# Age-Related Differences in the Medial Temporal Lobe Responses to Emotional Faces as Revealed by fMRI

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**ABSTRACT: Age-related differences involved in the neural substrates of emotional face perception were investigated in young and old healthy volunteers. The subjects were scanned using functional magnetic resonance imaging while they were judging the gender of faces with negative, positive, or neutral emotional valence. The results showed that both the predominant activation in young subjects and reduced activity in old subjects contributed to a significant age difference in the left amygdala during the perception of negative faces. Activity in the right parahippocampal gyrus during the perception of positive faces diminished with advancing age. Neural activity in the angular gyrus and lingual gyrus of the right hemisphere was reduced in the old subjects during the perception of positive faces. There was no region where old subjects had greater activity than young subjects during the task. In old subjects, the overall activity in the right hippocampus during the task correlated negatively with age, whereas the activity in the right parahippocampal gyrus correlated positively with neuropsychological performance. There was no significant correlation between subjects' characteristics and signal change in young subjects. These results indicate the age-associated vulnerability of the medial temporal lobe structures including the amygdala, hippocampus, and parahippocampal gyrus during face perception. The dissociation with reduced activity in the left amygdala and the right parahippocampal gyrus may suggest that aging differentially affects neural responses to faces with negative or positive emotional valence. The parieto-occipital lobe, which has been found to be involved in face processing, also showed a functional decline associated with aging. *Hippocampus 2002;12:352–362.***


**KEY WORDS: neuroimaging; activation; amygdala; parahippocampal gyrus; hippocampus**

## INTRODUCTION

The role of the medial temporal lobe structure, particularly the amygdala, in face perception has been investigated in monkeys (Leonard et al., 1985; Nishijo et al., 1988; Nakamura et al., 1992) and in humans (Adolphs et al., 1994). Recent neuroimaging studies also demonstrated activation of the amygdala during emotional face perception (Breiter et al., 1996; Morris et al., 1996, 1998a,b; Phillips et al., 1997; Schneider et al., 1997; Whalen et al., 1998; Blair et al., 1999; Hariri et al., 2000). Some studies suggest that the processing of negative faces specifically involves the left amygdala (Morris et al., 1996; Schneider et al., 1997; Blair et al., 1999). These findings imply that the amygdala plays a prominent role in stimulus-affective association, or in producing appropriate social behavior in primates, including humans (Aggleton, 1992). It is also important to note that functional decrements in the medial temporal lobe cause severe impairments in perception and memory (Squire and Zola-Mogan, 1991; Adolphs et al., 1994; Young et al., 1995). Local neuronal loss (Giannakopoulos et al., 1997; Jack et al., 1997) or decreased neuronal activity as measured by positron emission tomography (Martin et al., 1991; Murphy et al., 1996) in the medial temporal lobe has been shown to be associated with normal aging processes. Neuroimaging studies demonstrated that the hippocampus has significant activation during the encoding of faces or pictures in young adults, but that this activity is reduced in older adults (Grady et al., 1995, 1999). However, to date, age differences in the neural responses of the human amygdala have not been reported.

The aim of the present study was to explore age-related differences in the neural substrates involved in emotional face perception, focusing particularly on the functional

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neuroanatomy of the medial temporal lobe, including the amygdala, hippocampus, and parahippocampal gyrus. To do this, we acquired functional magnetic resonance imaging (fMRI) in healthy young and old subjects while they were performing gender discrimination tasks involving unfamiliar faces with negative, positive, or neutral emotional valence. Given that amygdala is thought to generally effect a stimulus-affective association based on learning and memory (Nishijo et al., 1988), and that the patients with bilateral amygdaloid damage were impaired at recognizing angry or disgusted faces as well as fearful faces (Adolphs et al., 1999), we used negative faces depicting an angry or disgusted expression. The imaging data were investigated on a voxel-by-voxel basis according to the random effect model (Friston et al., 1999). We predicted that the old subjects would show reduced activity in the medial temporal lobe regions, particularly in the amygdala.

In order to explore possible interactions between the subjects' characteristics and the neural responses that could not be identified by subtraction analysis, the participants' age and neuropsychological test scores were entered into correlation analysis. We predicted that activity in the medial temporal lobe structures would correlate negatively with age (Esposito et al., 1999). A PET study in patients with Alzheimer's disease during resting condition showed that cerebral glucose metabolism in the hippocampus positively correlated with performance of verbal episodic memory (Desgranges et al., 1998). A study with normal older adults and volumetric MRI analysis found that size of the hippocampal region was significantly associated with a decline in performance of verbal recall (Golomb et al., 1996). Raz et al. (1998) also reported that among subjects aged above 60, reduction in the volume of limbic structures predicted declines in explicit verbal memory. Therefore, we hypothesized that in old subjects, scores of neuropsychological tests, particularly those of verbal memory, would correlate with signal changes in the medial temporal lobe structures during the task condition.

## MATERIALS AND METHODS

### Subjects

Twelve young (6 male and 6 female, 19–39 years old, mean age 25.1 ± 5.0) and 12 old (6 male and 6 female, 62–72 years old, mean age 65.2 ± 2.6) subjects participated in the study after giving written informed consent. The subjects were community-dwelling, healthy, and independent-living adults. The subjects' physical health was verified in an interview before the study, and those who had a history of neurological diseases, psychiatric diseases, or drug or alcohol abuse were excluded. No subject was taking drugs that could affect the cerebral blood flow at the time of the study. No subject had incidental cerebral infarctions, as determined by high-resolution T2-weighted images. Except for one young adult who was ambidextrous, all subjects were strongly right-handed (assessed by the Edinburgh Handedness Inventory). This study was approved by the Ethics Committee at Fukui Medical University.

