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## Altered deactivation in individuals with genetic risk for Alzheimer's disease

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### Abstract

Regions that show task-induced deactivations may be part of a default-mode network related to processes that are more engaged during passive than active task conditions. Alteration of task-induced deactivations with age and dementia is indicated by atypical engagement of default-mode network regions. Genetic studies show a relation between the apolipoprotein E4 (*APOE4*) allele and the common form of Alzheimer's disease (AD), and altered functional brain activation has been observed in non-demented *APOE4* carriers compared to non-carriers. Here we investigate the hypothesis of altered default-mode network brain responses in individuals with genetic risk for AD. Functional MRI was used to assess task-induced deactivation in 60 subjects of which 30 carried at least one copy of the *APOE4* allele, and 30 non-carriers. Subjects were scanned while performing a semantic categorization task shown to promote episodic memory encoding. The results show patterns of deactivation consistent with the default-mode network. We also found reduced deactivation in non-demented *APOE4* carriers compared to non-carriers, suggesting alterations in the default-mode network in the absence of dementia. These results implicate possibilities for investigating altered properties of task-induced deactivations in individuals with genetic risk for AD, and may prove useful for pre-clinical identification of individuals susceptible to memory problems and AD. © 2008 Elsevier Ltd. All rights reserved.

**Keywords:** Deactivation; Aging; *APOE*; Genetic; AD; Alzheimer's disease; fMRI; Compensation; Memory encoding

Task-induced deactivations in neuroimaging studies can be characterized as decreases in the measured brain response during an experimental condition compared to a low-level rest baseline or a control condition. Such deactivations (task < baseline) may reflect active processes engaged during the resting state. Deactivations have consistently been found in a set of brain regions including the medial frontal, medial and lateral parietal, and posterior cingulate cortex (e.g. Binder et al., 1999; Mazoyer et al., 2001). The consistency of deactivation in these regions across tasks suggests that they are independent of task characteristics and study material. One hypothesis is that regions that show deactivations are part of a "default-mode network" related to processes that are more engaged during passive- than active-task

conditions (Raichle et al., 2001). According to the default-mode hypothesis, passive baseline is a state of structured processes that are interrupted when individuals engage in experimental tasks, resulting in relative deactivation when experimental conditions are compared with baseline conditions.

Recently, several studies have focused on to what extent task-related deactivation differs between young adults, healthy older adults, and patients with Alzheimer's disease (AD) (Grady, Springer, Hongwanishkul, McIntosh, & Winocur, 2006; Lustig et al., 2003; Persson, Lustig, Nelson, & Reuter-Lorenz, 2007; Rombouts, Barkhof, Goekoop, Stam, & Scheltens, 2005). For example, functional deactivation patterns in the medial PFC and PCC differ between patients with AD, healthy older adults, and young adults using a semantic classification task (Lustig et al., 2003). Lustig et al. (2003) found that deactivation in the medial PFC was reduced for both patients with AD and healthy older adults compared to young adults. Another intriguing finding was

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found in the PCC, in which young adults showed deactivation, while older adults without dementia showed a marginal increase in activation. Individuals with AD had positive activations that were greater than for older subjects without dementia. This suggests age-related changes in deactivation, and that these changes get more severe with the progression of dementia.

Genetic studies have identified a relation between the apolipoprotein E-4 (*APOE4*) allele and the common form of AD (Strittmatter et al., 1993). Measures of resting state glucose metabolism using FDG-PET have found altered patterns of brain activity in *APOE4* carriers compared to non-carriers (Reiman et al., 1996; Small et al., 2000). Typically, the characteristic changes observed in AD (reduced parietal, temporal, and posterior cingulate metabolism) appear in a less pronounced form in carriers of non-demented *APOE4* carriers. More recently, neuroimaging studies have investigated task-related brain activation patterns in non-demented *APOE4* carriers (Bookheimer et al., 2000; Lind et al., 2006b; Smith et al., 1999). A common finding is that increased risk for AD is associated with reduced activation in temporal, parietal and posterior cingulate regions (e.g. Lind et al., 2006b; Smith et al., 1999). In relation to the default-mode network, it is important to note that many of the regions that show reduced resting state metabolism in *APOE4* carriers and patients with AD are regions that show deactivation in young adults.

One possibility is that altered PCC activity in AD reflects disrupted connectivity with medial temporal lobe (MTL) structures that are the earliest and most affected sites for AD pathology (Braak & Braak, 1994). Also, human and animal lesion studies show that damage to MTL regions result in reduced PCC resting metabolism similar to what is observed in AD (Aupee et al., 2001; Meguro et al., 1999; Reed et al., 1999). The analysis of deactivations may be critical to the understanding of the neural dynamics and network activity that underlie efficient and optimal brain function. This may prove to be especially important for characterizing global alterations in neural functioning that accompany normal and abnormal aging.

