Gray Matter Volume Abnormalities in Depressive Patients With and Without Anxiety Disorders

Haochen Qi, BE, Yuping Ning, MD, Jie Li, MD, Shengwen Guo, DE, Minyue Chi, BE, Minjian Gao, BE, Yangbo Guo, MD, Yuling Yang, MD, Hongjun Peng, MD, and Kai Wu, MD

Abstract: Comorbidity with anxiety disorder is a relatively common occurrence in major depressive disorder. However, the unique and shared neuroanatomical characteristics of depression and anxiety disorders have not been fully identified. The aim of this study was to identify gray matter abnormalities and their clinical correlates in depressive patients with and without anxiety disorders.

We applied voxel-based morphometry and region-of-interest analyses of gray matter volume (GMV) in normal controls (NC group, n = 28), depressive patients without anxiety disorder (DP group, n = 18), and depressive patients with anxiety disorder (DPA group, n = 20). The correlations between regional GMV and clinical data were analyzed.

The DP group showed decreased GMV in the left insula (INS) and left triangular part of the inferior frontal gyrus when compared to the NC group. The DPA group showed greater GMV in the midbrain, medial prefrontal cortex, and primary motor/somatosensory cortex when compared to the NC group. Moreover, the DPA group showed greater GMV than the DP group in the frontal, INS, and temporal lobes. Most gray matter anomalies were significantly correlated with depression severity or anxiety symptoms. These correlations were categorized into 4 trend models, of which 3 trend models (ie, Models I, II, and IV) revealed the direction of the correlation between regional GMV and depression severity to be the opposite of that between regional GMV and anxiety symptoms. Importantly, the left INS showed a trend Model I, which might be critically important for distinguishing depressive patients with and without anxiety disorder.

Our findings of gray matter abnormalities, their correlations with clinical data, and the trend models showing opposite direction may reflect disorder-specific symptom characteristics and help explain the neurobiological differences between depression and anxiety disorder.

(Medicine 93(29):e345)

INTRODUCTION

A nxious depression is a common, clinically distinct subtype of major depressive disorder (MDD). It has been estimated that 40% to 50% of patients with MDD have at least 1 comorbid anxiety disorder. Twin studies have revealed a similar genetic risk and inheritance pattern for MDD and anxiety disorders. Additionally, family studies have indicated a cosegregation of MDD and anxiety disorders. Given that depression and anxiety respond to the same treatment strategies, they might also have a similar etiology. Many researchers have speculated that a common mechanism may underlie the development of anxious depression because high trait anxiety/neuroticism is a vulnerability factor for MDD and subjects with MDD exhibit enhanced fear conditioning. However, the onset of MDD is often preceded by the development of anxiety disorders, in both children and adults. The comorbid condition of depression and anxiety may differ from MDD in clinical course and characteristics because it has been associated both with worse outcomes and with more severe psychopathology. Thus, MDD patients with and without anxiety disorders may represent either distinguishable components of the depression spectrum or result...
from distinct combinations of genetic factors that contribute to both disorders.\textsuperscript{1,12} A large number of magnetic resonance imaging (MRI) studies have identified structural brain changes associated with MDD and anxiety. A previous selective review of T1-weighted structural MRI in adult samples of patients with MDD has demonstrated that volumetric reductions of the hippocampus (HIP), basal ganglia, and orbitofrontal and prefrontal cortex are consistently found in patients with MDD.\textsuperscript{13} More recently, a meta-analysis of studies applying voxel-based morphometry (VBM) to MDD indicated that gray matter is significantly reduced in a confined cluster located in the rostral anterior cingulate cortex, dorsolateral prefrontal cortex, and dorsomedial prefrontal cortex (DMPFC).\textsuperscript{12} Therefore, although decreased volumes are found in many brain structures, previous findings are not completely consistent.\textsuperscript{12,13} Conversely, adult patients with generalized anxiety disorder (GAD) show larger gray matter volume (GMV) in brain regions associated with anticipatory anxiety and emotion regulation, such as the amygdala (AMYG) and DMPFC.\textsuperscript{14} MDD patients with anxiety symptoms show greater GMV in the right temporal cortex when compared with MDD patients without anxiety symptoms.\textsuperscript{15} Specific involvement of the inferior frontal cortex in MDD and lateral temporal cortex in anxiety disorders without comorbid MDD has been demonstrated and may reflect disorder-specific symptom clusters.\textsuperscript{16} However, the unique and shared neuroanatomical characteristics of depression and anxiety have not been fully identified. In this study, we applied a VBM analysis of GMV in normal controls (NC) and depressive patients with and without anxiety disorder. Moreover, we analyzed the correlations between clinical characteristics and regional GMV in brain regions showing significant GMV differences in the group VBM comparisons.

