ELSEVIER

NeuroImage

www.elsevier.com/locate/ynimg
NeuroImage 30 (2006) 485 – 497

Cortical abnormalities in bipolar disorder investigated with MRI and voxel-based morphometry

Allison C. Nugent,[a,*] Michael P. Milham,[b] Earle E. Bain,[a] Linda Mah,[a] Dara M. Cannon,[a] Sean Marrett,[c] Carlos A. Zarate,[a] Daniel S. Pine,[a] Joseph L. Price,[d] and Wayne C. Drevets[a]

[a]*Section on Neuroimaging in Mood and Anxiety Disorders Program, National Institute of Mental Health, National Institutes of Health, 1 Center Drive, MSC 0135, Bethesda, MD 20892-0135, USA*
[b]*Beckman Institute, University of Illinois, Urbana, IL 61810, USA*
[c]*Functional Neuroimaging Facility, NIMH, NIH, Bethesda, MD 20892, USA*
[d]*Department of Anatomy and Neurobiology, Washington University School of Medicine, St. Louis, MO 63110, USA*

Received 8 April 2005; revised 16 September 2005; accepted 19 September 2005
Available online 26 October 2005

Bipolar disorder (BD) has been associated with abnormalities of brain structure. Specifically, in vivo volumetric MRI and/or post mortem studies of BD have reported abnormalities of gray matter (GM) volume in the medial prefrontal cortex (PFC), amygdala, hippocampal subiculum and ventral striatum. These structures share anatomical connections with each other and form part of a "visceromotor" network modulating emotional behavior. Areas of the lateral orbital, superior temporal and posterior cingulate cortices project to this network, but morphometric abnormalities in these areas have not been established in BD. The current study assessed tissue volumes within these areas in BD using MRI and voxel-based morphometry (VBM).

MRI images were obtained from 36 BD subjects and 65 healthy controls. To account for possible neurotrophic and neuroprotective effects of psychotropic medications, BD subjects were divided into medicated and unmedicated groups. Images were segmented into tissue compartments, which were examined on a voxel-wise basis to determine the location and extent of morphometric changes.

The GM was reduced in the posterior cingulate/retrosplenial cortex and superior temporal gyrus of unmedicated BD subjects relative to medicated BD subjects and in the lateral orbital cortex of medicated BD subjects relative to controls. White matter (WM) was increased in the orbital and posterior cingulate cortices, which most likely reflected alterations in gyral morphology resulting from the reductions in the associated GM.

The morphometric abnormalities in the posterior cingulate, superior temporal and lateral orbital cortices in BD support the hypothesis that the extended network of neuroanatomical structures subserving visceromotor regulation contains structural alterations in BD. Additionally, localization of morphometric abnormalities to areas known to exhibit increased metabolism in depression supports the hypothesis that repeated stress and elevated glucocorticoid secretion may result in neuroplastic changes in BD.

Keywords: Morphometry; Bipolar depression; Magnetic resonance imaging

Introduction

Bipolar disorder (BD) is a severe mental illness affecting approximately 1.3–1.6% of the population worldwide. BD is characterized by the presence of both depressive and manic episodes (Drevets and Todd, 1997). There are two diagnostic subtypes, BDI, in which the manic episodes result in pronounced impairment, and BDII, which is characterized by milder periods of hypomania, where elevated mood and activity are present without debilitation (American Psychiatric Association, 2000).

The etiology and pathophysiology of BD are unknown. Nevertheless, both in vivo neuroimaging and post mortem neuropathological studies have shown that individuals with BD have abnormal reductions in gray matter volume in parts of the medial prefrontal cortex (PFC), amygdala, hippocampal subiculum and ventral striatum (reviewed in Drevets et al., 2004; Strakowski et al., 2005). For example, within the medial PFC, morphometric MRI studies found reductions in gray matter volume in the anterior cingulate cortex (ACC) ventral and anterior to the genu of the corpus callosum (termed "subgenual" and "pregenual", respectively) in BD subjects relative to healthy controls (Drevets et al., 1997; Lochhead et al., 2004; Lyoo et al., 2004; Sassi et al., 2004). The results of these in vivo MRI studies guided post mortem histopathological studies of the subgenual ACC to identify abnormally reductions in neuronal somal size and glial cell counts and increases in neuronal density in BD (Chana et al., 2003), suggesting that the corresponding decrease in gray matter volume

* Corresponding author. Fax: +1 (301) 480 3610.
E-mail address: nugenta@intra.nimh.nih.gov (A.C. Nugent).
Available online on ScienceDirect (www.sciencedirect.com).

1053-8119/$ - see front matter © 2005 Elsevier Inc. All rights reserved.
doi:10.1016/j.neuroimage.2005.09.029

was accounted for by reductions in dendritic arborization (Chana et al., 2003; Drevets, 2004). Additionally, in the pregenual ACC and the Brodmann Area (BA) 9 cortex situated anterior to the ACC (anteromedial and dorsal anterolateral PFC), post mortem studies showed abnormal reductions in neuronal and glial cell density in BD subjects relative to controls (Rajkowska et al., 2001; Todtenkopf et al., 2005).

