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## Note

# Amygdalar enlargement associated with unique perception

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### ABSTRACT

Interference by amygdalar activity in perceptual processes has been reported in many previous studies. Consistent with these reports, previous clinical studies have shown amygdalar volume change in multiple types of psychotic disease presenting with unusual perception. However, the relationship between variation in amygdalar volume in the normal population and the tendency toward unusual or unique perception has never been investigated. To address this issue, we defined an index to represent the tendency toward unique perception using ambiguous stimuli: subjects were instructed to state what the figures looked like to them, and “unique responses” were defined depending on the appearance frequency of the same responses in an age- and gender-matched control group. The index was defined as the ratio of unique responses to total responses per subject. We obtained structural brain images and values of the index from sixty-eight normal subjects. Voxel-based morphometry analyses revealed a positive correlation between amygdalar volume and the index. Since previous reports have indicated that unique responses were observed at higher frequency in the artistic population than in the nonartistic normal population, this positive correlation suggests that amygdalar enlargement in the normal population might be related to creative mental activity.

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## 1. Introduction

Interference by emotion in perceptual processes ranges from everyday instances such as biased judgment in emotional situations (Mayer et al., 1992) to pathological instances such as exacerbation of hallucinatory symptoms under emotional stress in psychotic patients (Myin-Germeys et al., 2005). Because the limbic region, including its main component, the amygdala, has been considered to play critical roles in emotional processing (Vuilleumier, 2005), the modulatory roles of the amygdala in perceptual processes are easily expected

from these instances. In accordance with this view, numerous neurophysiological and neuroimaging studies have shown that signals from the amygdala strongly affect perceptual processing at the neuronal level (Kilpatrick and Cahill, 2003; Phelps and Ledoux, 2005). These findings suggest that if emotion-related circuits are recruited more frequently in a given subject than in others, his or her perception will be more easily affected by emotion than that in others. Therefore, assuming that frequent recruitment of a brain structure results in enlargement of its volume (Maguire et al., 2000), variation in volume of the amygdala might be observed in relation to individual difference

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in features of perception. However, few studies have focused on the relationship between the volume of the amygdala and individual characteristics of perception.

In previous clinical studies, structural abnormalities in amygdala have been reported in multiple types of psychotic disease associated with unusual perception. For example, amygdalar enlargement has been reported in temporal epilepsy patients with psychosis (Tebartz van Elst et al., 2002) and in patients with affective psychoses (Velakoulis et al., 2006). On the other hand, previous imaging studies of schizophrenia patients have reported volume deficits in multiple cerebral regions including the amygdala (Honea et al., 2005; Wright et al., 1999). In the normal population, variation in amygdalar volume has been reported in association with certain personality characteristics (Omura et al., 2005), but no previous imaging study has investigated variation in amygdalar volume in association with the tendency toward unusual perception. To address this issue, a quantitative index to represent the tendency toward unusual perception is required.

This type of index can be provided by a clinical test using ambiguous stimuli (Exner, 2003; Kircher et al., 2001; Perry et al., 1999; Rorschach, 1921). In this test, subjects are instructed to state what the figures look like to them. Quantitative evaluation of uniqueness in perception is enabled by classifying responses on the basis of appearance frequency of the same response in a control database. Specifically, responses found only at very low frequency in the control database can be considered as original or unique responses.

In our previous study (Asari et al., 2008), subjects viewed these ambiguous stimuli during functional MRI scans and instructed to say, vocally, what they look like to them. A subtraction analysis revealed the right temporopolar activation associated with unique versus frequent responses and the anterior prefrontal and bilateral occipitotemporal activation associated with frequent versus unique responses. Previous anatomical studies have shown the anatomical proximity between the temporal pole and the amygdala (Olson et al., 2007), thus the role of amygdala in production of unique responses is also suggested from this finding.

Some previous behavioral studies have reported that unique responses were observed at a higher frequency in the artistic population than in the nonartistic normal population (Ramachandra, 1994; Rorschach, 1921; Satterlee, 2006). The widely held belief that emotion may underpin imaginative or creative mental activities (Damasio, 2004; Modell, 1994) suggests that investigation of the correlation between this index and amygdalar volume might shed light on the neuronal basis of creative mental activities. In the present study, structural brain images of normal volunteers were obtained and voxel-based morphometry (VBM) analyses were performed to investigate the relationship between amygdalar volume and unique perception.