MATERIALS AND METHODS

Subjects

Thirty-eight depressive patients between the ages of 18 and 45 years were recruited from the inpatient and outpatient units at Guangzhou Psychiatric Hospital, Affiliated Hospital of Guangzhou Medical University, Guangdong, China. The structured clinical interview for DSM-IV diagnostic criteria was used to assess the presence or absence of MDD. A 17-item Hamilton Depressive Rating Scale (HAM-D) and Zung Self-Rating Depression Scale (SDS) were used to evaluate depression severity.\textsuperscript{17–19} A 14-item Hamilton Anxiety Scale (HAMA) and Self-Rating Anxiety Scale (SAS) were used to evaluate anxiety symptoms.\textsuperscript{20,21} We used Chinese versions of these measurements that show good reliability and validity.\textsuperscript{22} With the exception of anxiety disorder, patients were excluded from the study if they met any of the following criteria: having other psychiatric axis-I or axis-II disorders, another neurological disorder, any substance use within the past 6 months, electroconvulsive therapy, any type of contraindication for MRI, or any other clinically relevant abnormalities in their medical history or laboratory examinations. Therefore, 38 depressive patients were divided into 2 groups: 18 depressive patients without anxiety disorder (the DP group, SAS scores <40, HAMA scores <14) and 20 depressive patients with anxiety disorder (the DPA group, SAS scores ≥40, HAMA scores ≥14).

In addition, 28 sex and age-matched NC (the NC group) were recruited from the local community. Before enrollment, all subjects were fully informed of the details of the study and written informed consent was obtained. These studies were performed according to the Declaration of Helsinki and approved by the Guangzhou Psychiatric Hospital Ethics Committee.

Image Acquisition

Imaging data were acquired using Philips 3T MR systems (Philips, Best, The Netherlands) located at Guangzhou Psychiatric Hospital, Affiliated Hospital of Guangzhou Medical University. For each subject, an anatomical image was obtained using a sagittal 3-D gradient-echo T1-weighted sequence (TR = 7.6 ms, TED = 3.7 ms, TI = 795 ms, flip angle = 8°, 180 slices, slice thickness = 1 mm, gap = 0 mm, matrix = 256 × 256, and inversion time = 0).

Image Processing

All T1-weighted magnetic resonance (MR) images were analyzed using SPMS (Wellcome Institute of Neurology, University College London, UK; http://www.fil.ion.ucl.ac.uk/spm). First, the “New Segmentation” algorithm from SPMS was applied to each T1-weighted MR image to extract tissue maps corresponding to gray matter, white matter, and cerebrospinal fluid. Next, all segmented tissue maps were used to create a customized, population-specific template using the DARTELL template-creation tool.\textsuperscript{23} At the end of the process, gray matter map of each subject was warped using its corresponding smooth, reversible deformation parameters to the custom template space, and then to the MNI standard space. All warped gray matter images were then modulated by calculating the Jacobian determinants derived from the special normalization step and multiplying each voxel by the relative change in volume.\textsuperscript{24} Finally, all wrapped modulated gray matter images were smoothed with an 8-mm Gaussian kernel before voxelwise group comparisons.

Statistical Analysis

One-way analysis of variance (ANOVA) was used to test the group differences in clinical and demographic characteristics of all subjects using SPSS20.0 software. Post hoc pairwise comparisons were then performed using t tests. The gender data were analyzed using the $\chi^2$ test. A value of $P < 0.05$ was considered significant.

Smoothed modulated gray matter images were analyzed with SPM8 utilizing the framework of general linear model. Voxel-wise GMV differences among the 3 groups were investigated using the ANOVA model. Post hoc pairwise comparisons were used to compare differences in GMV between any 2 groups. The covariates included in the model were total gray matter volume (TGMV) calculated from modulated gray matter images, age, gender, and education years. The resulting statistical map was corrected for multiple comparisons to a significance level of $P < 0.05$ by combining individual voxels ($P < 0.001$) and using a cluster size of 173 voxels. This correction was confined within a whole brain mask and determined by Monte Carlo simulations using the AFNI AlphaSim program (http://afni.nih.gov/afni/docpdf/AlphaSim.pdf).

Moreover, we performed a region-of-interest (ROI) analysis of regional gray matter volume (RGMV) in which the ROIs were defined as the significant brain areas found in the voxel-wise analysis of GMV. The partial correlation between the clinical data and the RGMV of each ROI was calculated, controlling for the age, gender, education years, and TGMV.
TABLE 1. Statistical Analysis of Clinical and Demographic Characteristics Among Normal Controls and Depressive Patients With and Without Anxiety Disorders