Neuromorphometric abnormalities also have been identified in BD in basal ganglia and mesiotemporal lobe structures that are anatomically related to these medial PFC structures. In the striatum, a post mortem volumetric study found that the left accumbens area, bilateral external pallidum and right putamen were smaller in subjects with BD than in healthy controls (Baumann et al., 1999). In the hippocampal subiculum, BD samples showed abnormal reductions in the density of apical dendritic spines on pyramidal cells (Rosoklija et al., 2000), the concentrations of mRNA for synaptic proteins post mortem (Eastwood and Harrison, 2000) and the gray matter volume (Nugent et al., 2004), although the entire hippocampal volume generally has not been found to differ between BD and control samples. Finally, several neuroimaging studies reported that the amygdala volume is abnormal in BD, although disagreement exists regarding the direction of these differences with respect to healthy controls (reviewed in Drevets, 2004).

The regions implicated by these studies share extensive, monosynaptic, neuronal connections with each other and with the hypothalamus and periaqueductal gray to form a circuit that plays major roles in modulating emotional behavior (Ongur and Price, 2000). This circuit has been characterized as a "visceromotor network" based upon evidence that its projections function to modulate neuroendocrine, autonomic and behavioral responses to threatening, stressful and reward-related stimuli and contexts (Ongur and Price, 2000). Because BD is associated with disturbances in these domains of emotional behavior, the structural abnormalities found within this visceromotor circuit in BD conceivably could play an integral role in this condition's pathogenesis.

Anatomical studies in humans demonstrate that the medial PFC, ventral striatum, subiculum and amygdala regions involved in visceromotor function also share extensive, reciprocal, monosynaptic connections with areas of the orbitofrontal, posterior cingulate and superior temporal cortices (Ongur and Price, 2000; Kondo et al., 2004; Saleem et al., 2004; Price et al., 2004). These anatomical observations converge with neurophysiological data from human brain mapping studies of various emotional states (see Discussion) to suggest that specific orbitofrontal, posterior cingulate and superior temporal areas also may play roles in modulating emotional behavior. Whether morphometric abnormalities also exist in these regions in BD remains unclear, however.

Few post mortem neuropathological studies have assessed orbitofrontal, posterior cingulate or superior temporal cortices in BD, and preliminary results from morphometric MRI studies of BD either await replication or are in disagreement. In the ventral PFC, one voxel-based morphometry (VBM) study reported a reduction in GM density in the lateral orbital cortex situated on the inferior frontal gyrus (Lyoo et al., 2004), while another found a GM reduction located several centimeters away at the ventromedial pole of the superior frontal gyrus (Wilke et al., 2004). In the posterior cingulate cortex (PCC), one VBM study reported widespread deficits in gray matter "density" throughout the cingulate gyrus, including the PCC (Doris et al., 2004), of "poor outcome" BD, although this finding awaits replication and extension to BD subjects who are not poor outcome. Moreover, it remains unclear whether this difference in GM density is associated with a corresponding reduction in GM volume. In the superior temporal gyrus (STG), a post mortem histopathological study observed reduced glial cell density and reduced clustering of neurons into minicolumns in the planum temporale region in BD (Beasley et al., 2005). However, most MRI studies that employed manual segmentation of the entire STG found no significant volumetric differences between BD and control samples (Hirayasu et al., 2000; Schlaepfer et al., 1994; Brambilla et al., 2003; Kasai et al., 2003), although one reported abnormally decreased STG volume in children and adolescents with BD (Chen et al., 2004) and another found abnormally increased right anterior STG volume in adults with BD (Pearlson et al., 1997). These conflicting results regarding STG volume may reflect limitations in the sensitivity for detecting focally distributed differences between BD and control samples using neuromorphometric methods that sample an entire gyrus (see Discussion).

These conflicting results across studies also may reflect differences between the subject samples with respect to medication status or other clinical variables. In recent years, the mood stabilizing drug lithium was found to exert neurotrophic and neuroprotective effects in rodents and humans (Manji et al., 1999), and lithium treatment has been associated with increases in GM in longitudinal studies of BD (Moore et al., 2000 a,b). Other agents used for mood stabilization, such as valproic acid preparations, also exert neurotrophic and neuroprotective effects in rodents (Hao et al., 2004). Subjects with BD also are likely to receive antidepressant drugs, which have been associated with neurotrophic effects (Duman, 2004), and antipsychotic agents, which in some cases increase the GM volume of basal ganglia structures (Gur et al., 1998; Corson et al., 1999). The importance of taking such effects into account in neuromorphometric analyses has only recently been suggested by Sassi et al. (2004) and Drevets (2004), who found abnormal volumetric reductions in the ACC only in untreated BD subjects.

The current study is the first VBM study to assess morphometric abnormalities present in BD samples differentiated on the basis of recent medication status. Because the VBM approach is particularly useful for surveying large regions of cortex and revealing inherent differences in tissue volume between groups, this study specifically focused upon orbital and medial prefrontal cortical, PCC and STG regions that constitute the major cortical components of the visceromotor network.

Methods

Subjects

Thirty-six subjects (26 female, age = 39 ± 8.1) meeting Diagnostic and Statistical Manual of Mental Disorders-Version IV (DSM-IV) criteria for Bipolar Disorder Type I (n = 7) or II (n = 29) were imaged. Sixty-five healthy controls (46 female, age = 38 ± 11.8) with no personal or family history of psychiatric disorders in first-degree relatives also were imaged. Subjects were drawn from a larger pool of 39 BD subjects and 95 controls based upon having images of sufficient signal homogeneity to apply VBM (see Image analysis section). Subjects were between ages 18 and 60 years. Subjects were

excluded if they had major medical illnesses or neurological disorders met DSM-IV criteria for substance abuse within the previous 90 days or substance dependence within the past 5 years or had general exclusions from participating in MRI (e.g., pregnancy, pacemaker, etc.). Written consent was obtained from all subjects as approved by the NIMH Institutional Review Board.