In the present study, we investigate the hypothesis that the default-mode network is abnormal in subjects with increased genetic risk for AD. We used fMRI to assess task-induced deactivations in 60 subjects of which 30 carried at least one copy of the *APOE ε4* allele, and 30 non-carriers. Subjects were scanned while performing a semantic (abstract/concrete) categorization

task shown to promote episodic memory encoding (e.g. Kapur et al., 1994). Of main concern was whether task-induced deactivation in frontal and parietal regions would differ as a function of genetic risk. Here we investigated differences in deactivation between carriers and non-carriers of the *APOE4*, as well as dose-related differences between carriers of either one or two alleles (*APOE44* and *APOE34*), and non-carriers (*APOE33*), respectively.

1. Methods

1.1. Participants

Sixty cognitively intact persons between the ages 49 and 79 years participated in the present study. They were all recruited from *The Betula prospective cohort study: Memory, health, and aging* (Nilsson et al., 1997, 2004). Thirty subjects were carriers of at least one copy of the *APOE4*: 10 were homozygous (44) and 20 were heterozygous (34). The remaining thirty subjects carried two copies of *APOE3* and were considered as controls. The same participants were included in prior studies, and the results have been reported elsewhere (Lind et al., 2006a, 2006b, 2006c; Persson et al., 2006a, 2006b). To examine a possible dose-effect, three subgroups consisting of 10 subjects each were composed: *APOE44*, *APOE34* and *APOE33*. Participants were closely matched according to sex, age and years of education (see Table 1 for group characteristics). All subjects were non-demented and scored at or above the standard cut-off point of 25 on the mini-mental state examination (MMSE) (Folstein, Folstein, & McHugh, 1975). They were all right-handed, native Swedish speakers, and had no reported neurological problems that might cause dementia. Vision was normal or corrected to near normal using scanner compatible glasses or contact lenses. Subjects were paid for participation and informed consent was obtained in accordance with the guidelines of the Swedish Council for Research in the Humanities and Social Sciences.

Approximately 2 years after the reported MRI testing, 55 of the original 60 subjects were re-tested on a wide range of cognitive tasks as a part of the longitudinal Betula project and they still showed no signs of dementia. In addition, we compared the *APOE4* carriers' explicit memory performance (based on three tests—face recognition, verbal recall, and recall of actions, for detailed description of the tests, see Nilsson et al., 1997) with normative data available from the Betula database. Twenty-eight of the 30 *APOE4* carriers performed within 1 S.D. of the mean of their age group; two subjects scored below 1 S.D., but performed within 1 S.D. at the follow-up test (see above) 2 years after MRI testing. Together, these results provide evidence that all participants were cognitively intact.

1.2. *APOE* genotyping

A PCR was performed using 200 ng of genomic DNA as template in a 25-ml reaction mixture containing 20 pmol of PCR primers *APOE-A* (5'-TCC-AAG-GAG-CTG-CAG-GCG-GCG-CA-3') and *APOE-B* (5'-ACA-

Table 1  
 Group characteristics

	<i>APOE ε4</i> (n = 30)	<i>APOE ε3/3</i> (n = 30)	<i>APOE ε4/4</i> (n = 10)	<i>APOE ε3/4</i> (n = 10)	<i>APOE ε3/3</i> (n = 10)
Female/male	19/11	18/12	9/1	7/3	8/2
Age	65.6 (7.9)	66.6 (8.9)	63.1 (8.6)	65.6 (8.2)	64 (11.1)
Range	49–74	50–79	49–74	51–74	50–79
Education (years)	10.8 (3.6)	10.2 (3.2)	11.7 (3.1)	10.7 (4.0)	11.8 (3.1)
Range	6–17	6–16	8–16	6–17	9–16
MMSE	28.6 (1.3)	28.2 (1.3)	28.6 (1.2)	28.8 (1.2)	28 (1.2)
Range	25–30	26–30	27–30	26–30	26–29
SRB	24 (3.2)	21.6 (4.4)	22.8 (3.1)	24.7 (3.6)	22.2 (4.1)
Range	16–29	11–29	16–26	17–28	18–28

Note: Means and standard deviations (in parenthesis). MMSE = mini mental state examination (maximum = 30). SRB = word comprehension (maximum = 30).

GAA-TTC-GCC-CCG-GCC-TGG-TAC-ACT-GCC-A-3'), 0.2 U of Taq DNA polymerase (GibcoBRL, Gaithersburg, MD), 1.0 mM MgCl<sub>2</sub>, 75 mM Tris-HCl (pH 9.0), 20 mM (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, and 10% dimethyl sulfoxide. The PCR amplification consisted of 35 cycles of 30 s at 94 °C, 30 s at 65 °C, and 30 s at 72 °C. PCR products were digested using 5 U of *Hha*I (Life Technologies Inc., Rockville, MD) by incubating for 3 h at 37 °C. Bands were separated on a 5% agarose gel and visualized on an ultraviolet transilluminator after ethidium bromide staining. Alternatively, electrophoresis was performed using ExcellGel gels (Pharmacia, Piscataway, NJ) and the MultiphorII electrophoresis system (Pharmacia), and the bands were visualized by silver staining.