## 2. Methods

### 2.1. Subjects

Normal, right-handed, native Japanese volunteers were recruited by advertisement and were screened by structured

interview to exclude history of psychiatric or neurological illness. Sixty-eight subjects (41 females, age 20–36 years, each with more than 14-year educational history) participated in the imaging experiment. Most subjects (sixty-two out of sixty-eight) were undergraduate or graduate students with various specialties, and none of them had formal art education. A total of 217 subjects participated in the control experiment to create a control database. Age ( $25.1 \pm 4.6$ , mean  $\pm$  SD, range 20–39 years) and gender (female:male = 132:85) in the control group were matched with those in the experimental group (age  $23.3 \pm 3.5$ , range 20–36 years, female:male = 41:27), and the educational background in the control group (more than 14-year educational history) was similar to that in the experimental group. All subjects gave informed consent in keeping with experimental procedures approved by the Institutional Review Board of the University of Tokyo School of Medicine.

### 2.2. Task and imaging procedures

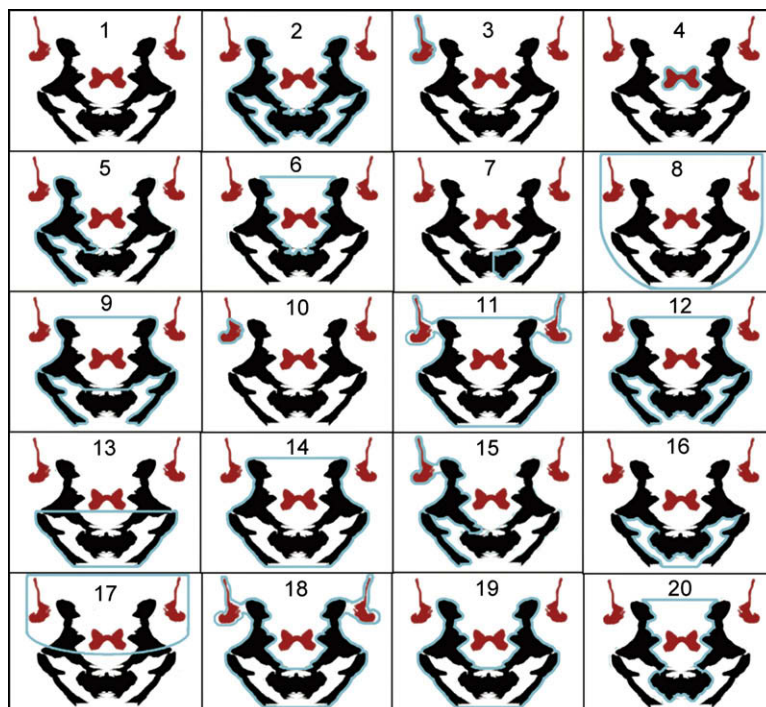
The experiments were conducted using a 1.5-T MRI system. T2-weighted spin-echo images were obtained for VBM analyses [repetition time (TR) = 5.5 sec; echo time (TE) = 30 msec; 75 slices, slice thickness = 2 mm; in-plane resolution =  $2 \times 2$  mm]. The task was performed in the MRI scanner during functional MRI scanning by projecting the ambiguous inkblot figures (Exner, 2003; Rorschach, 1921) onto a screen (one of the ten figures was presented for 3 min in each of ten functional runs). A reduced and blurred version of a stimulus example is presented in Fig. 1. The subjects were asked to report vocally what the figure looked like to them and were encouraged to provide as many answers as they wished. Their vocal responses were recorded using an MRI-compatible microphone. The results of functional imaging analyses were reported elsewhere (Asari et al., 2008; see also Section 1). In the control experiment, the same task was performed outside the scanner, and the subjects' responses were recorded to create the control database (see Section 2.3). Post-experimental interviews were conducted to confirm what the subjects saw and where they saw in the figures for each response in both the imaging and control experiments.

### 2.3. Definition of index

To compensate for the lack of normative data in the Japanese population, which is comparable to the data collected by Exner et al., and to exclude response bias due to difference in background including culture, education, age and gender, we classified the responses in terms of unique perception based on the frequency of responses in the matched control group. We defined "unique responses" in the experimental group as those not found in the control group. Note that "unique responses" were defined only for the experimental group. The "Unique Response Ratio" (URR) was then defined as the number of unique responses divided by the total number of responses per subject.