Of the 36 BD subjects, 16 had not received medication within the 4 months prior to scanning, and 20 either were currently medicated or had received medication within 4 months. The medications currently being taken by this latter group (and the number of subject taking each agent) were: lithium (n = 11), divalproex sodium (Depakote), valproic acid or valproate sodium (n = 6), chlorapramazine (n = 1). Two additional subjects were recently medicated (within 4 months), one with lithium and one with wellbutrin and Zoloft. Because the duration of the putative medication-induced neurotrophic effects is unknown, these subjects were included in the medicated group. In order to assess severity of illness, the bipolar type (I or II), age at illness onset and illness duration (current age minus age at onset) were determined.

Image acquisition

MRI scans were acquired on a GE 3T MRI scanner, running an approximately 11 min MP-RAGE pulse sequence optimized for gray matter/white matter contrast (TE = 2.1 ms, TR = 7.8 ms, prep time = 725 ms, flip angle = 6, TD = 1400 ms). Data were collected on one of two identical scanners, so scanner was used as a covariate in the statistical analysis. Prior studies have found that volumetric measurements from MP-RAGE images acquired on different scanners show an intra-class correlation greater that 0.9 (Quinn et al., 2005). Ten BD subjects (5 unmedicated, 5 medicated) and 7 controls were imaged on one scanner, and the remainder on the second scanner. Images were acquired as a 224 × 224 × 124 matrix, with a field-of-view of 22 cm and a slice thickness of 1.2 mm. Images were reconstructed on a 256 × 256 × 124 grid to an image voxel size of 0.85 × 0.85 × 1.2 mm.

Image analysis

Following acquisition, images were corrected for intensity non-uniformity using the minc tool N3 (McConnell Brain Imaging Centre Montreal Neurological Institute). Next, images were edited to remove non-brain material using the FSL "Brain Extraction Tool" (FMRIB, Oxford, UK) followed by manual editing using MEDx (Medical Numerics, Sterling, VA). Images were then segmented into binary gray matter (GM), white matter (WM) and CSF maps using the FSL tool FAST. Because the CSF surrounding the brain surface was removed prior to segmentation along with other non-brain matter, analysis of the CSF maps only provided assessment of the CSF spaces inside ventricles and sulci. Due to reduced contrast at the edges of the field-of-view, segmentations often failed at the dorsum of the brain, which would confound a VBM technique. Therefore, following a binary segmentation, all GM images were visually inspected for segmentation quality and graded on a one to four scale by raters blind to diagnosis. Segmentations resulting in a uniform GM ribbon on all slices were rated as "1", those with mild distortions of the GM ribbon on fewer than one-half of the slices were rated "2", and GM maps showing zeroed voxels ("holes") within the GM ribbon or areas of moderate GM thickening/thinning were rated "3". Images with moderate segmentation distortions (anatomically impossible thickening or thinning of GM) on >50% of slices, or interruptions of the GM ribbon, were rated "4". The vast majority of segmentation problems were confined to the vertex of the brain (near the edge of the field-of-view where GM/WM contrast was sometimes reduced) or the basal ganglia and thalamus, where differences in tissue composition across structures resulted in ambiguous differentiation of GM and WM. Because none of these regions were included within our a priori regions-of-interest, only images classified with the poorest rating 4 were excluded from analysis.

Among 4 raters trained on the method, reliability estimate for any single image rating was 0.91. Images for 13 BD subjects and 30 controls were excluded from analysis. For images included, the average grade for segmentation quality did not differ between the medicated BD, unmedicated BD and control groups. Additionally, the average grade was not significantly different across scanner.

Whole brain volumes were calculated by adding the GM and WM segments. The images were then re-segmented using the partial volume estimate option within FAST. This consists of an initial segmentation estimate followed by a further Markov random field estimate to increase the segmentation accuracy (Zhang et al., 2001). Segmentation was carried out on the images at their original spatial resolution.

VBM analysis was performed using SPM2 (Wellcome Department of Imaging Neuroscience, University College London, London, UK). Because our sample included a greater proportion of females than the average population, the optimized VBM procedure included generation of a local template (Good et al., 2001), in which images were registered to a standard template, resampled to the resolution of the standard template (2 × 2 × 2 mm), averaged across subjects and smoothed with an 8 mm Gaussian kernel. Following generation of the local template, the VBM procedure was applied by spatially normalizing each subject's original (higher) resolution tissue maps to the local template. Because previous studies showed that affine normalization is more sensitive and specific for detecting cortical abnormalities than less constrained nonlinear normalization routines (Wilke et al., 2003a), we chose to use an affine only normalization implemented in SPM2. Images were resampled to 2 × 2 × 2 mm (the resolution of the template) using trilinear interpolation following normalization. Normalized mages were smoothed using a Gaussian smoothing kernel with full-width at half-maximum = 12 mm. While smaller smoothing kernels may result in more sensitive detection of small volumetric abnormalities (Wilke et al., 2003a), the use of a 12 mm smoothing kernel assured that the data were sufficiently normal to justify the application of Gaussian random field theory in the statistical calculations (Salmond et al., 2002a).

