri et al. (2003). Participants encoded novel, complex scenes of neutral emotional valence derived from the International Affective Picture System (Lang et al., 1997).

Relational memory task

Relational memory was examined by assessing memory for face-scene pairings. Face stimuli consisted of 172 faces gathered from the adult face database from Dr. Denise Park's lab (Minear and Park, 2004). Scene stimuli consisted of 172 indoor and outdoor scenes gathered from the internet. Using Adobe Photoshop CS2 version 9.0.2 and Irfanview 4.0 (http://www.irfanview.com/), face stimuli were edited to a uniform size (320 × 240 pixels) and background (black), and scene stimuli were standardized to 576 × 432 pixels. Face-scene pairs were created using a custom MatLab (version 6.1) script that overlaid faces on scenes, and images were standardized to 576 × 432 pixels.

Procedure

Blood draws and genotyping, as well as neuropsychological testing, were completed on separate testing days prior to the scanning session. For the imaging session, stimuli were presented via a mirror in the head coil and a rear projection system using a PC computer running Cogent, a stimulus presentation toolbox within Matlab. Button responses and response

TABLE 2.

Greater Medial Temporal Lobe Activity in Met Carriers Compared to Val Homozygotes during Encoding and Retrieval

	H	BA	*x*	*y*	*z*	T	Voxels
Item task							
Encoding vs rest							
Anterior PHG	L	28/35	−23	−16	−22	4.05	12
Hippocampus/PHG	R		23	−19	−15	3.66	59
Retrieval vs rest							
Posterior Hippocampus/PHG	R		23	−30	−5	3.26	30
Relational task							
Encoding success activity							
Hippocampus/PHG	R		26	−27	−18	3.73	22
Retrieval success activity							
Anterior Hippocampus/PHG	L		−23	−12	−15	4.12	15
	R		19	−8	−19	2.89	14
Posterior Hippocampus/PHG	R		30	−30	−2	3.94	27

PHG, parahippocampal gyrus; H, hemisphere; L, left; R, right; BA, Brodmann's area.

times were recorded using an MRI-compatible four-button box held in the participant's right hand.

The item memory task consisted of two 3.23-min runs which alternated between encoding and retrieval blocks, interleaved with rest blocks (Hariri et al., 2003). Each block (rest, encoding, retrieval) lasted 20 s in duration. During encoding, participants viewed six images, presented serially for 3 s each, and were asked to make an indoor/outdoor decision about the image. During retrieval, participants again viewed six images serially for 3 s each (1/2 of the images were previously viewed images and half were new images) and were posed with the question "Have you seen this image before?" They were instructed to answer "yes" for previously viewed images and "no" for new images. During rest blocks, participants were instructed to fixate on a cross presented in the center of the computer screen. Before each block participants viewed an instruction screen (2 s) orienting them to the task at hand. Run order was counter-balanced across participants.

The relational memory task consisted of eight runs alternating between encoding and retrieval. During each encoding run participants saw 48 face-scene pairings and were asked to decide "How appropriate is the fit?" between the face and scene on a four-point scale ranging from "most" to "least" (see Figure 1). The goal of using the goodness-of-fit task during encoding was to assure that participants were attending to both the face and the scene, as well as their relationship with one another. During each retrieval run, 36 intact face-scene pairings were shown as well as 12 lures that were recombinations of previous face-scene pairings. Participants were told that they would be viewing both intact and recombined pairs. They were asked to judge whether the face-scene pair was the same pairing as that which they saw during encoding or whether it was a different face-scene pairing than what they originally viewed (see Figure 1). Participants made memory judgments to each pairing using a four-point scale: "definitely same," "probably same," "probably changed," or "definitely changed." Because the lures consisted of recombined studied items and never any novel items, participants had to encode and remember relationships among the items (i.e., specific or intact face-scene pairings)—memory for the items alone was not sufficient to support correct performance on the recognition test (as all individuals items presented at retrieval were "old" or presented during encoding). Stimuli were each presented for 3 s, and the intertrial jitter ranged from 750 to 1,500 μs to facilitate deconvolution of the hemodynamic response (Dale, 1999). Run order was counter-balanced across participants. The eight task runs each lasted 4 min 8 s, and a 2-min break was given between each run.