### 1.3. Procedure

Functional MRI was used to assess brain responses while participants performed an abstract/concrete semantic categorization task that promoted incidental encoding of a word list, containing in all 160 words. During fMRI, a blocked-task paradigm was used, alternating between the categorization task (30 s) and fixation (20 s). During fixation, subjects had been instructed to rest while watching a hair-cross that was constantly displayed on the screen. Each run began and ended with fixation scans (12 s). Four runs were used and they consisted of four categorization blocks containing 10 words each. In all, 160 words were presented during the fMRI session. Subject-responses were given by pressing one of two buttons, using the right index and middle fingers.

Subjects' behavioral performance was recorded for response reaction times and categorization accuracy. In addition, memory performance was tested after the scanning session using a self-paced yes/no recognition test in which participants indicated whether they saw a new or a previously studied word. Subjects made recognition decisions on 240 words: 160 previously presented words and 80 lures (not studied before), presented in a mixed order. Two subjects did not complete this session; hence the recognition results are based on 58 subjects only.

During all sessions, the same words were presented in the same order to all subjects.

### 1.4. fMRI data acquisition

Images were collected using a 1.5 T Philips Intera scanner (Philips Medical Systems, Netherlands) equipped for echo-planar imaging (EPI). A T<sub>2</sub>\*-weighted single-shot gradient echo EPI sequence was used to acquire blood oxygen level dependent (BOLD) contrast images. The following parameters were used: repetition time: 3000 ms (33 slices acquired), echo time: 50 ms, flip angle: 90°, field of view: 22 cm × 22 cm, 64 × 64 matrix and 3.9 mm slice thickness. To avoid signals arising from progressive saturation, five dummy scans were performed prior to the image acquisition. In the scanner, cushions and headphones were used to reduce movement, dampen scanner noise and communicate with the participant. Stimuli were displayed on a projection screen at the head of the bore, viewed by the subjects from within the magnet via a tilted mirror placed on the head coil.

Words were presented on the screen at the frequency of one every 3 s (ISI = 1 s), centered in lower case letters in white 60-point Courier New font on black background. Word presentation and registration of reaction time data was handled by a PC running E-Prime 1.0 (Psychology Software Tools). Responses were collected with a fiber-optic response box held in the right hand (Lumitouch reply system, Lightwave Medical Industries, Canada). High-resolution T<sub>1</sub>- and T<sub>2</sub>-weighted structural images were also acquired. The total time in the MR scanner was approximately 75 min per subject. For further details see Lind et al. (2006b).

### 1.5. fMRI data analysis

All images were sent to a PC and converted to analyse format. Functional images were pre-processed and analysed using SPM99 (Wellcome Department of Cognitive Neurology, UK, <http://www.fil.ion.ucl.ac.uk>) implemented in Matlab 6.1 (Mathworks Inc., MA, USA). Prior to analysis, all images were realigned to the first image volume acquired, then normalized to standard anatomic space defined by the MNI atlas (SPM99), and finally spatially

smoothed using a 6.0-mm full-width at half-maximum Gaussian filter kernel. The semantic categorization task was modelled as a fixed response (box-car) waveform convolved with the hemodynamic response function. Single-subject statistical contrasts were set up using the general linear model, and group data were analysed in a random-effects model. Statistical parametric maps (SPMs) were generated using *t* statistics to identify regions activated according to the model.

Two whole-brain analyses were carried out. As a first step, we used the contrast between baseline and semantic categorization (baseline–semantic categorization) across all participants in order to identify brain regions associated with the default-mode network. This procedure for investigating deactivations have been used in numerous previous studies (e.g. Lustig et al., 2003; Mazoyer et al., 2001; McKiernan, Kaufman, Kucera-Thompson, & Binder, 2003; Persson et al., 2007; Shulman et al., 1997), and generally results in robust involvement of default-mode regions. All reported across-subject whole-brain deactivations passed a threshold of  $P < 0.05$  corrected for multiple comparisons. To confirm that we were not biasing our results by only testing our hypothesis in voxels that showed robust deactivations across participants, we performed a second analysis at the group level using a 2 (group [*APOE*33 vs. *APOE*4]) × condition (baseline vs. semantic categorization) ANOVA to assess group-related differences in deactivation. All reported deactivations in the whole-brain group comparisons (*APOE*33 vs. *APOE*4) passed an uncorrected threshold of  $P < 0.005$ .

For the region-of-interest (ROI) analyses, we selected peak coordinates that have been associated with deactivation across different tasks in several previous neuroimaging studies, and which showed less activation (i.e. deactivation) in the encoding condition compared to the rest baseline for all participants (e.g. Binder et al., 1999; Mazoyer et al., 2001). Effect sizes (% signal change) for semantic categorization for each of the ROIs were then extracted for each of the subjects using the SPM ROI toolbox, and used for separate between-group ANOVAs for each of the ROIs. In order to assess whether deactivation in the default-mode network has implications for behavioral performance, we correlated deactivation magnitudes with post-scan memory performance (hits–false alarms) and reaction times across all participants. Thus, the fMRI analyses included (a) whole-brain analyses for all participants in order to identify the default-mode network and defining ROIs for subsequent analyses, (b) ROI analyses of between-groups differences in deactivation magnitude, (c) whole-brain analysis of group-differences in deactivation, and (d) analyses of performance–deactivation correlations and correlations between magnitude estimates in default-mode and prefrontal regions.