|                | NC (n = 28) | DP (n = 18) | DPA (n = 20) | F Value (χ²) | P Value |
|----------------|-------------|-------------|--------------|--------------|---------|
| Age, y         | 28.61 ± 5.45| 31.06 ± 7.39| 28.65 ± 8.18| 0.812        | 0.449   |
| Education, y   | 16.25 ± 4.04| 12.56 ± 3.48| 11.55 ± 3.38| 10.826       | <0.0001<sup>a,b</sup> |
| Gender (F/M)   | 13/15       | 11/7        | 9/11         | 1.232        | 0.540   |
| SDS            | 27.43 ± 5.85| 51.67 ± 5.88| 51.45 ± 9.19| 94.855       | <0.0001<sup>a,b</sup> |
| HAMD           | 13.50 ± 1.64| 22.28 ± 3.30| 20.25 ± 3.61| 62.047       | <0.0001<sup>a,b</sup> |
| SAS            | 27.46 ± 5.19| 27.67 ± 4.67| 47.10 ± 5.26| 103.495      | <0.0001<sup>b,c</sup> |
| HAMA           | 8.93 ± 1.98 | 8.11 ± 1.88 | 16.80 ± 3.52| 74.077       | <0.0001<sup>b,c</sup> |

DP = depressive patients without anxiety disorder, DPA = depressive patients with anxiety disorder, HAMA = Hamilton Anxiety Scale, HAMD = Hamilton Depression Rating Scale, NC = normal controls, SAS = Self-Rating Anxiety Scale, SDS = Zung Self-Rating Depression Scale. Values are showed as the mean ± standard deviation. The comparisons of clinical and demographic characteristics among the 3 groups (NC, DP, and DPA) were performed using a separate one-way ANOVA. Post hoc pairwise comparisons were then performed using t test. Statistical significance was set at P < 0.05. "For the gender distribution among the 3 groups, P value was obtained using χ² test." "Post hoc paired comparisons showed significant group differences between NC versus DP." "Post hoc paired comparisons showed significant group differences between DP versus DPA." "Post hoc paired comparisons showed significant group differences between NC versus DPA." "Post hoc paired comparisons showed significant group differences between DP versus DPA."  

TABLE 2. Gray Matter Volume Abnormalities Among Normal Controls, Depressive Patients Without Anxiety, and Depressive Patients With Anxiety

| Anatomical Region           | Hemisphere | Cluster Size (Voxels) | MNI Coordinates, mm |
|-----------------------------|------------|-----------------------|---------------------|
|                             |            |                       | x   | y   | z   | t Value |
| DP < NC Insula              | L          | 280                   | −38 | 17  | 15  | 3.327   |
| Precentral gyrus L          | 1845       | −44                   | −14 | 59  | 4.526 |
| Midbrain                    | L          | 596                   | −6  | −33 | 12  | 4.41    |
| Superior occipital gyrus    | L          | 581                   | 0   | −8  | −18 | 4.449   |
| Orbitofrontal cortex (medial) | L       | 276                   | −2  | 66  | −6  | 3.821   |
| Rolandic operculum          | L          | 224                   | −48 | −20 | 18  | 3.766   |
| Precentral gyrus R          | 525        | 27                    | −24 | 71  | 4.508 |
| Precentral gyrus R          | 283        | −6                    | 50  | 3.792 |
| Postcentral gyrus L         | 245        | 14                    | −45 | 77  | 3.855 |
| DP > NC Insula              | L          | 5362                  | −26 | 23  | −6  | 4.592   |
| Inferior temporal gyrus     | L          | 1987                  | −33 | −2  | −29 | 4.381   |
| Precentral gyrus L          | 1307       | −56                   | −26 | 56  | 4.261 |
| Rectal gyrus                | L          | 611                   | 0   | 60  | −18 | 4.118   |
| Superior frontal gyrus (dorsal) | L       | 361                   | −17 | 57  | 14  | 3.899   |
| Insula                      | L          | 6581                  | 36  | 35  | −5  | 5.214   |
| Lingual gyrus               | R          | 3045                  | 14  | −87 | −17 | 4.654   |
| Parahippocampal gyrus       | R          | 2708                  | 35  | 0   | −30 | 4.570   |
| Middle temporal gyrus       | R          | 970                   | 63  | 6   | −26 | 4.124   |
| Inferior temporal gyrus     | R          | 590                   | 56  | −48 | −11 | 4.079   |
| Rectal gyrus                | R          | 499                   | 8   | 35  | −32 | 4.404   |
| Supplementary motor area    | R          | 326                   | 11  | 12  | 54  | 3.800   |
| Angular gyrus               | R          | 244                   | 45  | −53 | 23  | 4.122   |

DP = depressive patients without anxiety disorder, DPA = depressive patients with anxiety disorder, L = left, NC = normal control, R = right. The resulting statistical map was corrected for multiple comparisons to a significance level of P < 0.05 by combining individual voxels (P < 0.001) and using a cluster size of 173 voxels. This correction was confined within a whole brain mask and was determined by Monte Carlo simulations using the AFNI AlphaSim program (http://afni.nimh.nih.gov/afni/docpdf/AlphaSim.pdf).
The volumes of extracted brain areas and the partial correlations were calculated by the in-house scripts using MATLAB 2010b.