Image Acquisition

Images were collected on a General Electric 3.0 Tesla Signa Excite HD short bore scanner (Milwaukee, WI) equipped with an 8-channel head coil. Following acquisition of the high-resolution anatomical images (450-μs repetition time (TR), a 3-μs echo time (TE), a 24-cm field of view (FOV), a 256^2 matrix, and a slice thickness of 1.9 mm), whole-brain functional images were acquired parallel to the anterior-posterior commissure plane using an inverse spiral sequence (direction = interleaved, matrix = 64^2, FOV = 24 cm, TR = 2,000 μs, TE = 30 μs, sections = 34, thickness = 3.8 mm, interscan spacing = 0).

Image Processing

Functional data were preprocessed and analyzed with SPM2 (Statistical Parametric Mapping; Wellcome Department of Cognitive Neurology, http://www.fil.ion.ucl.ac.uk/spm). Time-series data were corrected for differences in slice acquisition times and realigned. Functional images were spatially normalized to a standard stereotaxic space using the Montreal Neurological Institute (MNI) templates implemented in SPM2 and resliced to a resolution of 3.75 mm^3. The coordinates were later converted to Talairach space (Talairach and Tournoux, 1988). Finally, the volumes were spatially smoothed using an 8-mm isotropic Gaussian kernel. Head motion was assessed prior to preprocessing. No individual moved more than 3 mm in any direction, in any run. Thus, no data were eliminated in either group due to motion artifacts.

Statistical Analyses

Item task

In the blocked item memory task, the onset of each block was modeled as a boxcar function, with duration equal to the length of the block. Encoding, retrieval, and rest blocks associated with each task were modeled separately, and confounding factors, such as head motion and scanner drift, were also included in the model and treated as regressors of no interest.

Relational task

In the event-related relational memory task, trial-related fMRI activity was first modeled by convolving a vector of the onset times of each trial with a canonical hemodynamic response function (HRF) within the context of the general linear model (GLM), as implemented in SPM2. During encoding, subsequently remembered trials were defined as encoding trials that led to a "definitely same" recognition response, whereas subsequently forgotten trials were defined as encoding trials that led to a "changed" recognition response. These two trial types were modeled separately. Encoding success activity was identified by directly comparing subsequently remembered to subsequently forgotten trials. Retrieval trials were similarly defined and modeled, with retrieval success activity identified by directly comparing remembered to forgotten intact face-scene pairing. Thus, analyses focused on successful encoding and retrieval of intact pairs compared to failures to encode/retrieve intact pairs. Lures (and subsequent lures), trials leading to a "probably same" recognition response, as well as confounding factors such as head motion and scanner drift, were also included in the model and treated as regressors of no interest. The current analysis approach, isolating only high-confidence remember trials, allows for the investigation of neural activity associated with recollection-based memories, as opposed to

memories confounded by high levels of familiarity (e.g., Yonelinas, 2001; Diana et al., 2008).

Masking and thresholding

Because a primary goal was to replicate Hariri et al. (2003), we used similar statistical procedures. Based upon 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 an anatomical library available within SPM2 (Tzourio-Mazoyer et al., 2002). For the item memory task, within each group, encoding activity was identified by comparing encoding blocks to resting baseline activity at $P < 0.05$, uncorrected with a minimum cluster size of 10 contiguous voxels within the focal MTL ROI. These results were subsequently used as an inclusive mask for identifying group effects at an uncorrected $P < 0.05$ and a minimum cluster size of 10 contiguous voxels. Thus, activations were required to pass two thresholds: (1) they had to show a significant encoding task effect (encoding vs. rest) within one of the groups ($P < 0.05$ and $k = 10$ voxels); and (2) a group difference in the encoding effect ($P < 0.05$ and $k = 10$ voxels). The conjoint probability following inclusive masking approaches $P = 0.0025$ (Fisher, 1950; Lazar et al., 2002), but this estimate should be taken with caution given that the contrasts were not completely independent. This analysis approach was used to ensure that group differences were associated specifically with those activations exhibiting positive activity for task compared to resting baseline blocks. Similar analyses were conducted for retrieval, as well as the relational memory task, which assessed both encoding success activity and retrieval success activity in the MTL.