Before the experiment, a shorter version of the experimental task involving neutral faces was administered in order to confirm that subjects could perform at an average level. After the scanning, neuropsychological tests were conducted. The young adults outperformed the old adults in trail-making A (22.4 ± 6.2 vs. 31.5 ± 5.3, $P < 0.01$), digit symbol (82.1 ± 7.0 vs. 58.6 ± 6.2, $P < 0.01$), and cued recall of words (Wechsler Memory Scale, 19.3 ± 2.0 vs. 16.1 ± 3.6, $P < 0.05$). However, the performance of the old adults was within normal range (Spreen and Strauss, 1998). The immediate recall of figures results (Wechsler Memory Scale, 13.8 ± 0.3 vs. 12.4 ± 2.5), Self-Rating Depression Scale (SDS) scores (Zung, 1965; 30.5 ± 3.0 vs. 32.1 ± 6.1), and education years (16.0 ± 1.8 vs. 15.8 ± 0.5) did not differ between groups. The neuropsychological tests scores, except for those from the SDS, were subjected to principal component analysis in order to compute the score representing general cognitive function for each subject. The results showed that the digit symbol and word recall tests had a positive vector, and the trail-making results had a negative vector in the first principal component.

### Task Paradigm

Digitized grey-scale pictures of 24 unfamiliar faces (12 male and 12 female) with negative, positive, or neutral emotional valence were used. The validity and reliability of the portrayal of emotions in these pictures were tested in a new cohort (who did not participate in fMRI experiment) of young (n = 10; mean age, 36 years) and old (n = 10; mean age, 63 years) subjects. The subject was told to label the emotion expressed by the face from the six basic emotions (happy, angry, disgusted, fearful, sad, and surprised) and to rate the intensity of the emotion on a 5-point scale. The probability that a positive face was labeled as happy face was 99% for the young subjects and 88% for the old subjects. Mean (± SD) intensity rating of the happy face was 2.5 ± 0.74 and 2.36 ± 0.72 for young and old subjects, respectively. The young subjects labeled the negative faces as having angry (41%), disgusted (38%), sad (17%), or fearful (3%) faces. Mean intensity rating of these faces was 2.75 ± 0.3 for angry, 2.66 ± 0.25 for disgusted, 2.05 ± 0.44 for sad, and 2.5 ± 0.53 for fearful faces. The old subjects labeled the negative faces as having angry (25%), disgusted (35%), sad (28%), and fearful (10%) faces. The mean intensity rating of these faces was 2.87 ± 0.63 for angry, 2.81 ± 0.6 for disgusted, 2.88 ± 0.71 for sad, and 2.67 ± 0.78 for fearful faces. Mean intensity rating for the neutral faces was 0.1 ± 0.29 and −0.1 ± 0.53 for the young and old subjects, respectively. A 2 × 2 ANOVA performed on these rating scores, with group (young × old) and valence (positive × negative × neutral) as factors, revealed that the main effect of valence was significant ($F[2,138] = 340$, $P < 0.001$), but that the effect of age ($F[1,138] = 0.07$, $P = 0.78$) or their interaction ($F[2,138] = 2.34$, $P = 0.1$) was not significant.

Thus, in the present study, the neural substrate involved in perceiving a "negative" face condition is considered to be a reflection of the combination of cognitive processes for perceiving angry and disgusted faces for young subjects. Several behavioral studies have shown that among the six basic expressions, anger and disgust were perceived to be similar (Russell and Bullock, 1985; Adolphs et

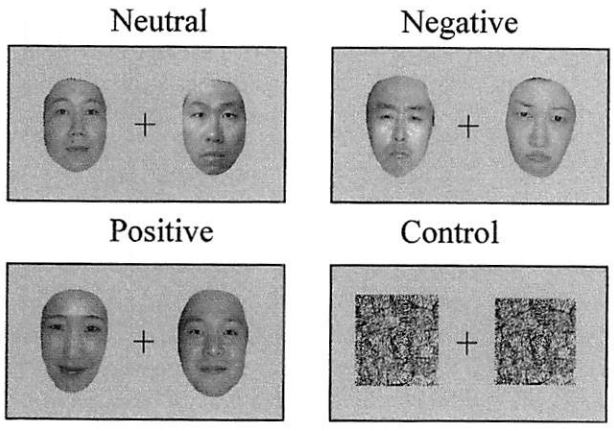


**FIGURE 1. Examples of stimuli used in the experiment.**

al., 1999). The results of a recognition task for these two expressions were considerably intermixed when the subjects grouped together faces which appeared to look alike (Russell and Bullock, 1985), as well as when they labeled the expressions and rated their intensities (Adolphs et al., 1999). Therefore, it is reasonable to assume that the perception of these two expressions may share common neural substrates. The old subjects labeled the negative faces as having sad faces more often than did the young subjects; however, the mean intensity rating of the emotional valence did not differ between young and old subjects.

A male face was paired with a female face within the same emotional category to create 12 pairs of faces (two faces were horizontally arranged on the screen) with negative, positive, or neutral emotional valence (Fig. 1). Each list of 12 face pairs was assigned to each experimental block (negative block, positive block, or neutral block). The task was to discriminate the gender of the faces. The subject was told to judge the side (left or right) of the target gender (male or female), and to respond by pressing the left or right button of the response box. The location (i.e., left side vs. right side) of the male face and the female face was randomized within the list. Half of the male and half of the female subjects responded to the male faces, and the other half of the male and half of the female subjects responded to the female faces. No particular instruction was given to pay attention to the emotional valence of the faces. During a control, the subject judged the size of two rectangles arranged horizontally on the screen (Fig. 1, lower right), and responded to the larger one by pressing the left or right button. Four repetitions of a control block (C) and three face blocks (F), with an additional control block at the end of the run, were presented to the subject (C-F-F-F-C-F-F-F-C-F-F-F-C-F-F-F-C). Each face was paired with a different face in each repetition. The order of face blocks was counterbalanced across all subjects (neutral-positive-negative, negative-neutral-positive, or positive-negative-neutral). The face pairs were presented at a rate of 2.5 s/pair with a 0.2-s intertrial interval. The stimuli were projected onto a half-transparent screen by an LCD projector connected to a personal computer in which the stimuli were generated. The subject saw the stimuli through a tilted mirror attached to the head coil of the scanner. Subjects' responses were recorded in order to compute the percentage of correct responses.