RESULTS

Clinical and Demographic Characteristics of the Subjects

The demographic characteristics of the subjects are shown in Table 1. The age and gender distribution were not different between the 3 groups (P > 0.05). However, the education years in the NC group were significantly higher than those in the DP and DPA groups (P < 0.0001). The clinical data for each group are also shown in Table 1. The ANOVAs demonstrated significant group effects in all the test scores (P < 0.0001). Specifically, the NC group showed significantly lower scores of SDS and HAMD when compared with the DP and DPA groups. Moreover, the DPA group showed significantly higher scores of SAS and HAMA compared with the NC and DP groups.

GMV Abnormalities

The DP group showed significant gray matter reductions in the left insula (INS) and the left triangular part of the inferior frontal gyrus (IFGtriang), when compared with the NC group (Table 2, Figure 1).

The DPA group showed significant gray matter increases relative to the NC group in the following brain areas: bilateral precentral gyrus (PreCG), bilateral midbrain, left superior occipital gyrus, left medial part of orbitofrontal cortex, left rolandic operculum (ROL), and right postcentral gyrus (PoCG) (Table 2, Figure 2).

The DPA group showed significant gray matter increases when compared with the DP group in the following brain areas: bilateral INS, bilateral inferior frontal gyrus (IFG), bilateral rectus (REC), bilateral ROL, bilateral inferior temporal gyrus (ITG), bilateral middle temporal gyrus (MTG), bilateral parahippocampal gyrus (PHG), bilateral HIP, left PreCG, left dorsal part of superior frontal gyrus (SFGdor), right lingual gyrus (LING), right PoCG, right supplementary motor area, right angular gyrus, and right AMYG (Table 2, Figure 3).

Correlation Between Clinical Data and GMV

We calculated the RGMV of 23 ROIs, which showed significant group differences in the voxel-wise analysis of GMV (as shown in Table 2). Significant partial correlations between the clinical data (including SDS, HAMD, SAS, and HAMA) and the RGMV of ROIs were found, controlling for age, sex, education years, and TGMV (as shown in Table 3). Most ROIs (17 of 23 ROIs) indicated significant correlations (P < 0.05) between the RGMV and at least 1 of the clinical evaluation metrics. A significant negative correlation was found only in ROI-3 (the left ITG) and ROI-9 (the right PHG) between the RGMV and HAMD values; all other significant correlations were positive.

Furthermore, we defined 4 trend models (I–IV, as shown in Table 3) according to the correlations between depression severity (SDS or HAMD) and RGMV and those between anxiety symptoms (SAS or HAMA) and RGMV (Figure 4).
patients with anxiety disorder, INS depression patients without anxiety disorder, DPA a cluster size of 173 voxels. All coordinates are in MNI space. DP

Correlation between anxiety symptoms and RGMV. Only the correlation between depression severity and RGMV and no significant trend model IV indicated a significant negative correlation between anxiety symptoms and RGMV and no significant trend model III, such as the left REC (Figure 4C). Finally, depression severity. Four brain regions were classified into correlation between RGMV and both anxiety symptoms and the left INS (Figure 4A). Trend model II indicated a significant positive correlation between anxiety symptoms and RGMV. (Figure 4B). Trend model III indicated a significant positive correlation between RGMV and both anxiety symptoms and depression severity. Four brain regions were classified into trend model III, such as the left REC (Figure 4C). Finally, trend model IV indicated a significant negative correlation between depression severity and RGMV and no significant correlation between anxiety symptoms and RGMV. Only the right PHG showed a pattern consistent with trend model IV (Figure 4D).

DISCUSSION

Our results indicated that the DP group had lower GMV in the left INS and left IFGtriang than the NC group. These results were consistent with several previous studies, which found that patients with MDD had significantly decreased GMV in the left INS\textsuperscript{25,26} and in the bilateral INS,\textsuperscript{27,28} when compared with healthy controls. However, many previous structural MRI studies have indicated that several brain regions (eg, anterior cingulate cortex, HIP, AMYG, and medial prefrontal cortex) are involved in the emotional and cognitive impairments in MDD.\textsuperscript{29–31} Dysfunction of these brain structures, in addition to the INS, forms the basis of the limbic-cortical theory of the pathogenesis of MDD.\textsuperscript{32,33} The INS is located deep within the lateral sulcus and has been included in the limbic lobe because of its intimate connections with the cingulate, AMYG, and orbitofrontal cortex.\textsuperscript{14} Theoretically, the INS is neuroanatomically well positioned to represent emotional experience because it receives interoceptive inputs from the whole body, and its connections with the prefrontal regions can provide contextual information.\textsuperscript{35} The inferior prefrontal cortex plays a major role in the orbitofrontal circuit, which allows the integration of limbic and emotional information into behavioral responses.\textsuperscript{36–39} Therefore, abnormalities in the INS and inferior prefrontal cortex might cause changes in social behavior and emotional experiences. Moreover, a previous combined functional and structural MRI study found that patients with MDD show GMV abnormalities in several parts of IFG and that the structural changes result in functional alterations within the emotional circuit.\textsuperscript{40}