Statistical analysis

Based upon the literature outlined in the introduction, a priori regions-of-interest (ROI) encompassed the cingulate, orbital and medial prefrontal and superior temporal cortices. The orbitomedial PFC ROI was delimited in coronal slices of the local template between the anterior- and posterior-most planes in which the medial orbital gyrus could be delimited from the lateral orbital gyri by the orbital sulcus. This ROI

Table 1
Demographic information

	Healthy controls			Bipolar disorder: n = 36 medicated			Unmedicated		
	M + F: n = 65	M: n = 19	F: n = 46	M + F: n = 20	M: n = 5	F: n = 15	M + F: n = 16	M: n = 5	F: n = 11
Age years (SD)	38 (11.8)	41 (11.0)	37 (11.9)	41 (8.3)	45 (7.0)	40 (7.8)	37 (7.5)	41 (6.5)	35 (7.5)
Age of onset years (SD)	–	–	–	18 (8.8)	18 (7.8)	18 (10.6)	21 (6.5)	20 (5.1)	21 (7.2)
Duration of illness years (SD)	–	–	–	23 (9.0)	27 (8.5)	22 (10.2)	17 (10.0)	21 (5.8)	15 (11.1)

extended laterally to include the lateral orbital and inferior frontal gyri and dorsally to include the anterior cingulate gyrus. The superior temporal ROI encompassed the STG in all coronal slices in which that gyrus was evident. The dorsal and posterior cingulate ROI were outlined on axial slices as the PCC (BA 23 26 27 29 30 31), situated on the posterior cingulate gyrus and the precuneate gyrus, anterior to the parietal–occipital sulcus. The three ROI were drawn on the local GM and WM templates prior to smoothing. The ROI volumes were 18.86 mL and 16.98 mL for the right and left STG, respectively; 47.68 mL and 47.85 mL for the right and left orbitomedial PFC, respectively; and 37.14 mL and 35.77 mL for the right and left dorsal and posterior cingulate regions, respectively.

Using SPM2, group differences in the GM and WM maps were compared between the control, medicated BD and unmedicated BD groups using an AnCova model with age, gender and scanner as covariates. The groups were compared pair-wise using t test contrasts within SPM2. Significant differences in volume in the areas encompassed within the a priori ROI were indicated either by a cluster of >25 voxels for which the voxel t values corresponded to $P < 0.001$ or by a cluster of any size that contained a voxel t value that remained significant after correction for multiple comparisons using the "small volume correction" option within SPM2 (Worsley et al., 1996).

To ensure that the differences identified within the ROI were not accounted for by larger differences which had their peak effect sizes located outside the ROI, whole brain VBM analyses were performed post hoc. These post hoc analyses also permitted exploration of possible differences in the remainder of the cortex. To reduce the possibility of Type I error, differences found in these analyses (i.e., outside the primary ROI defined a priori) were reported as significant only if they had $P < 0.05$ after correction for multiple comparisons over the whole brain volume (Worsley et al., 1996).

Results

Demographic and clinical information about the subject samples is presented in Table 1. The mean age and gender composition did not differ significantly between groups. All 36 of the BD subjects met criteria for a current major depressive episode at the time of scanning, and all had experienced multiple episodes (>2) of hypomania or mania in the past. Three unmedicated BD subjects and four recently medicated BD subjects were classified as Type I, and the remaining subjects were Type II. The difference in the age at illness onset did not reach statistical significance between the two BD samples (t = 1.06). The recently medicated BD subjects had a longer illness duration than the unmedicated BD subjects (P = 0.038).

Whole brain volumes for subjects whose images contained the entire cerebrum and cerebellum were 1125 ± 80.1, 1117 ± 123.8 and 1135 ± 103.3 mL, for unmedicated BD, medicated BD and healthy controls, respectively, did not differ significantly across groups (F = 0.214). Of the 101 images, 11 did not include either the ventral cerebellum or the vertex, so they were excluded from whole brain volume measures. These 11 images were included in the VBM analysis, however, because

Table 2
Significant differences in GM volume between the unmedicated bipolar disordered (BD), medicated BD and healthy control (HC) samples

GM volume				
Group contrast	BD medicated > BD unmedicated	HC > BD medicated	HC > BD unmedicated	
Region	Left posterior cingulate	Left orbital cortex	Left posterior cingulate	Left superior temporal gyrus
WB peak voxel	−10 −58 10	−42 27 −5	−10 −56 8	−46 −48 19
Extent (voxels)	63	92	16	28
t	4.13	3.88	3.77	3.47
SVC peak voxel	−10 −58 10	−42 27 −5	−10 −56 8	−46 −46 17
Extent (voxels)	43	92	14	4
t	4.13	3.88	3.77	
P corrected	0.017	0.033	0.049	0.065

Regional differences in GM volume were reported as significant if they contained 25 voxels at $P < 0.001$ uncorrected (peak voxel listed under whole brain (WB) peak voxel) or had a peak voxel with P(corrected) < 0.05 after corrections for multiple comparison using the small volume correction (SVC) within SPM2. Coordinates locate the voxel containing the highest ("peak") t value within the stereotaxic array of Talaraich and Tournoux (1988) and indicate distance in millimeters from the anterior commissure, with positive x = right, positive y = anterior and positive z = superior to a plane containing both the anterior and the posterior commissures. "Extent" refers to the number of voxels within the cluster of voxels which had $P < 0.001$. The differences in GM volume were identified within the ROI defined a priori as none of the results from the post hoc whole brain analysis (WB) remained significant after correction for multiple comparisons. Degrees of freedom = 95.

the primary regions-of-interest were entirely within the field-of-view in all subjects. Dependence of GM volume on the covariates was examined. Age, gender and, to a lesser degree, scanner accounted for a large portion of the variance and were thus retained in the statistical model. A correlation of total GM and age was significant at $P < 0.0001$, with $R^2 = 0.332$, and total GM significantly differed between the male and female subjects ($P < 0.0001$). Total GM volume was not significantly different between the two scanners ($t = 1.0$). The profound dependence on age and gender is consistent with previously published results (Good et al., 2001; Sato et al., 2003; Giedd, 2004).