## 2. Results

### 2.1. Behavioral data

The behavioral results have been described elsewhere (Lind et al., 2006b). In brief, both groups were accurate in classifying words as abstract or concrete (*APOE*33 carriers: 94.6%; *APOE*4 carriers: 97.2%). There were no significant between-group differences in classification accuracy or overall RT. The post-scan recognition data (hits–false alarms) revealed no significant difference between *APOE*33 carriers (62.5%) and *APOE*4 carriers (61.5%).

### 2.2. Whole-brain analyses of deactivation across all participants

To investigate deactivation related to memory encoding, we contrasted the rest baseline with the semantic classification task. The results from this contrast are presented in Fig. 1 and Table 2. Consistent with previous findings (e.g. Mazoyer et al., 2001; Raichle et al., 2001) task-induced deactivation was found

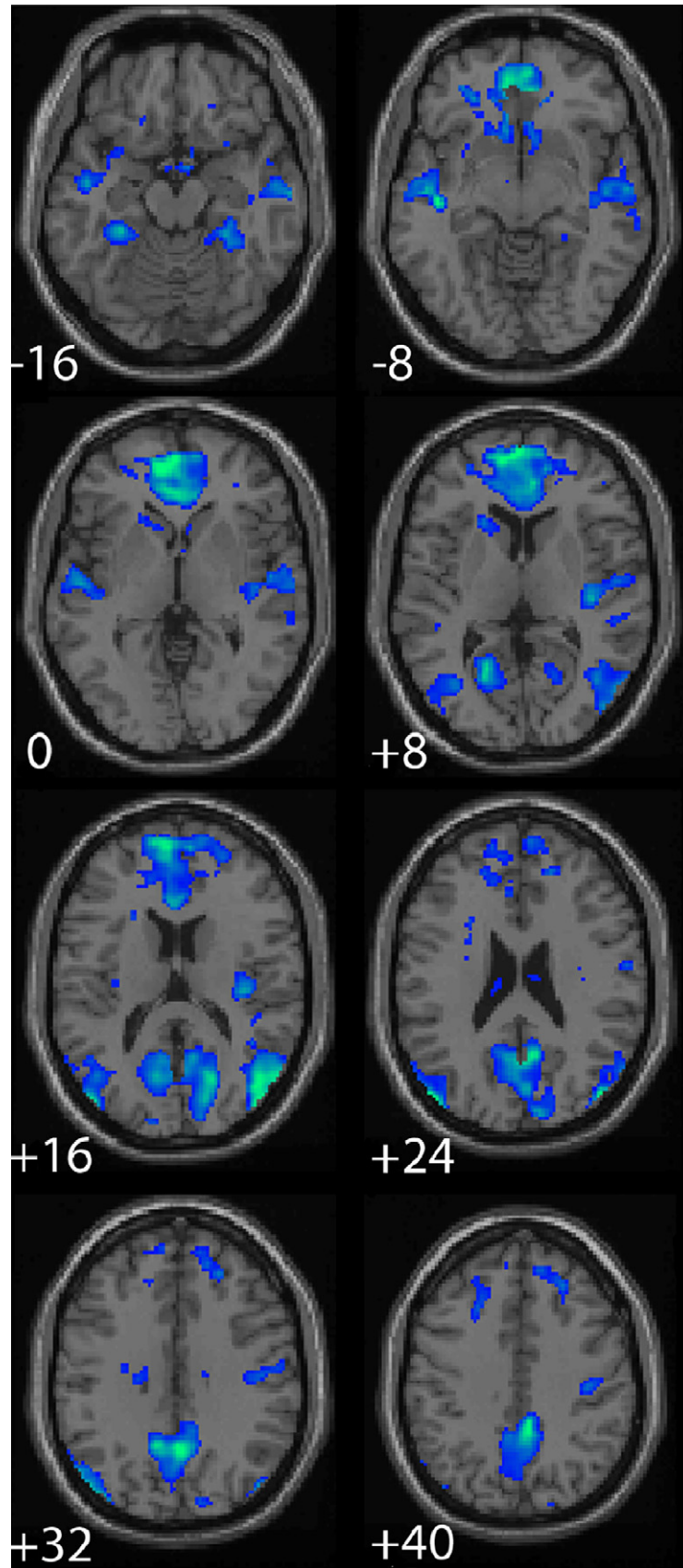


Fig. 1. Transverse sections show significant difference in rest baseline compared to semantic categorization (i.e. deactivation) at a corrected threshold ( $P < 0.05$ ). The anatomical template is used as the backdrop.