### 2.4. Voxel-based morphometry

VBM analyses were performed using SPM5 software (Ashburner and Friston, 2000, 2005; <http://www.fil.ion.ucl.ac.uk/>



**Fig. 1** – A stimulus example used in this study is presented in the cell 1. In the other cells (2–20), the location of each response example (see Appendix) in the stimuli is designated by blue lines. The Rorschach figure (Card III) was reprinted by kind permission of Verlag Hans Huber, Hogrefe AG, Bern/Switzerland.

spm/) according to an optimized protocol (Good et al., 2001), and then normalized segments of gray matter were generated and smoothed with a 10 mm full-width, half-maximum Gaussian kernel. Voxel values in the resulting images (unmodulated images) are considered to represent relative concentrations (volume concentrations) of gray matter in each voxel (Good et al., 2001). These images were further modulated by the Jacobian determinants derived from the spatial normalization steps, to obtain the supposed absolute volume of gray matter. In the present study, the unmodulated images were analyzed, since we were interested primarily in focal structures (such as the amygdala) and a previous study showed that the modulation step tends to reflect differences in global brain shape rather than in focal structures (Eckert et al., 2006). In keeping with previous VBM studies using normal subjects (Hulshoff Pol et al., 2006; Maguire et al., 2000; Omura et al., 2005), a multiple regression analysis was performed, including the URR score as the covariate of interest, and gender, age, and total gray matter volume as covariates of no interest. Significant peak voxels above  $P < .001$  (uncorrected) were reported (Table 1). The anatomically defined ROIs (regions of interest) used for small volume correction were generated by wfu\_pickatlas software (Maldjian et al., 2003; <http://www.fmri.wfubmc.edu/>).

### 3. Results

The mean numbers and standard deviations of total and unique responses by experimental subjects were  $39.4 \pm 17.6$

and  $11.6 \pm 8.6$ , respectively. Sixty-seven out of sixty-eight subjects provided at least one unique response. A list of example typical responses and unique responses for the example stimulus is presented in Appendix with the charts to designate the location of responses (Fig. 1). The contents of the responses ranged from ingenious ones in which fragmental visual features of the stimulus were successfully integrated to a whole elaborated response (see “a monkey reaching for a branch and a bird at the edge of water” in Appendix), to weird ones in which precise perception might have been interfered by the subject’s emotion or subconscious needs (see “underwear”, “bean paste soup in a china bowl”, or “birthday cake” in Appendix).

The structural brain images of all subjects were analyzed using a VBM analysis. The bilateral amygdalae exhibited significant positive correlations ( $P < .05$ , small volume correction using FDR) between volume concentration and URR (Fig. 2a; Table 1). The bilateral cingulate gyri, which are other components of the limbic system, also exhibited positive correlations at a less stringent threshold ( $P < .001$ , uncorrected). Positive correlations were also found in the other regions including the left lateral temporal regions, the right inferior occipital regions, the right precentral gyri, the right paracentral lobule, and the left caudate ( $P < .001$ , uncorrected). The individual volume concentration averaged in each of the independent anatomical ROIs for the left and right amygdala and the bilateral cingulate gyri (Fig. 2b) also exhibited significant partial correlations with URR ( $P < .05$  for the left amygdala and  $P < .01$  for the right amygdala and the bilateral cingulate gyri).