The VBM gray matter results are shown in Table 2. In the left PCC, an area was identified where GM in the unmedicated BD subjects was reduced relative both to the medicated BD subjects and the healthy controls (P(corrected) = 0.049, $t = 3.77$). In the left ventral PFC, GM was reduced in the medicated BD subjects relative to the healthy controls in the orbital cortex situated on the left inferior frontal gyrus (P(corrected) = 0.033, $t = 3.88$). Finally, in the left STG, unmedicated BD subjects showed decreased GM relative to healthy controls (based upon the criterion of having a cluster of >25 voxels for which P(uncorrected) < 0.001). Figs. 1a–c illustrate the areas of reduced GM superimposed on the local GM template.

WM results appear in Table 3. In the left ventral PFC, WM was increased in the medicated BD subjects relative to the healthy controls in the orbital cortex situated on the left inferior frontal gyrus (P(corrected) = 0.045, $t = 3.64$). In the left PCC, unmedicated BD subjects exhibited an area of increased WM relative to the medicated BD subjects (based again on the P(uncorrected) < 0.001 criteria). Notably, these areas of increased WM volume in the orbital cortex and PCC were situated adjacent to the corresponding GM decreases in the same group contrasts (Table 2). Finally, an area of increased WM volume was apparent

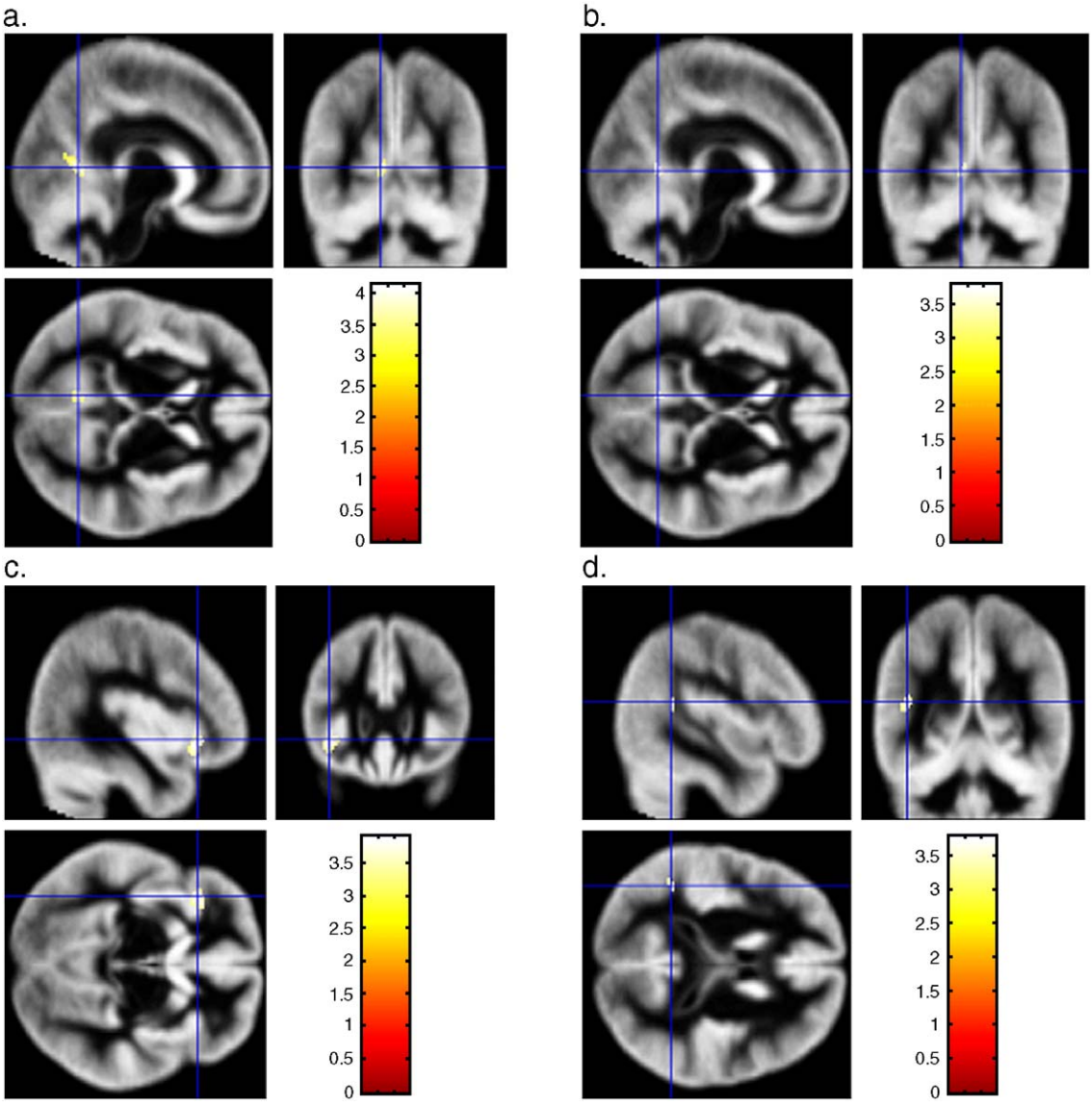

Fig. 1. Regions of decreased GM shown on the GM template in the left posterior cingulate cortex in (a) unmedicated BD subjects as compared to medicated BD subjects and (b) unmedicated BD subjects as compared to healthy control subjects; and regions of decreased GM in (c) left orbital cortex in medicated BD subjects as compared to healthy controls and in (d) left superior temporal gyrus in unmedicated BD subjects as compared to healthy controls.