Table 2

Talairach coordinates for areas that show maximal deactivation ( $P < 0.05$  corrected for multiple comparisons)

Anatomical localization	BA	x	y	z	T
<b>L medial frontal</b>	<b>23</b>	<b>-4</b>	<b>58</b>	<b>12</b>	<b>11.18</b>
<b>L middle temporal</b>	<b>21</b>	<b>-44</b>	<b>-20</b>	<b>-6</b>	<b>11.05</b>
<b>R posterior cingulate</b>	<b>31</b>	<b>2</b>	<b>-56</b>	<b>30</b>	<b>10.62</b>
<b>L ventral posterior cingulate</b>	<b>23</b>	<b>-18</b>	<b>-60</b>	<b>10</b>	<b>10.40</b>
<b>L lateral parietal</b>	<b>39</b>	<b>-46</b>	<b>-80</b>	<b>24</b>	<b>10.38</b>
<b>R lateral parietal</b>	<b>39</b>	<b>54</b>	<b>-68</b>	<b>14</b>	<b>10.26</b>
R middle/medial temporal	42	-18	-4	8.76	
R fusiform gyrus	20	28	-36	-18	8.05
R superior frontal gyrus	6	44	-18	36	7.99
L precuneus	7	-10	-62	56	6.05

L, left; R, right; BA, Brodmann's area; x, y, z, stereotactic coordinates. The regions in bold were selected for ROI analyses.

in several cortical regions including medial prefrontal cortex [Brodmann area (BA) 10], medial parietal cortex (posterior cingulate cortex/precuneus (PCC), BA 31; inferior PCC, BA 23/31), bilateral parietal cortex (BA 39), and the left middle/medial temporal gyrus (BA 21).

### 2.3. ROI analyses—between-group differences

The main objective for the ROI analyses was to examine *APOE* genotype-related differences in the magnitude of deactivation in regions related to the default-mode network. Based on the whole-brain analysis described previously (Fig. 1), six regions associated with the canonical default-mode brain network were selected for additional ROI analyses. The ROIs were defined from the clusters generated by the whole-brain analysis (baseline–semantic categorization) functional data and mean voxel values (% signal change) were extracted using Marsbar (<http://marsbar.sourceforge.net>). These regions have typically been associated with task-induced deactivations (Binder et al., 1999; Mazoyer et al., 2001; Shulman et al., 1997). For all subsequent ROI analyses, we focused on these six regions (Fig. 2, Table 2 in bold). First, using separate ANOVAs we tested for dose-dependent effects between *APOE44* carriers, *APOE34*, and *APOE33* carriers. These analyses yielded no significant results for any of the ROIs, suggesting that the differences in deactivation between *APOE44* carriers and *APOE34* carriers were not reliable. Second, separate ANOVAs (*APOE4* carriers vs. non-carriers) were performed for each of the ROIs. Below we report the findings from the analyses of deactivation difference between *APOE4* carriers vs. non-carriers.

Four of the six regions that were selected for the ROI analyses showed significantly less deactivation for *APOE4* carriers vs. non-carriers (medial PFC:  $F_{1,56} = 4.29$ ;  $P < .05$ , Fig. 2A; left middle temporal cortex:  $F_{1,56} = 9.48$ ;  $P < .005$ , Fig. 2B; medial parietal region:  $F_{1,56} = 4.24$ ;  $P < .05$ , Fig. 2C; right lateral parietal cortex:  $F_{1,56} = 4.38$ ;  $P < .05$ , Fig. 2F). These results suggest that genetic susceptibility for AD may affect the magnitude of deactivation in the default-mode-network. For the remaining

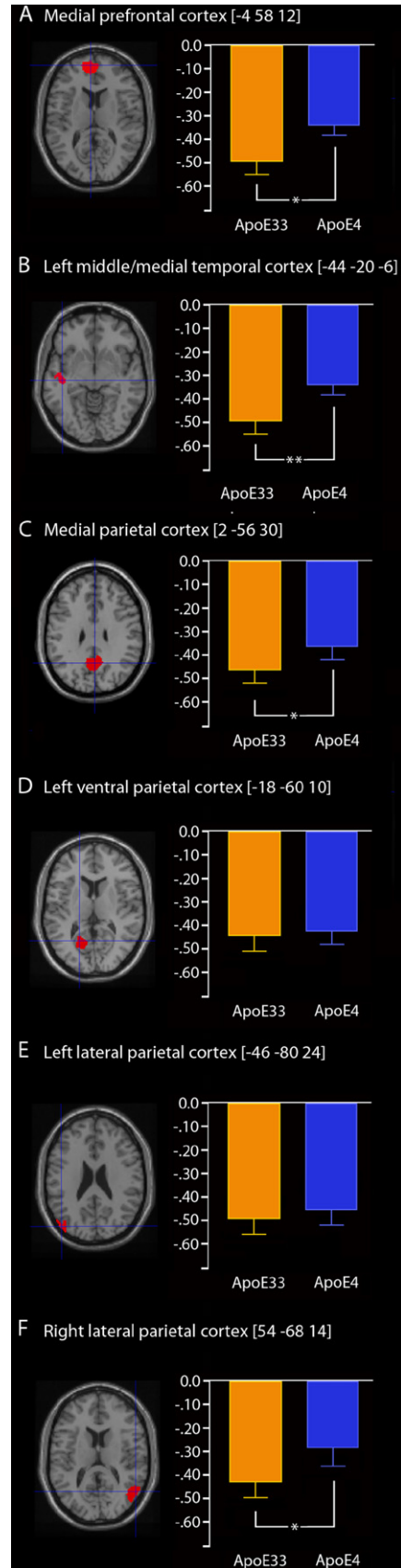


Fig. 2. Transverse sections show the location of the six ROIs used for the analysis of deactivations. Bars show average percent signal change for *APOE33* and *APOE4* participants, respectively. Error bars show standard error of the mean.