Interestingly, the DPA group showed increased GMV in a variety of cortical regions, compared to both the NC and DP groups. When compared with the NC group, the DPA group showed GMV increases primarily within areas of the midbrain, medial prefrontal cortex, and primary motor/somatosensory cortex. However, the DPA group showed greater GMV than the DP group principally within regions of the frontal, INS, and temporal lobes. Our results were consistent with previous findings that increased GMV in specific brain structures is associated with anxiety disorders.\textsuperscript{14,15,41,42} For example, among patients with MDD, those with anxiety display greater GMV than those without anxiety in the right temporal cortex extending from the mid-posterior superior temporal gyrus into the posterior middle and ITG.\textsuperscript{14} Moreover, our results may support the valence-arousal model, which states that depression correlates with decreased activity in the right parietotemporal brain regions associated with arousal properties, whereas anxiety correlates with increased activity.\textsuperscript{43} For example, patients with GAD show increased GMV in brain regions (eg, dorsomedial prefrontal cortex) associated with anticipatory anxiety and emotion regulation.\textsuperscript{14} In addition, a previous task-related functional MRI study observed frontal and limbic hypoactivation (eg, INS and medial frontal cortex) in patients with depression and comorbid anxiety.\textsuperscript{44} Thus, we speculated that abnormalities in these brain structures or the connections between them might result in pathological anxiety, which extensive fear conditioning research has suggested may arise from abnormal interactions between cortical and subcortical regions.\textsuperscript{45–47}

We also found significant correlations between gray matter anomalies and depression severity or anxiety symptoms. More importantly, significant correlations can be defined as 4 trend models. Three trend models (Models I, II, and IV; 13 of 17 ROIs) revealed an opposite direction of association between regional brain volumes and either depression or anxiety scores; the values of RGMV were decreased with depression severity but increased with anxiety symptoms. These results further support the findings from our VBM analysis, which showed an opposite direction of RGMV abnormalities for the DP and DPA groups when compared with the NC group. Anxiety disorders are marked by excessive fear (and avoidance), often in response to specific objects or situations and in the absence of

**FIGURE 3.** Gray matter volume increases in depression patients with anxiety disorder compared with depression patients without anxiety disorder. Depression patients with anxiety disorder showed significant gray matter increases in bilateral INS, bilateral inferior frontal gyrus, bilateral rectus, bilateral Rolandic operculum, bilateral inferior temporal gyrus, bilateral middle temporal gyrus, bilateral parahippocampal gyrus, bilateral hippocampus, left precentral gyrus, left dorsal part of superior frontal gyrus, right lingual gyrus, right postcentral gyrus, right supplementary motor area, right angular gyrus, and right amygdala. The resulting statistical map was corrected for multiple comparisons to a significance level of $P < 0.05$ by combining individual voxels ($P < 0.001$) and using a cluster size of 173 voxels. All coordinates are in MNI space. DP = depression patients without anxiety disorder, DPA = depression patients with anxiety disorder, INS = insula.
true danger.\textsuperscript{47} However, MDD is a prevalent mental health concern and is associated with significant disability and suffering.\textsuperscript{48,49} Thus, our trend model findings (Models I, II, and IV) may help to explain the neurobiological differences between depressive patients with and without anxiety. Importantly, the left INS cortex showed a trend Model I with a strong positive correlation between the RGMV and HAMA. The INS cortex is known to play a key role in modulating subjective feeling states and interpersonal awareness.\textsuperscript{50} Moreover, the INS is known to play a key role in modulating subjective feeling states and interpersonal awareness.\textsuperscript{50} The partial correlation between the clinical data and the RGMV of each ROI was calculated, controlling for the age, gender, education years, and TGMV. The partial correlation was set at P < 0.05 (shown in bold).