## Image Acquisition and Analysis

Functional images of the whole brain were acquired using the 3 Tesla MRI system (GE, Milwaukee, WI) equipped with single-shot EPI (TR = 4 s, TE = 30 ms, flip angle = 90°, 64 × 64 matrix and 44 slices, 2.7-mm slice thickness with a 0.3-mm gap). After discarding the first six images, the successive 136 images (eight images in each block) were subjected to analysis. Each block lasted 32 s. A high-resolution anatomical image (T2-weighted) was also acquired (TR = 6 s, TE = 68 ms, flip angle = 90°, 256 × 256 matrix, 2D-FSE, 1.5 mm interleaved in 7 mm). The functional images were realigned to the final image by SPM99 (Statistical parametric mapping; Wellcome Department of Cognitive Neurology, http://www.fil.ion.ucl.ac.uk/spm), and normalized to the standard space of Talairach and Tournoux (1988) by using parameters obtained from the normalization of a coregistered anatomical image to the MNI T2-weighted template. Finally, the images were smoothed by 8-mm Gaussian kernel.

## Subtraction Analysis

Statistical analysis in the present study was conducted according to the random effect model (Friston et al., 1999) so that inferences could be made at the population level. First, the signal time-course for each subject was modeled with a boxcar function convolved with a hemodynamic response function and high-pass filtering (256 s). The signal was proportionally scaled by setting the whole brain mean value to 100 arbitrary units. In the separate group analysis for young and old subjects, each of the negative, positive, or neutral face conditions was contrasted with the control condition, thereby creating one contrast image per subject for each face condition. These images were entered into a one-sample *t*-test to investigate the significant activation during each task. Significant signal changes for each contrast were assessed using *t*-statistics on a voxel-by-voxel basis (Friston et al., 1995). The resulting areas of activation were characterized in terms of their peak height and spatial extent. The statistical threshold was set to $P = 0.001$ ($T = 4.02$) for height, and clusters larger than 10 contiguous voxels were reported. In the between-group analysis, the contrast images created in the separate group analysis were entered into a two-sample *t*-test (masked by activation during three face conditions in 24 subjects at $P = 0.1$). The statistical threshold was set to $P = 0.001$ ($T = 3.50$) for height, and clusters larger than 5 contiguous voxels were reported. In the between-group analysis, the results for subtraction comparing the face conditions to each other (i.e., negative minus neutral, positive minus neutral, and negative minus positive) were also reported for a descriptive purpose. In these contrasts, a lenient threshold was used, and the search region was restricted to the medial temporal lobe.

## Correlation Analysis

The aim of the analysis was to explore the relationship between overall activation during the tasks and the subjects' characteristics. Age (mean corrected), and the score on the first principal component of the four neuropsychological tests (digit symbol, trail-making, word recall, and figure recall), were entered into a simple

*TABLE 1.*

*Significant Activation During Negative Face Condition*[†]

| | L/R | Region (BA) | T value | Coordinate |
|---|---|---|---|---|
| Young | R | Fusiform gyrus (37) | 14.25* | 40, −78, −16 |
| | L | Fusiform gyrus (37) | 8.50* | −40, −76, −16 |
| | R | Inferior frontal gyrus (46) | 9.48* | 50, 28, 18 |
| | L | Inferior frontal gyrus (46) | 6.40* | −40, 22, 22 |
| | R | Amygdala | 6.58* | 22, −8, −16 |
| | L | Amygdala | 8.64* | −18, −6, −16 |
| | R | Angular gyrus (39) | 6.36 | 32, −66, 36 |
| | L | Midbrain | 6.25 | −2, −36, −8 |
| | R | Inferior temporal gyrus (20) | 5.75 | 30, −10, −34 |
| | L | Lingual gyrus (18) | 5.70* | −16, −74, −16 |
| | R | Lingual gyrus (18) | 5.00 | 16, −72, 0 |
| Old | R | Fusiform gyrus (37) | 9.81* | 44, −52, −26 |
| | L | Fusiform gyrus (37) | 9.58* | −42, −68, −24 |
| | R | Inferior frontal gyrus (44) | 7.54* | 40, 16, 24 |
| | L | Inferior frontal gyrus (44) | 8.69* | −40, 16, 24 |
| | R | Cingulate gyrus (32) | 5.88 | 2, 18, 40 |
| | R | Middle temporal gyrus (37) | 5.49 | 52, −66, 4 |

[†]L, left; R, right; BA, Brodmann area. Degrees of freedom = (1,11).
*Significance ($P$ = 0.05) after multiple comparisons.

regression analysis as covariates. Because the two groups differed significantly in age and neuropsychological performance, these analyses were conducted separately for young and old subjects. Then, to clarify which of the four neuropsychological tests was critically involved in brain activation, each test score (z-transformed) was separately entered into a regression analysis. We hypothesized that signal change in the medial temporal lobe would negatively correlate with age (Esposito et al., 1999), and positively correlate with a neuropsychological test score reflecting the subject's verbal memory (Golomb et al., 1996; Desgranges et al., 1998; Raz et al., 1998). Therefore, the analysis was restricted to regions in the amygdala, hippocampus, and parahippocampal gyrus. The statistical threshold was set at $P = 0.001$ (T = 4.14) for height, and clusters larger than 5 contiguous voxels were reported.