Table 3
Significant differences in WM volume between the BD and control groups

WM volume			
Group contrast	BD unmedicated > BD medicated	BD medicated > HC	BD unmedicated > HC
Region	Left posterior cingulate	Left orbital cortex	Left posterior cingulate/medial parietal cortex interface
WB peak voxel	−10 −67 20	−36 29 −3	−14 −38 50
Extent (voxels)	60	39	39
t	3.5	3.64	3.67
SVC peak voxel	−10 −67 20	−36 29 −3	−14 −36 48
Extent (voxels)	60	38	9
t	3.5	3.64	3.44
P corrected	0.092	0.045	0.107

The criteria for assigning statistical significance and the interpretation of the coordinates are as in Table 2.

in the left medial parietal cortex lying along the PCC in the unmedicated BD subjects relative to the healthy controls based on the same criteria. No significant differences in WM between groups were found in the STG. Figs. 2a–c show areas of reduced WM on the local WM template.

The CSF maps showed no significant differences between groups.

The post hoc whole brain VBM analyses established that the differences identified within the primary ROI were not accounted for by larger differences which had their peak effect sizes located outside those ROI as the peak voxel locations identified by the a priori small volume ROI analyses and the post hoc whole brain analyses were virtually identical in all cases (Tables 2, 3). No differences were identified between groups which remained significant after applying conservative corrections for multiple comparisons over the whole brain. However, to guide hypotheses for future studies and comparisons between our results and those of previous studies, the cortical GM regions where differences approached significance (based upon having a cluster of >25 voxels for which $P < 0.005$) are listed in Table 4 and illustrated in maximum intensity projections for the medicated BD versus unmedicated BD, control versus medicated BD and control versus unmedicated BD contrasts (Figs. 3a–c; clusters found within basal ganglia and thalamic regions were not listed in Table 4 for the reasons listed in the Image analysis section above).

Discussion

The VBM analysis showed left lateralized reductions in GM in BD patients relative to controls in the PCC, lateral orbital cortex and STG. In the PCC and orbital cortex, local increases in WM were identified in the vicinity of the areas where GM was reduced. The reciprocal pattern of differences between the GM and WM tissue compartments within the PCC and orbital cortex most likely reflects differences in gyral morphology between BD and healthy brains that would be expected to result from the GM reductions in BD.

This interpretation of the WM differences as reflecting secondary changes in gyral morphology was followed from the observations that the reductions in GM identified herein were situated inside sulci. In such locations, the relatively small CSF spaces that would form in association with the subtle reductions in GM arising in BD (the magnitudes of abnormal cortex GM reduction in BD have ranged from 8% to 38%; Drevets et al., 1997; Nugent et al., 2004) would presumably be obliterated as gravity pulled down the malleable, adjacent, remaining brain tissue into the ex vacuo CSF space. As the remaining thinned GM displaces toward the sulcus to fill this space, the corresponding WM would be pulled along into the position normally occupied by GM, resulting in an apparent increase in local WM in the corresponding VBM images. This physical phenomenon likely accounts for why increases in WM, but not in CSF, appear in the vicinity of the GM reductions (Tables 1, 2). For example, the GM images in Fig. 1 exhibit no visible CSF within the sulci of the GM image (an effect that also partly results from the modest blurring of the MRI images, the anatomical variability across subjects and the relatively subtle magnitude of GM volume reductions in BD).

Alternative explanations for the reciprocal pattern of GM and WM changes in BD also may conceivably exist, however. An apparent increase in one tissue type occurring simultaneously with a decrease in another may indicate the systematic misclassification of voxels in the structure due to changes in the NMR signal. For example, the neuropathological changes associated with multiple sclerosis (Parry et al., 2003) and ischemia (Calamante et al., 1999) have been shown to alter NMR relaxation times in a manner that changes the T_1 measure from the corresponding tissue.

Methodological strengths and limitations influencing comparisons with previous studies

Two limitations of the VBM technique merit comment. First, the computation of the VBM statistical parametric map requires many thousands of independent statistical comparisons, increasing the likelihood of Type I error. The determination of significance in VBM studies must, therefore, protect against Type I error by applying appropriate corrections, analogous to those developed for PET and functional MRI that employ either the Gaussian random field theory or the cluster test approaches. In the case of VBM, cluster-corrected *P* values are not strictly valid as VBM data are not spatially smooth; large clusters will appear in spatially smooth regions, while small clusters will appear in small or irregular regions. As clusters are therefore biased to extreme values, *P* values for clusters are inexact (Ashburner and Friston, 2000). Voxel-wise corrections, however, remain valid but may be overly conservative (Wilke et al., 2003a). To preserve sensitivity while circumventing the use of cluster tests, we set criteria similar to but more stringent than those previously applied in the VBM literature (Wilke et al., 2001, 2003b; Sowell et al., 1999). As a result, however, only areas where volumetric differences across groups