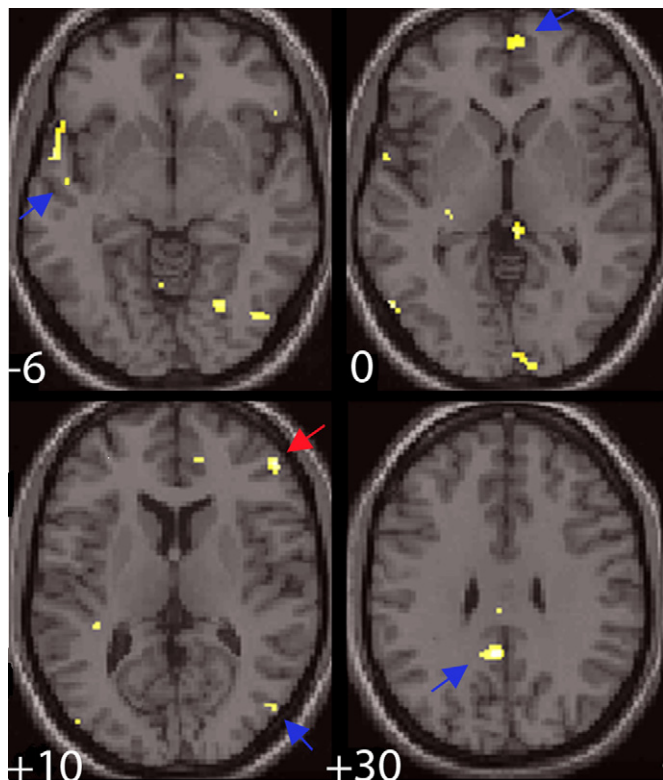


Fig. 3. Map of statistically significant group differences across the brain (baseline > semantic categorization). Note that exploratory maps based on ANOVA do not indicate the direction of the effects.

two regions, no differences were found between *APOE33* and *APOE4* carriers (Fig. 2D and E).

#### 2.4. Confirmatory group analyses

Although ROI analyses permit the stringency of assessing a priori hypotheses and increased sensitivity, they run the risk of omitting effects elsewhere in the brain. We therefore used exploratory whole-brain analyses to confirm and extend the a priori ROI analyses. A 2 (group [*APOE33* vs. *APOE4*]) × condition (baseline vs. semantic categorization) ANOVA was used to assess group-related differences in deactivation. This contrast revealed significant group-related differences in the medial PFC, medial PCC, the left middle/medial temporal cortex, and right lateral parietal cortex (Fig. 3). Although these differences were found using a liberal threshold they were consistent with the findings from the ROI analyses. A significant difference was also found in right inferior prefrontal cortex (Fig. 4). Additional findings included small differences in premotor and visual regions, and the lateral temporal cortex which will not be discussed further. Note, however, that exploratory maps based on ANOVA do not indicate the direction of the effects. Therefore, we examined the group difference in the right inferior prefrontal region by extracting the magnitude estimates for this particular region (Fig. 4). This analysis revealed that *APOE33* carriers showed *deactivation* in this region, while *APOE4* carriers showed *activation* in this region.

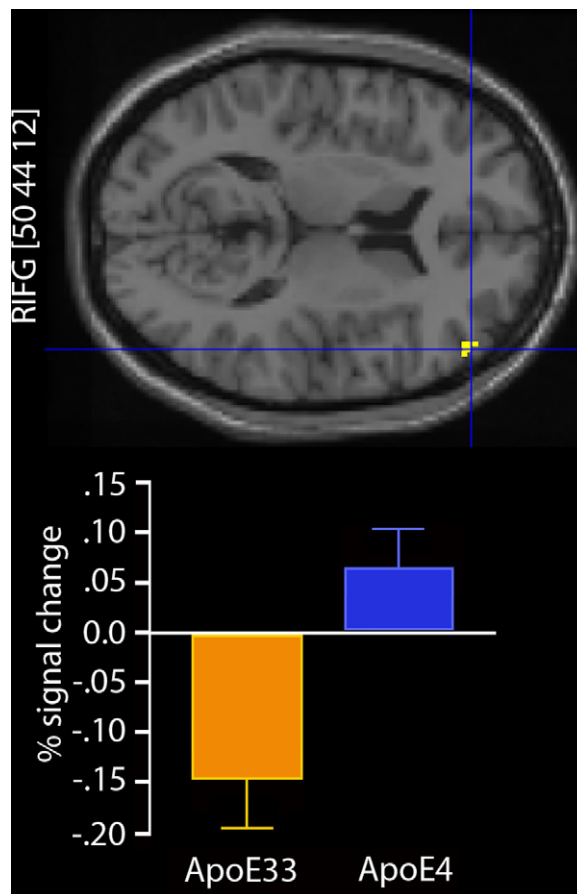


Fig. 4. Transverse section depicts the location of the right frontal region. Bars show average percent signal change for the *APOE33* and *APOE4* groups, respectively. Error bars show standard error of the mean.