To identify activations outside of our a priori ROI that also exhibited between-group differences, whole-brain between group contrasts were conducted at an uncorrected $P < 0.001$ with a minimum cluster size of 20 contiguous voxels. We note that no activation was found that met this criteria.

Voxel-based morphometry (VBM)

Given that several previous studies have identified an effect of the *BDNF* genotype on volumetric differences within the MTL (e.g., Bueller et al., 2006), and differences in volume have been associated with differences in functional activity (e.g., Remy et al., 2005), we assessed differences in MTL gray matter concentration between groups using a VBM analysis (for further description of methods, see Good et al., 2001). High-resolution anatomical images of all subjects were normalized and segmented in SPM2. These images were used to create a gray matter template, which was smoothed with an 8-mm kernel for further processing. Every T1 image was segmented, and resulting gray matter images were normalized (nonlinear cutoff = 25 mm) to the specific gray matter template. Estimated normalization parameters were applied to the original T1 images, which were then segmented a final time. The resulting gray matter images were smoothed with an 8-mm kernel and used for statistical evaluation. Between-group contrasts were conducted on the modulated gray matter volumes within the MTL at $P < 0.001$, uncorrected, with a minimum cluster size of 10 contiguous voxels.

RESULTS

Elimination of Behavioral Confounds

Table 1 reports both the hit and false alarm rate for each group. Paired *t*-tests revealed that, overall, both groups made significantly more hits than false alarms across both memory tasks (item task: *BDNF* met carriers: $t(10) = 18.54$, $P < 0.001$; *BDNF* val/val homozygotes: $t(10) = 34.66$, $P < 0.001$; relational task: *BDNF* met carriers: $t(10) = 7.90$, $P < 0.001$; *BDNF* val/val homozygotes: $t(10) = 13.59$, $P < 0.001$), indicating that both groups were successful in correctly recognizing previously viewed images and face-scene associations. Unpaired *t*-tests on each measure revealed no significant between-group differences in hit rate, false alarm rate, or response bias (d') in either task. Moreover, a breakdown of trial types by confidence also revealed no significant differences between groups. The results indicate that both groups exhibited similar levels of memory performance. Thus the goal of equating cognitive performance across groups based on our selection criteria was met, and any differences in fMRI activity between *BDNF* met carriers and noncarriers cannot be attributed to differences in cognitive performance.

Attempted Replication of Reduced MTL Activation During Item Encoding and Retrieval in Met Carriers

Compared with *BDNF* val homozygotes, met carriers exhibited significantly greater MTL activity associated with both the encoding and retrieval tasks (see Table 2; Fig. 2). Specifically, these effects were seen for encoding in the left anterior parahippocampal gyrus (PHG) and right hippocampus/PHG, and for retrieval in right posterior hippocampus/PHG. No MTL region showed more activity for *BDNF* val homozygotes compared to met carriers, for either encoding or retrieval. In sum, the results of the blocked item memory tasks not only failed to replicate Hariri et al.'s (2003) findings but they actually showed the opposite result, with more MTL activity for met than val cariers.

Examination of *BDNF* val66met Genotypic Differences in MTL Activity Denoting Encoding and Retrieval Success During the Relational Memory Task

Compared with *BDNF val* homozygotes, met carriers exhibited significantly greater MTL activity associated with both encoding and retrieval success defined as the difference between remember and forgotten trials (see Table 2; Fig. 3). Specifically, these effects were seen during encoding in the right hippocampus/PHG and during retrieval in bilateral anterior hippocampus/PHG and right posterior hippocampus/PHG. No MTL region