In the between-group analysis, amygdaloid activation was considered significant at $P = 0.05$ after correction for multiple comparison in a 2 × 2 × 2 cm search region (Morris et al., 1996). The region names (according to the human brain atlas of Duvernoy, 1999), T-values, and coordinates of activated foci are listed in Tables 1–3. The results of significant activation under positive and neutral conditions for the young and old subjects are listed in Tables 4 and 5. Activated clusters were superimposed on the mean image of the high-resolution anatomical images (T2-weighted) of each subject (Figs. 2–4 and 6). The signal changes in the left amygdala, right parahippocampal gyrus, right prefrontal cortex, and left fusiform gyrus in young and old subjects are plotted in Figure 5. The results of the correlation analysis are also plotted in Figure 7.

## RESULTS

### Behavioral Data

The mean (± SD) percentage of correct responses for the gender discrimination task given by the young subjects was 98 ± 4% for the negative face condition, 99 ± 1% for the positive face condition, and 99 ± 2% for the neutral face condition. The old subjects performed equally well in recognizing each face (98 ± 3% for the negative, 99 ± 2% for the positive, and 97 ± 4% for the neutral condition). A two-way ANOVA showed no significant main effect of age (F[1,70] = 0.91, $P = 0.34$), task condition (F[2,69] = 1.24, $P = 0.29$), or interaction (F[2,69] = 0.53, $P = 0.59$). Both groups performed at ceiling level in responding to the control condition (over 99% correct).

### Neuroimaging Data

Young subjects activated the bilateral amygdala, prefrontal cortex (BA46), fusiform gyrus (BA37), and lingual gyrus (BA18) in responding to the negative face condition (Table 1). The angular gyrus (BA39) and the inferior temporal gyrus (BA20) in the right hemisphere also showed significant activation. The distribution of activation during the positive or neutral condition was similar to that during the negative condition (Tables 4 and 5). A particularly notable finding was that the activation of the left amygdala was greater during the negative condition than during the other conditions (Fig. 2, left). The activation in the posterior part of the right parahippocampal gyrus was predominant during the positive con-

TABLE 2.

*Age-Related Differences During Negative, Positive, and Neutral Face Conditions*[†]

| | L/R | Region (BA) | T value | Coordinate |
|---|---|---|---|---|
| Young > old | | | | |
| Negative condition | | | | |
| | L | Amygdala | 3.97* | −16, −6, −20 |
| Positive condition | | | | |
| | R | Parahippocampal gyrus | 4.77* | 14, −38, −6 |
| | R | Lingual gyrus (18) | 4.61 | 12, −66, −2 |
| | R | Angular gyrus (39) | 4.57 | 34, −68, 40 |
| Neutral condition | | | | |
| | L | Midbrain | 5.03 | −4, −38, −6 |
| | R | Midbrain | 4.43 | 8, −38, −8 |
| | L | Lingual gyrus (18) | 4.46* | −14, −74, −14 |
| Old > young | | | | |
| | No significant voxel | | | |

[†]L, left; R, right; BA, Brodmann area. Degrees of freedom = (1, 22).
*Significance ($p$ = 0.05) after correction for multiple comparison (under the negative condition, search region was restricted to the amygdala).

dition (Fig. 3, left). In older subjects, the prefrontal cortices and fusiform gyri were also significantly activated during the tasks (Table 1), while the amygdalae were weakly activated at the threshold of $P = 0.05$ (T = 1.80, Fig. 2, right). The older subjects did not show significant activation in the right parahippocampal gyrus, even at a low threshold ($P = 0.05$, Fig. 3, right).

A direct comparison between groups revealed age-related differences in the left amygdala in response to the negative face condition, and in the right parahippocampal gyrus in response to the positive face condition (Table 2). Both of these activations survived a spatial multiple comparison at $P = 0.05$ (under the negative condition, the search region was restricted to the amygdala; Morris et al., 1996). These clusters were superimposed on the mean anatomical image of the 24 subjects (Fig. 4). A plot of the adjusted signal change in comparison with the control condition demonstrated that the young subjects showed greater activation during both tasks (Fig. 5A,B). The young subjects had significantly greater activity in the right angular gyrus (BA39) and lingual gyrus (BA18) during the positive condition. Age differences were also found in the posterior part of the midbrain and lingual gyrus (BA18) during the neutral condition. The magnitudes of signal change in the right prefrontal cortex and left fusiform gyrus of both groups are plotted in Figure 5C,D for purpose of comparison. Age difference in subtraction comparing the face conditions to each other did not survive our statistical threshold; however, young subjects had greater activation in the bilateral amygdala in the negative minus neutral contrast than did old subjects (left, x, y, z = −18, −4, −20, T = 2.42, $P < 0.05$; right, x, y, z = 16, 0, −18, T = 2.66, $P < 0.01$). Young subjects had greater activation in the right parahippocampal gyrus in the positive minus neutral contrast (x, y, z = 12, −32, −12, T = 2.63, $P < 0.01$), and in the left parahippocampal gyrus in the negative minus positive contrast (x, y, z = −12, −46, −8, T = 2.92, $P < 0.01$) than did old subjects. When three face conditions were combined together and contrasted with the control condition, the young subjects had significantly greater activation in the left amygdala (x, y, z = −14, −8, −20, T = 4.18, $P <$ 0.001), right parahippocampal gyrus (x, y, z = 12, −40, −8, T = 4.32, $P < 0.001$), and bilateral lingual gyri (x, y, z = 12, −66, −2,

TABLE 3.

*Significant Correlation Between Signal Change and Subjects' Characteristics*[*]

| | Group | Pos/Neg | L/R | Region (BA) | T value | Coordiante |
|---|---|---|---|---|---|---|
| Age | | | | | | |
| | Old | Neg | R | Hippocampus | 4.94 | 30, −18, −22 |
| Neuropsychological performance | | | | | | |
| | Old | Pos | R | Parahippocampal gyrus | 6.93 | 14, −8, −26 |

[*]Pos, positive correlation; Neg, negative correlation; Neuropsychological performance, score of first principal component of neuropsychological tests. Degrees of freedom = (1, 10) for neuropsychological performance. Degree of freedom = (1, 9) for age.