#### 2.5. Correlation analyses of fMRI and behavioral data

##### 2.5.1. Performance–deactivation correlations

Do individual differences in performance correspond to individual differences in deactivation magnitude? In order to assess whether deactivation in the default-mode network has implications for behavioral performance, we correlated the magnitude of deactivation in the six ROIs previously described (Fig. 2) with post-scan memory performance (hits–false alarms) and reaction times across all participants. The results from these analyses did not reveal any significant correlations, suggesting weak relationship between deactivation and behavioral performance.

Also, in order to integrate the current results with previous findings we decided to analyse the deactivation data with respect to whether altered deactivation may predict negative outcome among at-risk individuals. Eighteen *APOE4* carriers were divided into two groups on the basis of their longitudinal memory performance (decline vs. stable), measured at two test occasions approximately 5 years apart (see Lind et al., 2006a for further details). Between-group comparisons for each of the ROIs previously described revealed that one region, the medial PFC, showed less deactivation in individuals with declining memory performance (decline:  $-0.32$ , stable:  $-0.45$ ;  $t(16) = 2.35$ ,  $P = 0.032$ ). This suggests a relationship between altered deacti-

vation in medial PFC and future cognitive decline in individuals at risk for Alzheimer's disease.

### 2.5.2. Inter-regional correlations of fMRI data

Given the finding of group-differences in right inferior frontal gyrus (Fig. 4), we investigated the relationship between brain responses in this particular region and default-mode deactivation. This analysis was carried out by correlating the deactivation magnitudes (% signal change) obtained from the ROI analyses with the magnitude estimates (% signal change) in the right inferior frontal gyrus. The results from these analyses showed that two regions, the medial PFC ( $r = .44, P < .005$ ) and the left middle/medial temporal cortex ( $r = .34, P < .05$ ), correlated positively with the right inferior PFC. Thus, less deactivation was associated with more activation in right inferior PFC. When these correlations were investigated for each *APOE* group separately, we found that these correlations were only significant for the *APOE4* group. This suggests more pronounced default-mode–prefrontal interactions for *APOE4* carriers compared to non-carriers.

We also performed additional analyses of correlations between default-mode regions and regions that were previously reported as showing strong activations (Lind et al., 2006b; Persson et al., 2006a). These regions included left parietal cortex, left and right DLPFC, and left and right VLPFC. We did not find evidence for correlations between activation in any of these regions and deactivation in default-mode regions.

## 3. Discussion

The present data reveal several important findings. First, across group contrasts show patterns of deactivation that are consistent with the canonical default-mode network. Additionally, reduced deactivation was observed in non-demented *APOE4* carriers compared to non-carriers. To our knowledge, this is the first study that link changes in deactivation to genetic risk for developing AD, and suggests alterations in deactivation in the absence of dementia. Furthermore, there was no difference in deactivation between *APOE44* and *APOE34* carriers, suggesting a lack of dose-dependent effects. Also, the magnitude of brain responses in a right inferior PFC region was higher for *APOE4* carriers compared to non-carriers, and correlations with this region and default-mode deactivation suggest default-mode network–prefrontal interactions.

Our finding of altered default-mode network activation is consistent with previous observations showing reduced deactivation in patients with AD and mild cognitive impairment (Lustig et al., 2003; Rombouts et al., 2005; see also Celone et al., 2006 for recent findings), as well as in studies of healthy older adults (Grady et al., 2006; Lustig et al., 2003; Persson et al., 2007), and extends their work to show that changes in default-mode activity can occur in healthy individuals at risk for AD. Our findings may also relate to observations that regions associated with the default-mode network show early perfusion and metabolic abnormalities. This is especially true for the posterior cingulate cortex. For example, in very early stages of AD, even before a clinical diagnosis, reduced regional cerebral blood flow as

well as glucose metabolism in the posterior cingulate gyrus and precuneus has been reported using PET (Minoshima, Foster, & Kuhl, 1994; Minoshima et al., 1997) and SPECT (Johnson et al., 1998; Kogure et al., 2000). Reduced blood flow or metabolism has also been reported in pre-symptomatic non-demented individuals with at least a single *APOE4* allele.