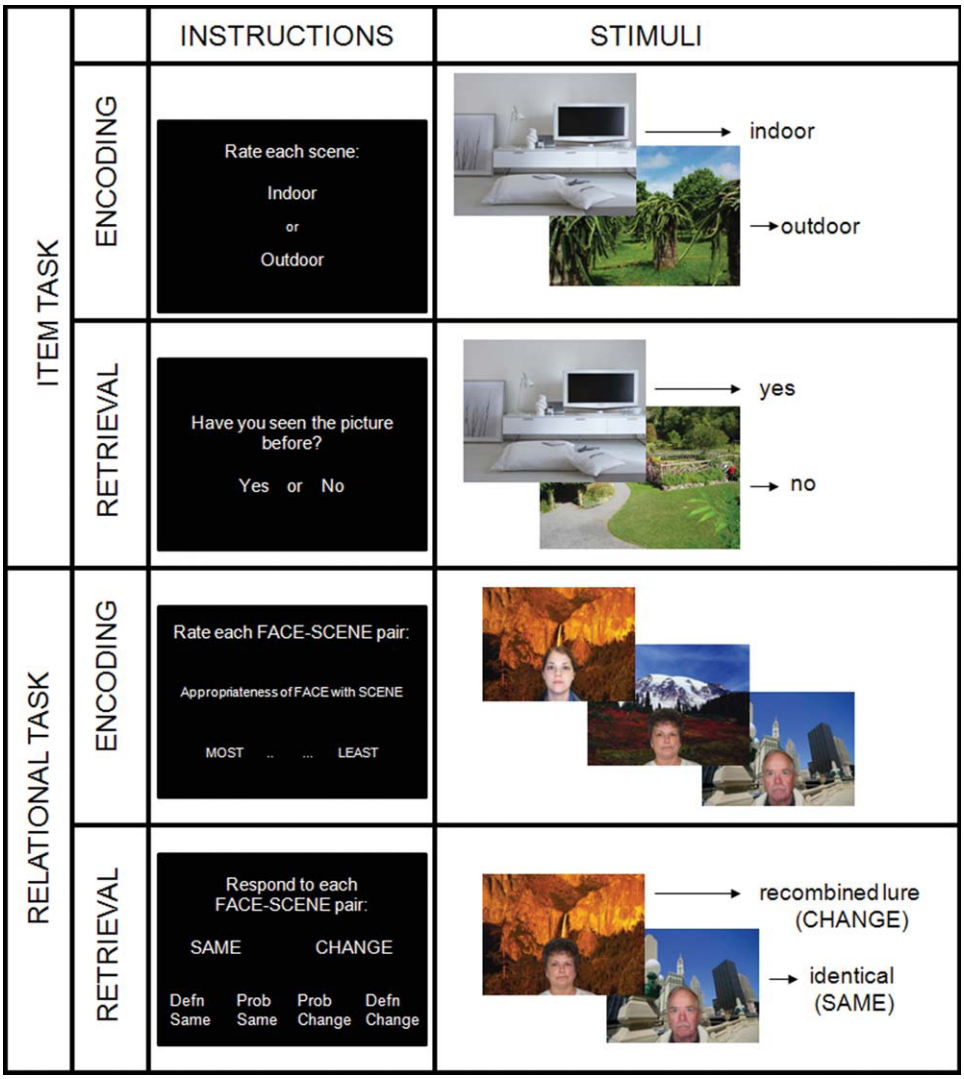

FIGURE 1. Task instructions and example of stimuli used in the item and relational memory tasks (Please note that actual stimuli in item task consisted of neutral pictures taken from the International Affective Picture System dataset.). Item memory task: During encoding participants were asked to determine whether each scene depicted an "indoor" or "outdoor" scene. During retrieval they were asked whether they had viewed that scene previously during encoding, and to respond simply, "yes" or "no." Relational memory task: During encoding participants were asked to rate the appropriateness of the face-scene pair, on a four-point scale of most-to-least appropriate. During retrieval they were asked to determine whether the face-scene pair they were viewing was the same or changed from that which was presented during encoding. They were asked to respond either "definitely same," "probably same," "probably change," or "definitely change." Note: during the Item Memory Task, images consisted of neutral-valence pictures. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

showed more activity for *BDNF* val homozygotes compared with met carriers, for either encoding or retrieval success activity.