*TABLE 4.*

*Significant Activation During Positive Face Condition*[†]

| | L/R | Region (BA) | T value | Coordinate |
|---|---|---|---|---|
| Young | R | Fusiform gyrus (37) | 12.40* | 48, −62, −24 |
| | L | Fusiform gyrus (37) | 9.20* | −40, −76, −16 |
| | R | Parahippocampal gyrus | 11.13* | 14, −42, 0 |
| | R | Inferior frontal gyrus (44) | 10.92* | 44, 12, 26 |
| | L | Inferior frontal gyrus (44) | 5.07 | −36, 6, 32 |
| | R | Angular gyrus (39) | 5.87 | 34, −68, 38 |
| | L | Angular gyrus (39) | 4.77 | −28, −66, 36 |
| | R | Amygdala | 5.66* | 18, −2, −18 |
| | L | Amygdala | 5.13 | −14, −4, −20 |
| | L | Cerebellum | 5.50 | −2, −82, −26 |
| Old | L | Fusiform gyrus (37) | 9.77* | −42, −70, −22 |
| | R | Fusiform gyrus (37) | 9.26* | 40, −72, −22 |
| | L | Lingual gyrus (18) | 7.83 | −10, −80, −6 |
| | L | Inferior frontal gyrus (44) | 7.50* | −38, 16, 24 |
| | R | Inferior frontal gyrus (46) | 7.15 | 50, 30, 18 |
| | L | Middle temporal gyrus (39) | 6.05 | −34, −80, 14 |

[†]L, left; R, right; BA, Brodmann area.
*Significance ($P$ = 0.05) after multiple comparisons.

T = 4.24, $P < 0.001$; x, y, z = −14, −76, −16, T = 4.04, $P <$ 0.001).

The results of the correlation analysis are summarized in Table 3. There was a significant negative correlation between age and adjusted response in the right medial temporal lobe in the 12 old subjects; this correlation, however, may have been derived from the oldest subject in the group. Therefore, we reanalyzed the data after excluding this subject, and again found a significant negative cor-

*TABLE 5.*

*Significant Activation During Neutral Face Condition*[†]

| | L/R | Region (BA) | T value | Coordinate |
|---|---|---|---|---|
| Young | R | Fusiform gyrus (37) | 12.28* | 40, −78, −16 |
| | L | Fusiform gyrus (37) | 10.91* | −38, −74, −14 |
| | R | Inferior frontal gyrus (46) | 12.67* | 48, 28, 18 |
| | L | Inferior frontal gyrus (44) | 6.59* | −38, 10, 30 |
| | R | Midbrain | 8.00* | 6, −36, −8 |
| | R | Amygdala | 6.73* | 24, 2, −20 |
| | L | Amygdala | 6.99 | −16, −8, −18 |
| | R | Angular gyrus (39) | 6.49 | 34, −66, 36 |
| | L | Cerebellum | 6.39 | −14, −48, −50 |
| | R | Lingual gyrus (18) | 5.27 | 10, −70, −8 |
| | R | Superior temporal gyrus (38) | 5.04 | 38, 12, −30 |
| Old | L | Fusiform gyrus (37) | 10.69* | −42, −64, −24 |
| | R | Fusiform gyrus (37) | 10.37* | 40, −72, −22 |
| | R | Middle temporal gyrus (39) | 8.64 | 34, −66, 12 |
| | R | Middle frontal gyrus (9/46) | 8.07* | 50, 28, 30 |
| | R | Inferior frontal gyrus (46) | 6.38 | 48, 32, 16 |
| | L | Middle frontal gyrus (9) | 5.99* | −46, 22, 32 |
| | L | Superior frontal gyrus (6) | 6.46 | −2, 14, 56 |

[†]L, left; R, right; BA, Brodmann area.
*Significance ($P$ = 0.05) after multiple comparisons.

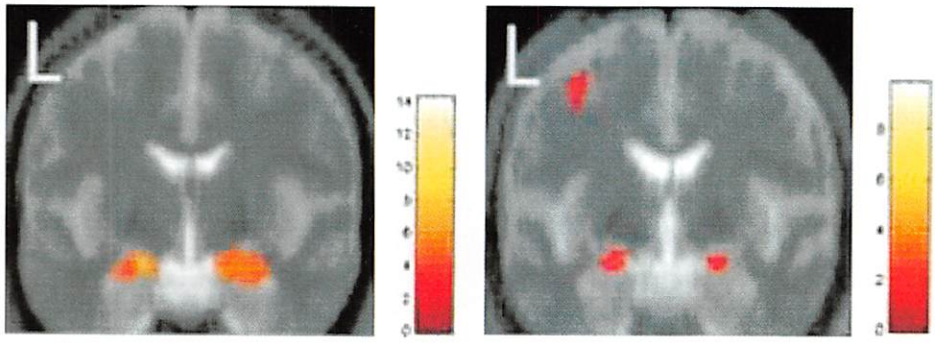

FIGURE 2. Activation in amygdalae in response to negative face condition for young (left) and old (right) subjects. Activation was superimposed on the mean T2-weighted images for each group. Coronal images were shown at y = −6, where a significant age difference was observed. Note that the statistical threshold is set at $P = 0.001$ for young subjects and at $P = 0.05$ for old subjects. Bilateral amygdaloid activation was observed in young subjects, with greater activity in the left amygdala during the negative face condition (left).