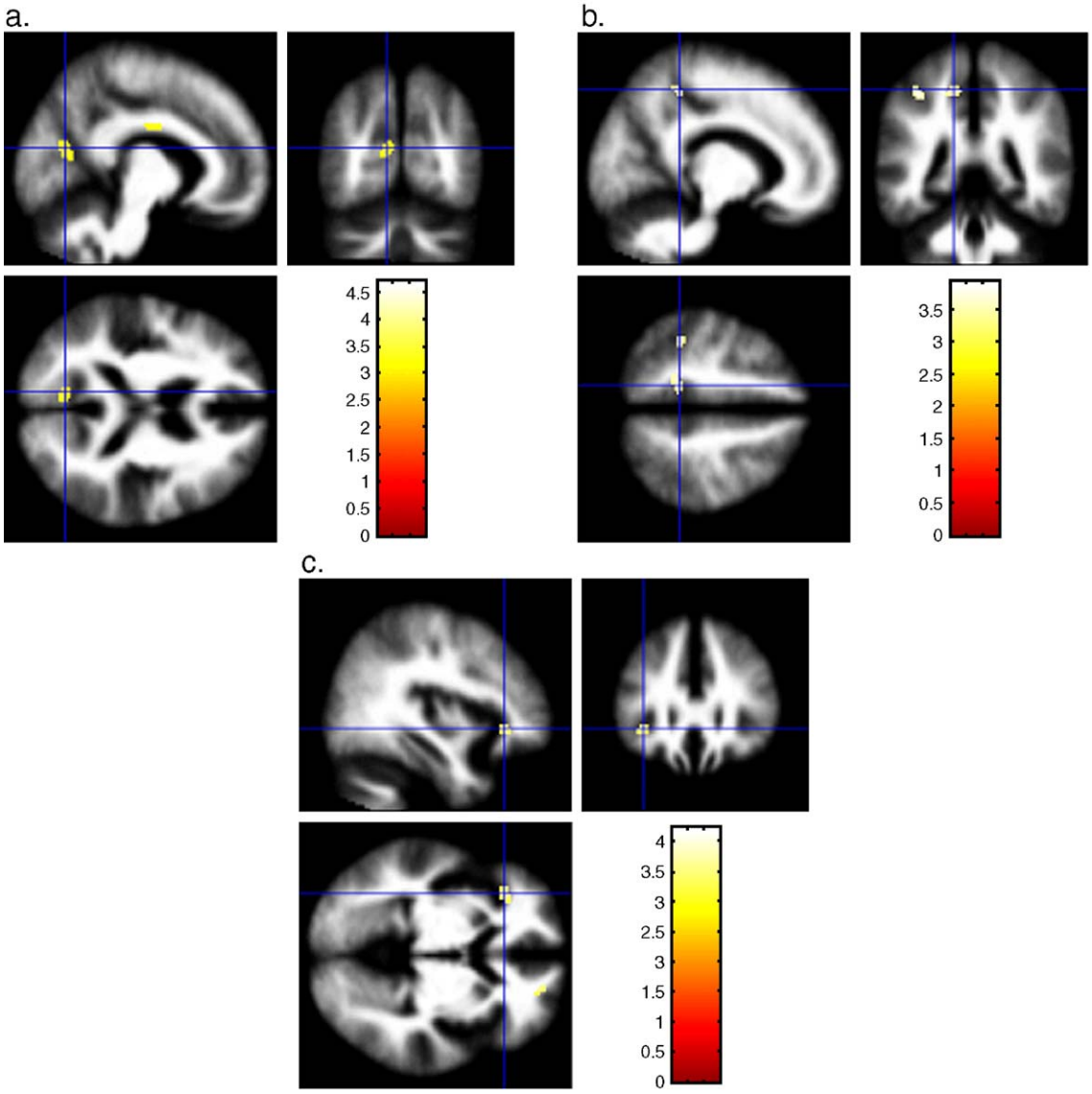

Fig. 2. Regions of increased WM shown on the WM template in the left posterior cingulate cortex in unmedicated BD subjects as compared to medicated BD subjects (a) and in unmedicated BD subjects as compared to healthy controls (b) and in the left orbital cortex of medicated BD subjects as compared to healthy controls (c).

reached very large effect sizes were detected as significant, yielding substantial Type II error rates in comparisons involving the relatively small subject samples studied herein. Moreover, regions in the vicinity of the basal ganglia and thalamus were not assessed in the current study because the limited tissue contrast resolution between GM and WM in these structures renders the tissue segmentations unreliable. The current study thus had very low power for detecting abnormalities previously reported in BD in the amygdala, subiculum, accumbens area and subgenual and pregenual ACC.

Second, the sensitivity for detecting differences in these relatively small regions, which had been shown to contain morphometric MRI abnormalities in BD by studies applying manual segmentation techniques, was further reduced by the dependence of VBM upon spatial normalization of the primary images. Because substantial variability normally exists across humans in the relative size and positioning of structures within the brain, techniques that rely upon spatial normalization are subject to misalignment error. This error particularly reduces the sensitivity of VBM for detecting volumetric abnormalities in small structures such as the amygdala, subiculum, accumbens area and subgenual ACC because they cannot be precisely aligned across subjects using extant spatial normalization algorithms.