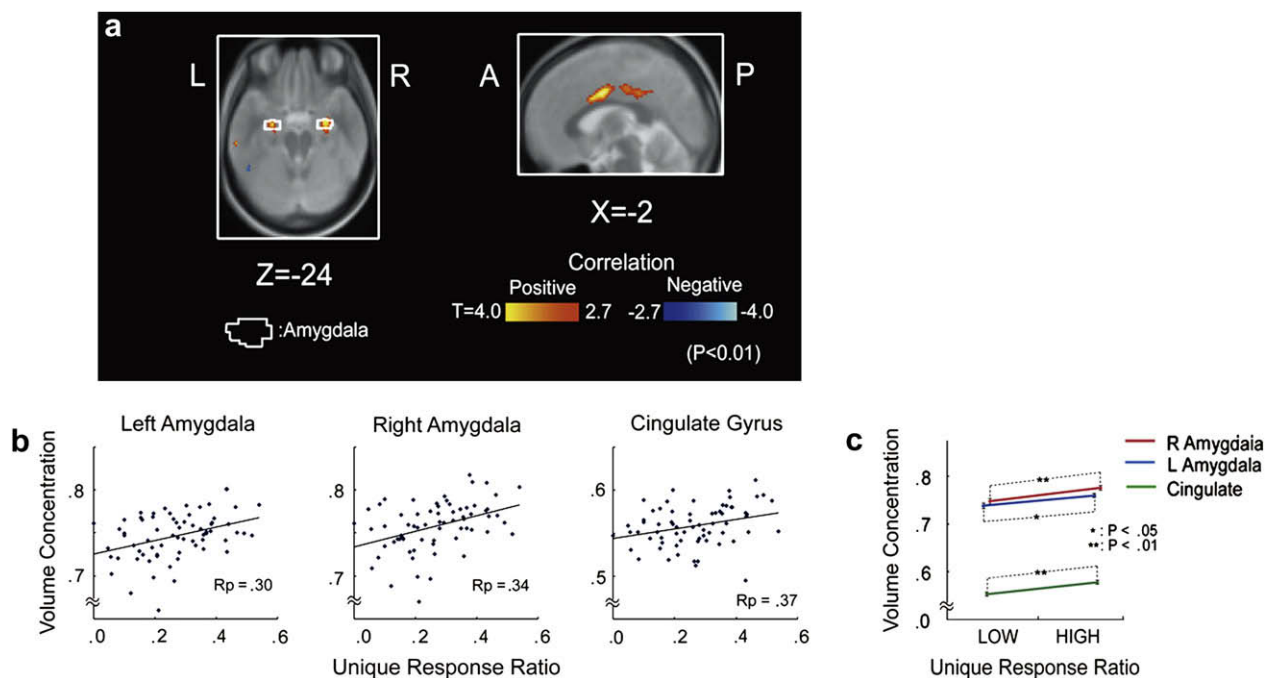
**Table 1 – Peak coordinates in VBM analysis**

Correlation	Hemisphere	Region	Coordinates			t-value	BA
			x	y	z		
Positive	Left	Limbic	-4	0	32	3.92	24
			-8	-46	32	3.68	31
			-26	-4	-24	3.45*	Amygdala
		Temporal	-64	-24	-20	3.77	20
			-58	-8	-32	3.62	21
	Basal Ganglia	-22	20	14	3.63	Caudate	
		Right	Limbic	26	-2	-24	3.70*
	4			-4	36	3.49	24
	Frontal		38	-8	42	3.61	6
			50	-10	26	3.58	6
Occipital	10		-32	62	3.65	4	
22	-72	12	3.87	17			
Negative		None					

Covariates of no interest (gender, age, and total gray matter volume) were included in the analysis.  
BA, Brodmann area.  
\*P < .05 small volume correction for bilateral amygdala.

Since there is a great variance in URR across the subjects (see also the scattergrams in Fig. 2b), an alternative analysis was performed to show individual difference in volume concentrations: the subjects group was divided into quartiles on the basis of URR, then the volume concentrations of the uppermost quartile group and the lowermost quartile group

was compared using a t-test. The volume concentrations were significantly different between the two groups (see Fig. 2c;  $T = 2.9$ ,  $P < .01$  for the right amygdala;  $T = 2.1$ ,  $P < .05$  for the left amygdala;  $T = 3.2$ ,  $P < .01$  for the cingulate gyri). A one-way ANOVA has also shown that the volume concentrations were significantly different across all the four groups (see Fig. 2c; F



**Fig. 2 – Results of VBM analyses. A. Statistical correlation map showing correlations between gray matter volume and URR (Unique Response Ratio). B. Individual volume concentrations averaged in the independent anatomical ROIs for the left and right amygdala and the bilateral cingulate gyri plotted against URR. L, left; R, right; Rp, partial correlation coefficient. C. Mean of volume concentrations across subjects belonging to the uppermost quartile group (right side) and the lowermost quartile group (left side) in terms of URR (Unique Response Ratio), averaged in the independent anatomical ROIs for the left and right amygdala and the cingulate gyri. The error bars represent standard errors across subjects. Significant difference is indicated by asterisks (\*P < .05; \*\*P < .01). L, left; R, right.**



(3, 16) = 8.8,  $P < .00001$  for the right amygdala;  $F(3, 16) = 3.8$ ,  $P < .05$  for the left amygdala;  $F(3, 16) = 6.8$ ,  $P < .01$  for the cingulate gyri).