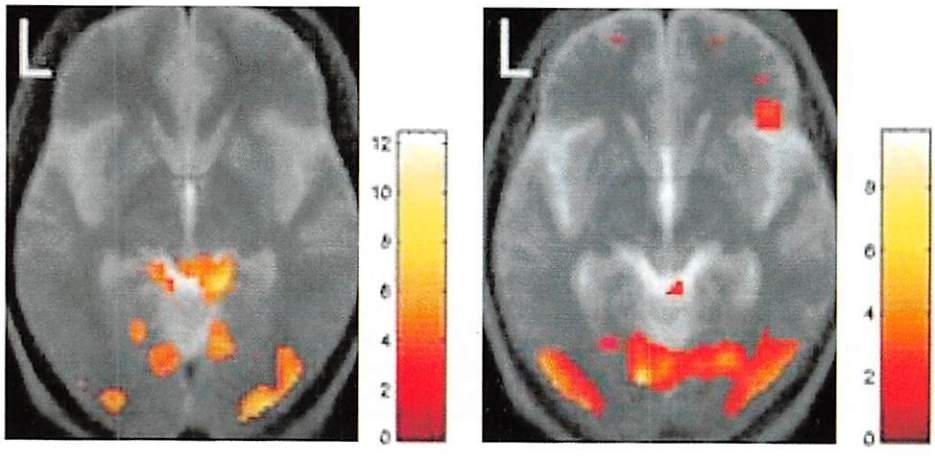

FIGURE 3. Activation in posterior part of parahippocampal gyrus in response to the positive face condition for young (left) and old (right) subjects. Activation was superimposed on the mean T2-weighted images for each group. Axial images were shown at z = −6, where a significant age difference was observed. Note that the statistical threshold is set at $P = 0.001$ for young subjects and at $P = 0.05$ for old subjects. Significant activation in the right parahippocampal gyrus was observed in young subjects, but not in old subjects.

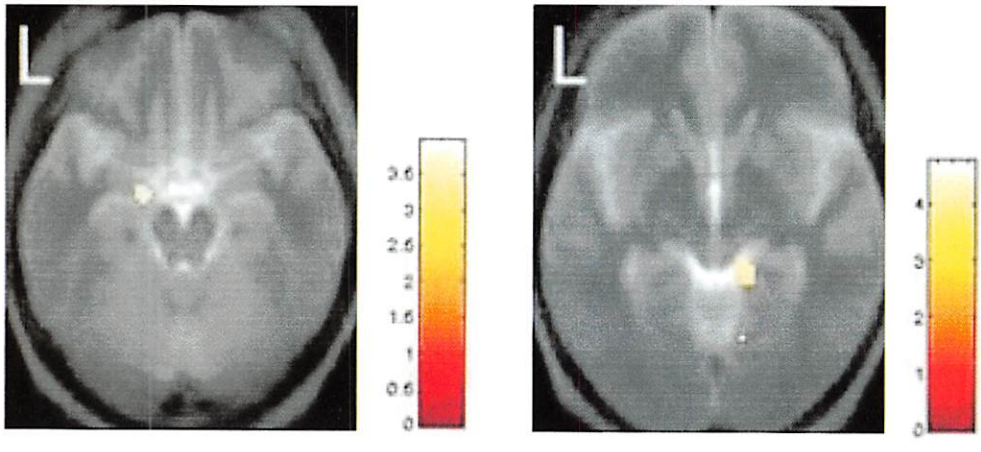

FIGURE 4. Results of between-group comparison. The cluster was superimposed on the mean T2-weighted images of the 24 subjects. Significant age differences were observed in the left amygdala in response to the negative condition (left, z = −20) and in the right parahippocampal gyrus in response to the positive condition (right, z = −6). Peak coordinates are shown in Table 2. The statistical threshold was set at $P = 0.001$. These clusters survived correction for multiple comparison at $P = 0.05$.

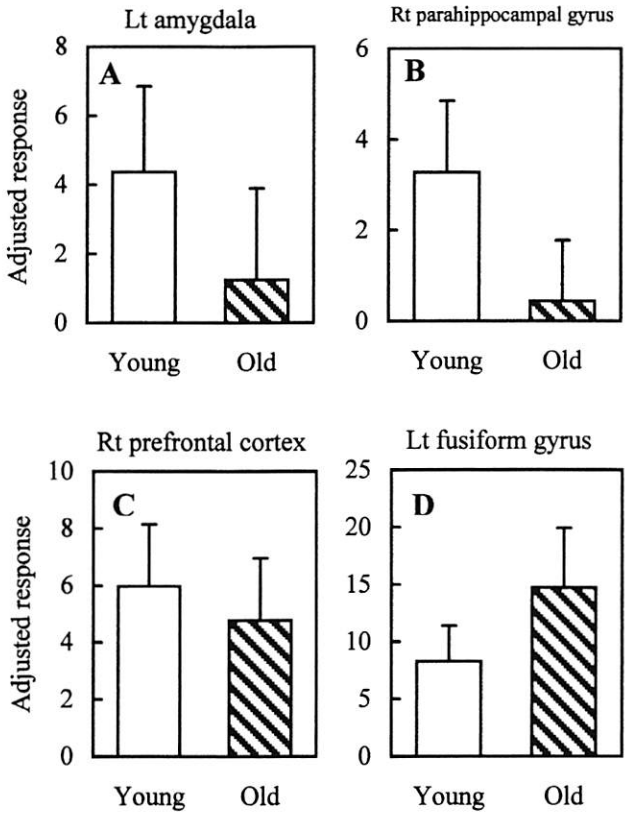


**FIGURE 5. Signal changes from control condition in left amygdala under the negative condition (A), right parahippocampal gyrus under the positive condition (B), right prefrontal cortex under the negative condition (C), and left fusiform gyrus under the positive condition (D). Columns represent mean adjusted response (bar: SD) in the peak voxel of each cluster for both age groups.**

relation in the right hippocampus (Fig. 6, left; Fig. 7A, left). There was no significant correlation between age and neural response in the young subjects. The neuropsychological tests score showed a positive correlation with adjusted response in the right parahippocampal gyrus in the old subjects (Fig. 6, right; Fig. 7A, right). Among the four neuropsychological tests, only the score of word recall had significant positive correlation with right parahippocampal activity (x, y, z = 16, −12, −26, T = 10.67, $P < 0.001$). The score of digit symbol (x, y, z = 14, −8, −26, T = 2.98, $P = 0.007$) and that of trail-making (x, y, z = 14, −6, −24, T = 2.88, $P = 0.008$) had positive correlation with activity in this region; however, they were not statistically significant. The subject's score and signal change in the parahippocampal gyrus are plotted in Figure 7B separately for each neuropsychological test. There was no significant correlation between test scores and neural response in the young subjects.