Disrupted hippocampal functionality has been suggested as a potential mechanism underlying PCC hypometabolism/hypoperfusion (Matsuda, 2001), and changes in deactivation in early AD and mild cognitive impairment (Celone et al., 2006). Although we did not find support for a direct relationship between hippocampal atrophy or white-matter integrity and deactivation magnitudes (data not shown), previous reports using the same subjects indicate both altered white-matter disruption in a left MTL region (Persson et al., 2006b), and reduced right hippocampus volume (Lind et al., 2006c) in *APOE4* carriers compared to non-carriers. A recent PET study also indicated task-specific alterations in PCC–MTL interactions in older adults compared to young adults (Della-Maggiore et al., 2003). Additional evidence for this hypothesis comes from a recent study that reported a marked reciprocal relationship between the degree of activation within the hippocampus and deactivation in medial and lateral parietal regions during an episodic memory task (Celone et al., 2006). Indeed, previous observations in this sample showed substantial differences in hippocampal engagement between *APOE4* carriers and non-carriers (Lind et al., 2006b). Taken together, these patterns suggest that pre-clinical disruption of hippocampal function might be related to a reduced deactivation in individuals with genetic risk for AD.

In addition to a possible link between structural changes, resting state metabolism, and deactivation, recent investigations on default-mode brain responses in young and older adults have proposed a more process-oriented approach to age-differences in deactivation (Grady et al., 2006; Persson et al., 2007), which may also apply to the present results. In one study (Persson et al., 2007), young and older participants' brain responses at rest were compared to a read condition, and a semantic retrieval task with increasing demands on cognitive control. In line with previous findings (McKiernan et al., 2003), it was found that increasing task difficulty was associated with increased deactivation in the default-mode network. Of main importance, however, was the finding that the difference in deactivation between young and older adults was greatest in the condition with highest task demands, smaller in the low-task demand condition, and non-significant in the read-only condition. The authors suggest that reduced deactivation for older adults in cognitive tasks may indicate a reduced cognitive efficiency stemming from difficulties in disengaging from or inhibiting internal processes in order to reallocate resources to the experimental task. One possibility is that the present findings of reduced deactivation in *APOE4* carriers compared to controls reflect that individuals with increased genetic risk for AD have a reduced ability to suspend activation related to default-mode processes during the active task.

At first glance, the absence of a difference in performance between *APOE4* carriers and non-carriers might argue against this conclusion. Recent findings of correlations between deactivation and performance provide evidence for a functional role

of default-mode regions in successful task execution (Persson et al., 2007). The relationship between deactivation and performance indicates that at a specific level of task difficulty, greater deactivation may be related to more efficient task performance. Previous studies have suggested that spared performance could result from older adults recruiting additional (prefrontal) regions to compensate for changes that occur with aging (e.g. Cabeza, Anderson, Locantore, & McIntosh, 2002; Grady et al., 1994; Reuter-Lorenz et al., 2000). Indeed, the finding that *APOE4* non-carriers showed right PFC deactivation while *APOE4* carriers showed right PFC activation may reflect such compensation. This finding is supported by two recent papers showing increased activation in right PFC regions in *APOE4* carriers and individuals with familial risk for AD, compared to controls during verbal encoding and verbal paired associate learning (Bassett et al., 2006; Han et al., 2007). This idea is further supported by our findings of correlations between deactivation in medial PFC and left middle temporal cortex indicating interactions between PFC and default-mode network regions.

Our previous finding of a reduced task-related response in left parietal cortex for *APOE4* carriers compared to controls in the same sample of participants (Lind et al., 2006b) might first seem to contradict the current finding of reduced right lateral parietal deactivation in the current study. It should be noted, however, that the parietal region previously reported is located more anterior and dorsal compared to the parietal region reported here, and do not overlap with regions showing deactivations. This suggests that the previously reported parietal region is not a part of the default-mode network. Indeed, these two regions might constitute parts of two different networks involved in entirely diverse cognitive processes. Thus, previous findings of less activation in *APOE4* carriers compared to non-carriers in parietal, anterior cingulate and hippocampal regions in the same participants, together with the current findings of reduced deactivation may constitute a brain pattern preceding behavioral indications of dementia in individuals with genetic risk for AD.

Measures of functional change in aging can be complicated by the presence of cerebral atrophy. Although methods for standardization are applied in fMRI to correct for individual differences in brain volume and morphology, such methods may not adequately compensate for local tissue loss and thus confound measurements of cortical function in these regions. Previous quantitative analyses, however, show minimal relationship between BOLD fMRI signal and atrophy in normal aging (Johnson et al., 2000). Likewise, a recent study showed significant differences in spontaneous resting state fluctuations between AD patients and controls after correcting for cerebral atrophy (He et al., 2007). Although these previous observations do not rule out the possibility that local gray matter concentration relate to deactivation in the present study, it speaks against the possibility that atrophy is responsible in a direct way for the fMRI findings.

Taken together, these findings indicate altered deactivation in the brain default-mode network in individuals with genetic risk for AD. The reduced deactivation observed in *APOE4* carriers compared to non-carriers, may be related to reduced resting state

metabolism, structural changes, or both. It may also indicate that *APOE4* carriers have a reduced ability to suspend activation related to default-mode processes during the active task. The present results extend the literature on *APOE* genotypic differences among non-demented adults, and present a promising approach for early identification of pre-clinical AD. Further research using longitudinal follow-up measurements may be needed to clarify the relationship between genetic risk for AD, cognitive performance, and changes in functional brain deactivation.

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