| ID of ROIs | Anatomical Region | Hemisphere | Cluster Size (Voxels) | SDS | HAMD | SAS | HAMA | Trend model |
|-----------|-------------------|------------|----------------------|-----|------|-----|------|-------------|
| 1 Insula  | L                 | 280        | n.s. n.s. 0.285* 0.382** | I   |
| 2 Insula  | L                 | 5362       | n.s. n.s. 0.311* | II  |
| 3 Inferior temporal gyrus | L | 1987 | n.s. 0.290* 0.303* 0.250* | III |
| 4 Precentral gyrus | L | 1307 | n.s. 0.330** 0.279* | I   |
| 5 Rectal gyrus | L | 611  | 0.272* 0.397** 0.301* 0.342** | III |
| 6 Superior frontal gyrus (dorsal) | L | 361  | 0.290* 0.295* 0.303* 0.250* | III |
| 7 Insula  | R                 | 6581       | n.s. n.s. 0.330** 0.334** | I   |
| 8 Lingual gyrus | R | 3045  | n.s. 0.296* 0.346* | I   |
| 9 Parahippocampal gyrus | R | 2708  | n.s. 0.322* n.s. n.s. | IV  |
| 10 Middle temporal gyrus | R | 970  | n.s. n.s. 0.260* n.s. | I   |
| 11 Inferior temporal gyrus | R | 590  | n.s. n.s. 0.364* | I   |
| 12 Rectal gyrus | R | 499  | n.s. n.s. n.s. | n.s. |
| 13 Supplementary motor area | R | 326  | n.s. n.s. n.s. | n.s. |
| 14 Angular gyrus | R | 244  | n.s. n.s. 0.369** | I   |
| 15 Precentral gyrus L | 1845 | n.s. n.s. 0.266* | n.s. I |
| 16 Midbrain | L | 596  | n.s. n.s. n.s. | n.s. |
| 17 Midbrain | L | 581  | n.s. n.s. 0.322* 0.327* | I   |
| 18 Superior occipital gyrus | L | 350  | 0.310* n.s. 0.281* 0.264* | III |
| 19 Orbitofrontal cortex (medial) | L | 276  | 0.398** 0.543** 0.323* 0.345** | III |
| 20 Rolandic operculum | L | 224  | n.s. n.s. 0.268* n.s. | I   |
| 21 Precentral gyrus | R | 525  | n.s. n.s. n.s. | n.s. |
| 22 Precentral gyrus | R | 283  | n.s. n.s. n.s. | n.s. |
| 23 Postcentral gyrus | R | 245  | n.s. n.s. n.s. | n.s. |

MDD.\textsuperscript{52} To better explore the neuroanatomical characteristics of depression and anxiety disorders, it would be useful to recruit drug-naive, first-episode patients of depression and anxiety disorders in the future study. Second, the results of our VBM analyses were inconsistent with many previous studies. However, comparing data across studies is difficult because of variations in study design, assessment, and the use of inconsistent definitions to diagnose anxious depression.\textsuperscript{1} Therefore, the inconsistent results between the current and previous studies might be attributed to several factors: the VBM analyses with relatively small sample sizes in this study might have insufficient statistical power.\textsuperscript{53} Third, we defined 4 trend models with different patterns of correlation between RGMV and depression severity or anxiety symptoms. Although the analysis of trend models was not quantitative, further studies could apply a data-driven technique, such as multivariate pattern analysis, to discriminate psychiatric patients from healthy controls.\textsuperscript{54–57}

CONCLUSION

In this study, we demonstrated that depressive patients with and without anxiety disorder showed gray matter abnormalities in a variety of brain structures and that the abnormalities...
of some brain structures were significantly correlated with clinical data. Based on the correlations with either HAMD or HAMA, 4 trend models were defined and might reflect disorder-specific symptom characteristics. The existence of distinct neuroanatomical profiles associated with depressive patients with or without anxiety disorder may provide a potential biomarker for disease diagnosis.

REFERENCES

1. Ionescu DF, Niciu MJ, Mathews DC, et al. Neurobiology of anxious depression: a review. *Depress Anxiety*. 2013;30:374–385.

2. Silberg J, Rutter M, Neale M, et al. Genetic moderation of environmental risk for depression and anxiety in adolescent girls. *Br J Psychiatry*. 2001;179:116–121.

3. Williamson DE, Forbes EE, Dahl RE, et al. A genetic epidemiologic perspective on comorbidity of depression and anxiety. *Child Adolesc Psychiatr Clin N Am*. 2005;14:707–726.

4. Ressler KJ, Mayberg HS. Targeting abnormal neural circuits in mood and anxiety disorders: from the laboratory to the clinic. *Nat Neurosci*. 2007;10:1116–1124.

5. Sandi C, Richter-Levin G. From high anxiety trait to depression: a neurocognitive hypothesis. *Trends Neurosci*. 2009;32:312–320.

6. Nissen C, Holz J, Blechert J, et al. Learning as a model for neural plasticity in major depression. *Biol Psychiatry*. 2010;68:544–552.

7. Gorman JM. Comorbid depression and anxiety spectrum disorders. *Depress Anxiety*. 1996;4:160–168.

8. Gorman JM, Coplan JD. Comorbidity of depression and panic disorder. *J Clin Psychiatry*. 1996;57(Suppl 10):34–41.

9. Roy-Byrne PP, Stang P, Wittchen HU, et al. Lifetime panic-depression comorbidity in the National Comorbidity Survey. Association with symptoms, impairment, course and help-seeking. *Br J Psychiatry*. 2000;176:229–235.