VBM Analysis

We note that no regions within the MTL exhibited volumetric differences between groups in the current sample.

DISCUSSION

This study investigated the effects of a naturally occurring functional variant of the *BDNF val66met* genotype on both item and relational memory processes within the MTL. After eliminating differences in cognitive performance and MTL volume, and contrary to previous results by Hariri et al. (2003), we found that *BDNF* met allele carriers exhibited significantly greater MTL activity associated with both encoding and retrieval of complex scenes, compared to noncarriers. Additionally, we showed for the first time that these group differences in MTL activation for item memory extended to successful encoding and retrieval of relational memories.

The main difference between the current item memory study and that of Hariri et al. (2003) is that the present study controlled for potentially confounding influences of group differences in cognitive behavior. In Hariri et al. (2003), met carriers

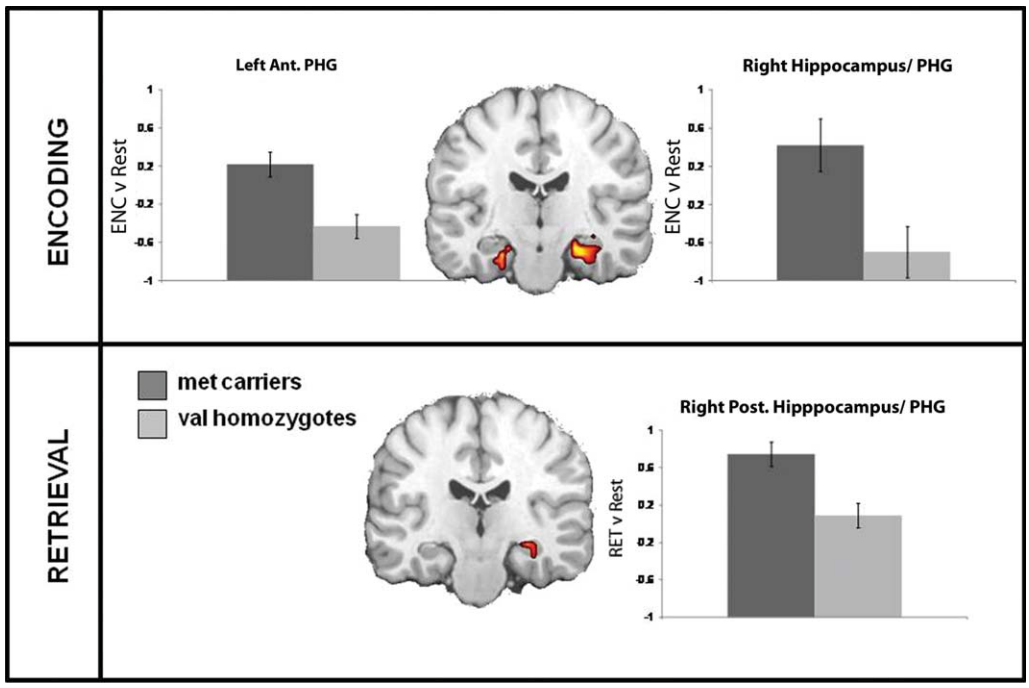

FIGURE 2. Greater item memory task activity in the medial temporal lobes for both encoding (a) and retrieval (b) for *BDNF* met carriers compared to val homozygotes. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

showed both poorer behavioral performance and reduced MTL activation during both encoding and retrieval tasks. Such confound limits the interpretation of neural differences. For example, in the aging literature, it has often been found that by separating older individuals into high and low performers (based on their cognitive behavior in a given task), one observes different patterns of neural activity for the same cognitive process (e.g., Cabeza et al., 2002; Daselaar et al., 2003). Reduced activations in the low, compared with high performers, have been attributed to inability of the low performers to recruit the necessary neural mechanisms for successful task performance. By matching participants on a subset of neuropsychological tests ahead of the fMRI phase, this was also able to match memory performance across both groups in both the item and relational memory tasks performed in the scanner. In the absence of memory differences, met allele carriers, compared with noncarriers, exhibited greater (not reduced) hippocampal activation during both encoding and retrieval of item memory.