## DISCUSSION

The involvement of the human amygdala in a face perception task was investigated using PET (Morris et al., 1996, 1998a,b; Blair et al., 1999) and fMRI (Breiter et al., 1996; Phillips et al., 1997; Schneider et al., 1997; Whalen et al., 1998; Hariri et al., 2000). Most previous studies found that greater blood flow or signal changes occurred in response to fearful face conditions than in response to neutral or happy face conditions. A recent fMRI study showed that the gender judgment task for emotional faces, as employed in the present study, activated the human amygdala more than did explicit judgment of expressions (Critchley et al., 2000). A lesion study in humans also demonstrated significant involvement of the amygdala in the recognition of fearful face expressions (Adolphs et al., 1994). However, the involvement of the amygdala in the processing of disgusted or angry faces has not been reported in neuroimaging studies (Phillips et al., 1997; Sprengelmeyer et al., 1998). In studies with monkeys, neurons in the amygdala responded to the presentation of human faces (Leonard et al., 1985; Nakamura et al., 1992) and objects with affective significance (Nishijo et al., 1988), indicating that the amygdala

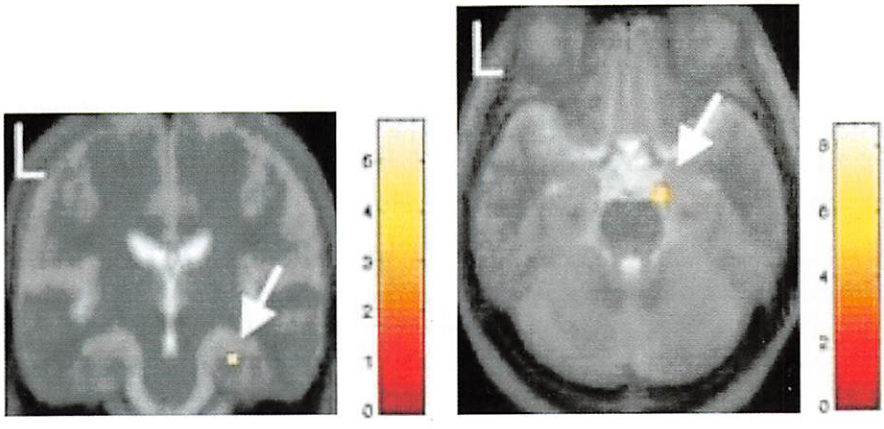


**FIGURE 6. Results of correlation analysis. Activity was superimposed on the mean T2-weighted images for old subjects. Left: Activity in the right hippocampus in old subjects had a negative correlation with age. Right: Activity in the right parahippocampal gyrus in old subjects had a positive correlation with neuropsychological performance. Peak coordinates are shown in Table 3.**

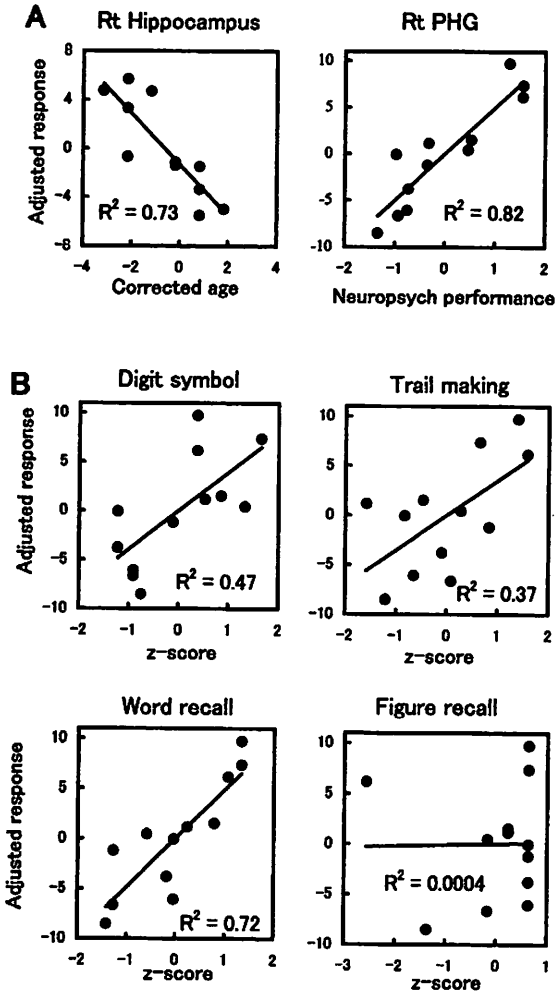


**FIGURE 7.** **A: Plots of significant correlations between old subjects' characteristics and the adjusted response (left, age vs. right hippocampus; right, neuropsychological performance vs. right parahippocampal gyrus). The x-axis represents the characteristics, and the y-axis represents the adjusted response. Note that the correlation between age and response was computed in 11 old subjects. PHG, parahippocampal gyrus. B: Plots of correlation between the z-score of each neuropsychological test and the adjusted response in the right parahippocampal gyrus of old subjects. Note that scores for the trail-making test is multiplied by −1 to show positive correlation with signal. The regression coefficient ($R^2$) is shown in each graph.**

generally effects a stimulus-affective association based on learning and memory. A behavioral study of nine individuals with bilateral amygdala damage showed that they did not correctly recognize angry or disgusted faces (Adolphs et al., 1999). This result in humans strongly suggests an involvement of the human amygdala in perceiving angry and disgusted faces as well as fearful ones, and supports the present finding with predominant activation in the amygdala under the negative condition.