**FIGURE 4.** Partial correlation between the clinical data and the RGMV of selected ROIs was calculated, controlling for the age, gender, education years, and TGMV. (A) ROI-2: left insula. (B) ROI-3: left inferior temporal gyrus. (C) ROI-5: left rectal gyrus. (D) ROI-9: right parahippocampal gyrus. The solid lines indicate significant correlations with $P < 0.05$. The dashed lines indicate insignificant correlations. HAMD = Hamilton Depressive Rating Scale, HAMA = Hamilton Anxiety Scale, RGMV = regional gray matter volume, ROI = region of interest, TGMV = total gray matter volume.
10. Rush AJ, Zimmerman M, Wisniewski SR, et al. Comorbid psychiatric disorders in depressed outpatients: demographic and clinical features. *J Affect Disord*. 2005;87:43–55.

11. Kessler RC, Chiu WT, Demler O, et al. Prevalence, severity, and comorbidity of 12-month DSM-IV disorders in the National Comorbidity Survey Replication. *Arch Gen Psychiatry*. 2005;62:617–627.

12. Bora E, Fornito A, Pantelis C, et al. Gray matter abnormalities in major depressive disorder: a meta-analysis of voxel based morphometry studies. *J Affect Disord*. 2012;138:9–18.

13. Lorenzetti V, Allen NB, Fornito A, et al. Structural brain abnormalities in major depressive disorder: a selective review of recent MRI studies. *J Affect Disord*. 2009;117:1–17.

14. Schienle A, Ebner F, Schafer A. Localized gray matter volume abnormalities in generalized anxiety disorder. *Eur Arch Psychiatry Clin Neurosci*. 2011;261:303–307.

15. Inkster B, Rao AW, Ridler K, et al. Structural brain changes in patients with recurrent major depressive disorder presenting with anxiety symptoms. *J Neuroimag* 21. 2011:375–382.

16. van Tol MJ, van der Wee NJ, van den Heuvel OA, et al. Regional brain volume in depression and anxiety disorders. *Arch General Psychiatry*. 2010;67:1002–1011.

17. Hamilton M. A rating scale for depression. *J Neurol Neurosurg Psychiatry*. 1960;23:56–62.

18. Zung WWK. A self-rating depression scale. *Arch General Psychiatry*. 1965;12:63–70.

19. Zung WWK, Richards CB, Short MJ. Self-rating depression scale in an outpatient clinic. Further validation of the SDS. *Arch General Psychiatry*. 1965;13:508–515.

20. Hamilton M. The assessment of anxiety by rating scale. *Brit J Med Psychol*. 1959;50:55.

21. Zung WWK. A rating instrument for anxiety disorders. *Psychosomatics*. 1971;371–379.

22. Zhang MY. Handbook of Psychiatric Measures. Changsha, Hunan, China: Hunan Science and Technology Press; 1995.

23. Ashburner J. A fast diffeomorphic image registration algorithm. *Neuroimage*. 2007;38:95–113.

24. Good CD, Johnsrude I, Ashburner J, et al. Cerebral asymmetry and the effects of sex and handedness on brain structure: a voxel-based morphometric analysis of 465 normal adult human brains. *NeuroImage*. 2001;14:685–700.

25. Liu CH, Jing B, Ma X, et al. Voxel-based morphometry study of the insular cortex in female patients with current and remitted depression. *Neuroscience*. 2014;262:190–199.

26. Soriano-Mas C, Hernandez-Ribas R, Pujol J, et al. Cross-sectional and longitudinal assessment of structural brain alterations in melancholic depression. *Biolog Psychiatry*. 2011;69:318–325.

27. Peng J, Liu J, Nie B, et al. Cerebral and cerebellar gray matter reduction in first-episode patients with major depressive disorder: a voxel-based morphometry study. *Eur J Radiol*. 2011;80:395–399.

28. Sprengelmeyer R, Steele JD, Mwangi B, et al. The insular cortex and the neuroanatomy of major depression. *J Affect Disord*. 2011;133:120–127.

29. Abe O, Yamase H, Kasi K, et al. Voxel-based analyses of gray/white matter volume and diffusion tensor data in major depression. *Psychiatry Res*. 2010;181:64–70.

30. Frodl T, Reinhold E, Kourtoulener N, et al. Interaction of childhood stress with hippocampus and prefrontal cortex volume reduction in major depression. *J Psychiatr Res*. 2010;44:799–807.

31. Koolschathe PC, van Haren NE, Lensvelt-Mulders GJ, et al. Brain volume abnormalities in major depressive disorder: a meta-analysis of magnetic resonance imaging studies. *Hum Brain Map*. 2009;30:3719–3735.