The current results are also contrary to a recent study by Hashimoto et al. (2008), which identified encoding-related genotype differences in hippocampal activity in an Asian sample—in the absence of behavioral memory differences. While only partially replicating Hariri et al.'s original findings (2003), Hashimoto et al., point to sample differences as well as methodological differences and differences in allelic heterogeneity as potential mediating factors for the inconsistency in results. Given that the stimuli, study design and analysis were all closely matched between the current study and that of Hariri (2003) and Hashimoto (2008), we must consider possible genetic differences (e.g., Asian vs. Caucasian) and age differences (average age in current sample: ~22 years; average age in Hashimoto et al.: ~37 years) as potential causal factor mediating the inconsistencies in results.

This study also investigated the role of the *BDNF* genotype in memory performance and MTL functioning in a more difficult relational memory task. Like the item memory task, relational memory also engages the hippocampus and PHG, but research has shown that it places a greater demands on hippocampal processing than item memory alone (e.g., Davachi and Wagner, 2002; Sperling et al., 2001; Dennis et al., 2008) to bind item and context into a complex memory trace (Eichenbaum et al., 2007; Johnson et al., 1993). An additional difference between the item and relational memory tasks was related to neural measurement. The current item-memory study [and the previous study by Hariri et al. (2003)] probed MTL involvement during episodic encoding using a blocked design in which activity is summed across all portions of the memory blocks. While a potentially useful design, this procedure confounds such processes as stimulus identification, effort, and, in the case of memory, encoding (or retrieval) attempts as well as successes. The relational task, on the other hand, specifically measured successful memory activations using an event-related design time-locked to stimulus onsets. As noted, event-related designs allow for the direct comparison of neural activity associated with remembered vs. forgotten trials, thus allowing for an assessment of neural processing leading to successful memory while filtering out other aspects of task processing. Despite differences in functional measurement and task design, the results were the same—greater activation