The main aim of the present study was to investigate age differences in the neural substrates of emotional face perception. Direct comparison between groups of older and younger subjects revealed that activity in the left amygdala in response to a negative face condition was significantly greater in young subjects than in old subjects. The present results suggest that the old subjects' neural response to the negative faces was impaired, although their performance during the gender discrimination task was comparable to that of the young subjects. It is well-established that structures in the medial temporal lobe are particularly vulnerable to aging, as confirmed by neuropathological (Giannakopoulos et al., 1997; Davis et al., 1999) and MRI volumetric (Jack et al., 1997) studies. Therefore, the impairment of neural response in the left amygdala in old subjects is probably attributable to the neuronal loss or reduced vascular responses associated with aging. To date, several neuroimaging studies have used faces or pictures as stimuli to investigate age differences in the human medial temporal lobes (Grady et al., 1995, 1999). However, the tasks used in these studies did not involve stimuli that were laden with emotional content, and did not show any activation of the amygdala. The present study, to our knowledge, is the first to demonstrate a reduced neural response in the human amygdala in association with aging. We do not claim that the activity in the amygdala is specific to the recognition of faces, because several PET studies have shown a significant relationship between the amygdala and the perception of emotional pictures (Taylor et al., 1998; Hamann et al., 1999). The present results, however, apparently show that a significant age-related difference during negative face perception derived from the predominant neural activity in the left amygdala in young subjects and the reduced activity in old subjects.

In contrast to the negative condition, an age difference was observed in response to the positive faces in the right parahippocampal gyrus, where young subjects showed robust activation. The old subjects had significantly lower activity in this region, as shown in Figure 3. Behavioral studies have shown that happy faces are recognized more accurately and rapidly than faces exhibiting other emotions (Kirouak and Dore, 1983), particularly if the stimuli are presented to the left visual filed-right hemisphere (Duda and Brown, 1984; Hugdahl et al., 1989). The tendency to recognize happy faces more quickly, as observed in these earlier studies, might be related to the prominent activity in the right parahippocampal gyrus that was observed in the present study during positive face condition. It is notable that the peak coordinate of the parahippocampal activation in young subjects (x, y, z = 14, −42, 0) was in the vicinity of that reported by Kelley et al. (1998; x, y, z = 19, −35, 0) during the face-encoding task. Another neuroimaging study using PET and a face working-memory task found significant involvement of the right medial temporal lobe (Haxby et al., 1995). Although the task in the present study did not require mnemonic processing, it is possible that incidental encoding of the stimuli might have occurred while the subjects were judging the gender of the faces. Age-related differences in right medial temporal lobe activity were reported during tasks involving faces (Grady et al., 1995) and pictures (Grady et al., 1999). In line with these previous reports, the present study indicates that a vulnerability of the medial temporal lobe is associated with aging.

The old subjects exhibited reduced activity in the lingual gyrus in response to both the positive and neutral conditions, and showed reduced activity in the right angular gyrus in response to the positive condition. It has been well-documented that lesions in the medial surface of the occipito-temporal lobe could result in

prosopagnosia in humans (Damasio et al., 1982). Localized cortical removal of the parietal lobe in epileptic patients resulted in poor performance during a face-matching task (Kolb and Taylor, 1981), a finding that suggests that the parietal lobe plays a role in face perception. These studies support our results that signal changes during affective processing of face stimuli in the parieto-occipital regions were reduced in old subjects.

Several neuroimaging studies indicate that the relationship between brain activation and blood oxygen level dependent (BOLD) signal responses may be altered in elderly subjects. The amplitude of signal response in elderly subjects was significantly decreased compared to younger subjects during photic stimulation (Ross et al., 1997). The time lag between onset of task and rise of signal response was prolonged with increasing age (Taoka et al., 1998). Age differences were also observed in the signal-to-noise ratio and motion artifacts of imaging data (D'Esposito et al., 1999). These findings may represent reduced neuronal activation, reduced vascular response to normal activation, or an alteration in the coupling of blood oxygenation changes in response to focal activation in elderly subjects. It is argued that the age difference reported in the present study may simply relate to general group differences in overall BOLD activation sensitivity. However, as shown in Figure 5C, the magnitude of activation in the right prefrontal cortex of old subjects was comparable with that of young subjects. Moreover, old subjects activated the left fusiform gyrus more than did young subjects (Fig. 5D), although the difference was not significant in SPM analysis. Therefore, the present results indicate that age effect was the most significant in the medial temporal lobe structures under the negative or positive face condition, even though general group differences in BOLD response may exist.

The correlation analysis revealed significant relationships between activity in the medial temporal lobe and age and cognitive function of the old subjects. In the old subjects, a significant negative correlation existed between age and overall activity in the right hippocampus during the tasks. Esposito et al. (1999) found a significant negative correlation between age and parahippocampal activity during visuospatial tasks. In subjects at rest, age was associated both with decreased blood flow in the posterior part of the parahippocampal gyrus (Martin et al., 1991) and with decreased glucose metabolism in the hippocampus (Murphy et al., 1996). Another significant finding was that old subjects' neuropsychological performance positively correlated with signal changes in the right parahippocampal gyrus. This result implies that old subjects with higher cognitive functioning activated the medial temporal lobe during the task, whereas those with poorer cognitive functioning did not. We predicted from the studies reported by Golomb et al. (1996), Desgranges et al. (1998), and Raz et al. (1998) that verbal memory function would correlate with medial temporal lobe function in old subjects. The present results showed that among the four neuropsychological tests, the score of the word recall test was most sensitive to medial temporal lobe activity, as found in the separate regression analysis. Although previous studies investigated cerebral glucose metabolism in patients with Alzheimer's disease during resting condition (Desgranges et al., 1998), or size of hippocampal (Golomb et al., 1996) or limbic regions (Raz et al., 1998) by volumetric MRI analysis, these significant correlations, including the present one, that were observed in old subjects could run parallel with age differences in the subtraction analysis. The present study suggests a further possibility of using fMRI as a tool for noninvasive measurement of the medial temporal lobe function in humans.

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