32. Mayberg HS. Modulating dysfunctional limbic-cortical circuits in depression: towards development of brain-based algorithms for diagnosis and optimised treatment. *Brit Med Bull*. 2003;65:193–207.

33. Sheline YI. 3D MRI studies of neuroanatomic changes in unipolar major depression: the role of stress and medical comorbidity. *Biol Psychiatry*. 2000;48:791–800.

34. Paulus MP, Stein MB. An insular view of anxiety. *Biol Psychiatry*. 2006;60:383–387.

35. Suzuki A. Emotional functions of the insula [in Japanese]. *Ann Rev Neurosci*. 1986;9:357–381.

36. Bonelli RM, Cummings JL. Frontal-subcortical circuitry and behavior. *Dialog Clin Neurosci*. 2007;9:141–151.

37. Burruss JW, Hurley RA, Taber KH, et al. Functional neuroanatomy of the frontal lobe circuits. *Radiology*. 2000;214:227–230.

38. Tekin S, Cummings JL. Frontal-subcortical neuronal circuits and clinical neuropsychiatry: an update. *J Psychiatr Res*. 2002;53:647–654.

39. Scheuerlcker J, Meisenzahl EM, Koutsouleris N, et al. Orbitofrontal volume reductions during emotion recognition in patients with major depression. *J Psychiatry Neurosci*. 2010;35:311–320.

40. Benetti S, McCrory E, Arulanantham S, et al. Attachment style, affective loss and gray matter volume: a voxel-based morphometry study. *Hum Brain Map*. 2010;31:1482–1489.

41. Liao M, Yang F, Zhang Y, et al. Childhood maltreatment is associated with larger left thalamic gray matter volume in adolescents with generalized anxiety disorder. *PloS One*. 2013;8:e71898.

42. Heller W, Etienne MA, Miller GA. Patterns of perceptual asymmetry in depression and anxiety: implications for neuropsychological models of emotion and psychopathology. *J Abnorm Psychol*. 1995;104:327–333.

43. Schlund MW, Verduzzo G, Cataldo MF, et al. Generalized anxiety modulates frontal and limbic activation in major depression. *Behav Brain Funct*. 2012;8:8.

44. Boatman JA, Kim JJ. A thalamo-cortico-amygdala pathway mediates auditory fear conditioning in the intact brain. *Eur J Neurosci*. 2006;24:894–900.

45. Cannistraro PA, Rauch SL. Neural circuitry of anxiety: evidence from structural and functional neuroimaging studies. *Psychopharmacol Rev*. 2007;31:169–191.

46. Shin LM, Lieberson I. The neurocircuitry of fear, stress, and anxiety disorders. *Neuropsychopharmacology*. 2010;35:169–191.

47. Hasin DS, Goodwin RD, Stinson FS, et al. Epidemiology of major depressive disorder: results from the National Epidemiologic Survey Replication (NCS-R). *J Am Med Assoc*. 2003;289:3095–3105.

48. Nagai M, Kishi K, Kato S. Insular cortex and neuropsychiatric comorbidity of 12-month DSM-IV disorders in the National Comorbidity Survey Replication (NCS-R). *J Psychiatr Res*. 2009;43:940–949.

49. Kessler RC, Berglund P, Demler O, et al. The epidemiology of major depressive disorder: results from the National Comorbidity Survey Replication (NCS-R). *J Am Med Assoc*. 2003;289:3095–3105.

50. Nagai M, Kishi K, Kato S. Insular cortex and neuropsychiatric disorders: a review of recent literature. *Eur Psychiatry*. 2007;22:387–394.
52. Park SC, Hahn SW, Hwang TY, et al. Does age at onset of first major depressive episode indicate the subtype of major depressive disorder? The clinical research center for depression study. *Yonsei Med J.* 2014;55:1712–1720.

53. Lai CH. Gray matter volume in major depressive disorder: a meta-analysis of voxel-based morphometry studies. *Psychiatry Res.* 2013;211:37–46.

54. Anderson JS, Nielsen JA, Froehlich AL, et al. Functional connectivity magnetic resonance imaging classification of autism. *Brain.* 2011;134 (Pt 12):3742–3754.

55. Dai Z, Yan C, Wang Z, et al. Discriminative analysis of early Alzheimer’s disease using multi-modal imaging and multi-level characterization with multi-classifier (M3). *NeuroImage.* 2012;59:2187–2195.

56. Liu F, Guo W, Yu D, et al. Classification of different therapeutic responses of major depressive disorder with multivariate pattern analysis method based on structural MR scans. *PloS One.* 2012;7:e40968.

57. Uddin LQ, Menon V, Young CB, et al. Multivariate searchlight classification of structural magnetic resonance imaging in children and adolescents with autism. *Biol Psychiatry.* 2011;70:833–841.