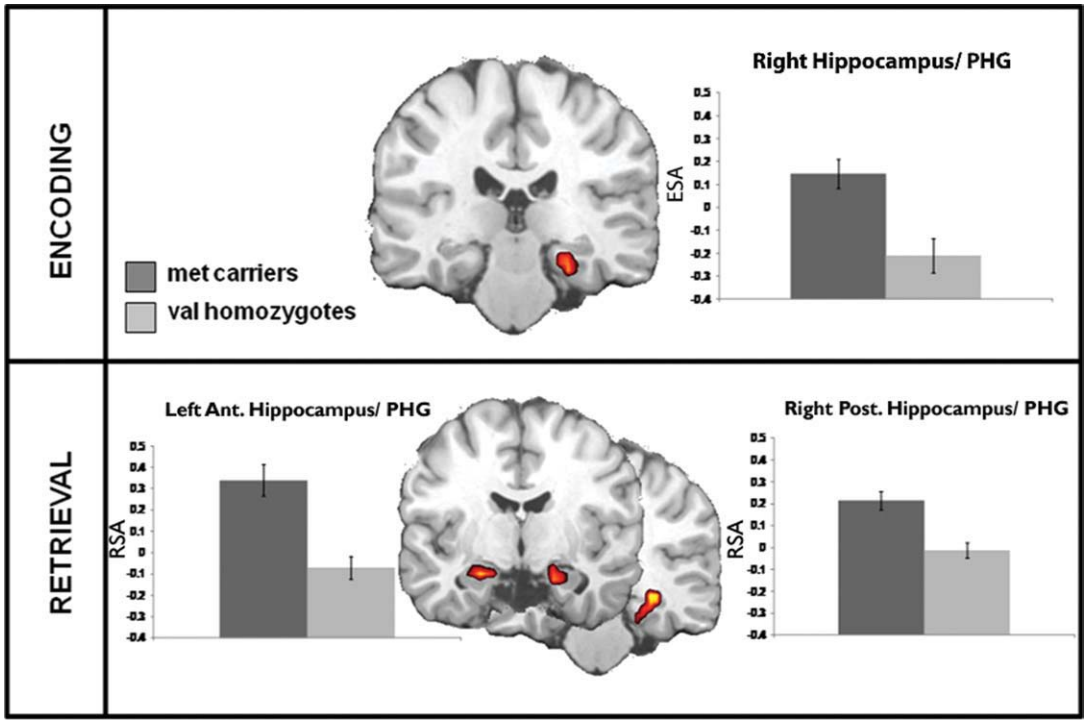

FIGURE 3. Greater medial temporal lobe activity indexing encoding success (a) and retrieval success (b) during the relational memory task, for *BDNF* met carriers compared to val homozygotes. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

of the hippocampus was observed in met carriers during both successful encoding and retrieval of relational memories. Thus, not only do the relational memory findings support the item memory results, but they also suggest that increased engagement of the hippocampus and PHG in met allele carriers occurs across a range of episodic memory tasks. Interestingly, hippocampal differences were observed in both anterior and posterior hippocampus during retrieval. In concert with results from Giovanello et al. (2009), these results suggest differences in neural activity responsible for retrieval of both flexible and exact memory associations.

Importantly, these results complement those of Hariri et al. (2003) in that the current results suggest that previously observed reductions in hippocampal activity in met carriers may be a result of poorer performance, and not an intrinsic inability to perform the task based upon limitations of the MTL structure and function. As noted above, the relationship between external measures of cognition (i.e., memory score) and internal processes of cognition (i.e., neural activations) can be difficult to interpret. Often researchers are left to determine whether group differences in neural activity are a result of differences in cognitive performance or whether differences in cognitive performance arise from differences in neural activations. However, differences in neural activity in the absence of overt behavioral differences, as in the current set of results, may suggest several interpretations. One interpretation is that met allele carriers, to match noncarriers on cognitive performance measures, need to recruit and utilize greater MTL processing to do so. And when they fail to recruit the MTL in such a manner, performance suffers (as found in Hariri et al., 2003). Alternatively, the increased MTL activation in met carriers compared to noncarriers may represent enhanced episodic memory that simply was not measured in the current behavioral paradigm (e.g., memory for greater details or representation of longer lasting memory traces). Finally, the increased MTL activation may represent neural inefficiency, with increases leading to no memory advantage and equitable performance across groups. Increased MTL activity in the absence of enhanced memory performance has been observed in other populations, known for impairments in memory, such as older adults (e.g., Duverne et al., 2008) and patients with Mild Cognitive Impairment (e.g., Dickerson et al., 2004). Like previous studies examining the effects of the *BDNF* val66met genotype on episodic memory-related neural recruitment (Hariri et al., 2003; Hashimoto et al., 2008), the current results require replication, as this is the first time that met carriers have been shown to exhibit increased MTL activity during performance of memory tasks.

CONCLUSIONS

Taken together, converging results from both item and relational memory tasks suggest that the increase in hippocampal and PHG activation for *BDNF* met allele carriers is not simply associated with task-related processing, but rather successful memory performance. The findings were strikingly similar across both encoding and retrieval stages of memory processing. These results indicate that met carriers are able to recruit MTL activity

to support equitable memory performance. Moreover, reductions in cognitive performance are not a ubiquitous effect associated with the *BDNF val66met* genotype of these individuals or group differences in MTL volume. These findings combined with that of previous studies also underscore the variability in results across fMRI studies attempting to measure complex cognitive functioning. Future studies attempting to link genetic variants with cognitive activation patterns using fMRI should take care to examine the contribution of task performance when interpreting results. Further studies should aim to determine whether the increased MTL, and specifically hippocampal, activation is advantageous or compensatory, and to elucidate the functionality of this mechanism across the lifespan and in patient populations who exhibit compromised MTL activity.

Acknowledgments

The authors thank Phil Kragel and Lauren Warren for their help in preparation of this manuscript.

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