#### 4. Discussion

The VBM analyses revealed positive correlations between gray matter volume concentration and unique perception in the bilateral amygdala, as well as in other important limbic components, the cingulate gyri. The reciprocal anatomical connections between the amygdala and the cingulate gyri have been reported in previous anatomical studies (Pandya et al., 1973) and the involvement of the cingulate gyri in emotional processing has also repeatedly been shown in previous neurophysiological and neuroimaging studies (Bush et al., 2000). Therefore, these results suggest that emotion-related neural circuits might underlie generation of unique perception. Positive correlations were also found in other cerebral regions including the left lateral temporal regions and the occipital region. These results might suggest more developed neural circuits involved in objects recognition (Grill-Spector, 2003; Martin and Chao, 2001) in subjects with more frequent generation of unique responses.

Some methodological concerns might remain regarding possible confounding factors and the use of the unmodulated images: effects of possible confounding factors including gender, age and total gray matter volume were minimized using a multiple regression analysis. Recent VBM studies have been divided over the choice of whether modulated or unmodulated images should be used for analysis (Eckert et al., 2006; Gong et al., 2005; Hulshoff Pol et al., 2006; Omura et al., 2005). We used unmodulated images in the present analysis, since we were primarily interested in focal structures including the amygdala (Eckert et al., 2006), though a similar analysis using modulated images also showed significant positive correlations with URR in the left and right amygdala ( $P < .05$ , small volume correction using FDR) when the smoothing kernel was reduced to 8 mm to increase sensitivity to local anatomical differences, demonstrating the robustness of our results independent of the methods used.

The stimuli used in the present study have also been used in a clinical test (Exner, 2003; Rorschach, 1921), and previous reports have shown that unique or original responses are observed at higher frequency in the artistic than in the nonartistic population in this test (Ramachandra, 1994; Rorschach, 1921; Satterlee, 2006). On the other hand, unique responses that cannot easily be recognized by others are also known to appear more frequently in the psychotic population, and the ratio of unique responses to total responses per subject has been thought to represent deviancy in perception or psychotic tendency (Exner, 2003; Ganellen, 1996; Hilsenroth et al., 1998; Jorgensen et al., 2000). In addition, this ratio is also known to correlate with other indices of unusual perception based on questionnaire methods (Perry et al., 2003).

Our findings of a positive correlation between amygdalar volume and unusualness of perception is thus consistent with those of previous studies showing amygdalar enlargement in temporal epilepsy patients with psychosis (Tebartz van Elst

et al., 2002) and in patients with affective psychoses (Velakoulis et al., 2006). However, it should be noted here that only unique perception exhibited by normal subjects was addressed in the present study. Different from our findings, previous studies of schizophrenia patients have reported volume deficits in multiple cerebral regions including the amygdala (Wright et al., 1999; Honea et al., 2005), though disease-specific abnormal neurodevelopmental or neurodegenerative processes possibly due to genetic deficits have been suggested to be causes of such structural abnormalities (Ross et al., 2006).

Since the present study involved only normal subjects, the variation in amygdalar volume shown here might reflect individual differences in terms of perception within normal range rather than pathological traits. Our results and those of previous reports on certain types of psychoses together suggest that the enlargements of the amygdala and the cingulate gyri correlated with unusual perception might reflect frequent recruitment of emotion-related circuits (Maguire et al., 2000), which might be involved in generation of psychotic symptoms in the psychoses and might also contribute to generation of unique perception in the normal population.

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#### Appendix

Examples of typical responses and unique responses for one stimulus are presented below. Numbers in the parentheses designate location of the responses (see Fig. 1). If the response was produced when the stimuli was rotated by 180 degrees or 90 degrees, the mark “r” or “s” is added to the number, respectively.

##### Typical responses

A human (5), a ribbon (4), the upper part of an insect (2r), fire (3), a butterfly (4), a bow tie (4), a monster (9r).

##### Unique responses

A palace with torches (1r), underwear (1), a car spewing gas (front view) (2), a crown (2r), a flamingo (3), a duck wearing a bow tie (6), a squirrel (7), a clown’s face (8), a cat’s face (8), the head of a stag beetle (9r), a bird (10), a dog’s face (11), a cat with swords in both hands (12r), crab (12r), a monkey reaching for a branch and a bird at the edge of water (12s), bean paste soup in a china bowl (13), a mushroom cloud (14r), a bird reflecting on the surface of water

(15s), a bat (16), a human's face (17), a birthday cake (18), a human's lower body (19r), a plastic bottle (20r).

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