The post hoc assessments at less conservative significance thresholds (Table 4, Fig. 3) partly addressed these limitations in sensitivity and the likelihood of Type II error. These data contained replications of the reductions in GM found in the perigenual anterior cingulate cortex in unmedicated BD subjects relative to healthy controls (Drevets et al., 1997; Lochhead et al., 2004; Lyoo et al., 2004; Sassi et al., 2004) and in unmedicated BD subjects relative to medicated BD subjects (Drevets et al., 1997; Lochhead et al., 2004; Lyoo et al., 2004; Sassi et al., 2004). These results also implicated additional areas of the orbitofrontal cortex, PCC and STG in BD. Most notably, these data identified an area of the left

Table 4
Cortical regions where reductions in GM volume approached significance based upon containing a cluster of >25 voxels with $P < 0.005$ uncorrected

Region	*t*	*P*(uncorrected)	Coordinates (*x y z*)
NC > unmedicated BD			
R perigenual anterior cingulate/frontal polar C	3.45	<0.001	20 51 5
R dorsal temporal polar c of the STG	3.26	0.001	59 7 −5
R posterior cingulate c	3.24	0.001	24 −33 2
L posterior cingulate c	3.20	0.001	−16 −31 5
L parieto occipital c	3.06	0.001	−42 −72 31
R inferior parietal c	3.02	0.002	34 −25 42
L perigenual anterior cingulate/frontal polar c	3.00	0.002	−20 52 1
L lateral orbital c	2.98	0.002	−40 52 −14
L inferior parietal c	2.88	0.002	−40 −31 38
NC > medicated BD			
R dorsolateral prefrontal c	3.45	<0.001	22 35 30
R ventrolateral prefrontal c	3.31	0.001	42 53 16
R frontal polar c	3.18	0.001	20 52 −8
L sensorimotor c	3.10	0.001	−22 −20 56
L sensorimotor c	3.00	0.002	−32 −10 41
R sensorimotor c	2.85	0.003	22 −20 58
Medicated BD > unmedicated BD			
R perigenual anterior cingulate/frontal polar c	3.25	0.001	18 51 5
L parieto occipital c	3.11	0.001	−42 −74 29
L lateral orbital c	3.08	0.001	−36 52 −16
L perigenual anterior cingulate c	2.98	0.002	−6 33 8

Although the data shown were from the post hoc whole brain analysis (see Fig. 3), several of these differences also were located within the primary ROI. Abbreviations, c—cortex, L—left, R—right, STG superior temporal gyrus. FEW-corrected *P* values are reported, although these corrections are probably excessively conservative. The degrees of freedom was 95.

lateral orbital cortex where GM was reduced in the unmedicated BD subjects relative both to the healthy controls and the medicated BD subjects and implicated the dorsal temporopolar area of the STG and the frontal polar area of the ventral PFC that specifically have been shown to participate in the visceromotor network (Ongur and Price, 2000).

A methodological strength of our approach was that the VBM technique is more sensitive than manual segmentation for detecting focal morphometric abnormalities in large regions of cortex which may contain affected areas that cannot yet be reliably delimited a priori by clearly evident anatomical landmarks. This capability is particularly advantageous for investigations of cortical abnormalities in BD, in which volumetric abnormalities appear to be focally rather than globally distributed (as would be expected for an illness that involves specific neural circuits rather than entire gyri) (Drevets, 2004). This enhanced sensitivity for detecting focally distributed reductions in GM may account for the differences between our findings and those of studies which detected no difference in STG volume in BD (Hirayasu et al., 2000; Schlaepfer et al., 1994; Brambilla et al., 2003; Kasai et al., 2003). These latter studies had limited sensitivity for detecting differences between BD and control samples because they relied upon an STG ROI that encompassed the entire STG, which would have diluted the effect of the relatively focal abnormalities identified herein (Tables 2, 4) on the volumetric measure by including tissues where volume did not differ between BD and control samples.

A particularly critical difference between our study and previous studies that may account for discrepancies in the results across studies was the selection of a BD group who had not been medicated recently. To our knowledge, ours is the only VBM study to address the recent discovery that some drugs to which BD subjects are commonly exposed (especially mood stabilizers) exert potent neurotrophic effects by assessing a BD sample that was not treated recently. Because these neurotrophic effects appear to induce neuromorphometric changes in humans with BD (Drevets et al., 1997; Lochhead et al., 2004; Lyoo et al., 2004; Sassi et al., 2004), our study sets a new standard for VBM analyses of BD. For example, some studies of predominantly medicated BD subjects reported abnormal increases in GM volume in BD which conceivably may have been produced by medication-induced neurotrophic effects (e.g., Drevets et al., 1997; Lochhead et al., 2004; Lyoo et al., 2004; Sassi et al., 2004; Adler et al., 2005).

Our findings that both the medicated BD subjects and the healthy controls had significantly more GM than the unmedicated BD subjects in the PCC provide additional evidence that medications shown to have neurotrophic effects in rodents can influence cerebral volumes in humans with BD. These data also appear compatible with the hypothesis that mood stabilizing agents exert protective or restorative effects on the neuropathological changes associated with untreated BD (Drevets et al., 1997; Lochhead et al., 2004; Lyoo et al., 2004; Sassi et al., 2004). Although the medication status of the BD subject samples may have been confounded by selection bias as more severely ill subjects presumably would have been more likely to receive treatment (for example, the medicated subjects had a significantly longer illness duration than the unmedicated subjects), this clinical difference would presumably bias the current results toward finding a greater reduction in GM in the medicated sample. An additional methodological limitation is the inclusion of subjects with a remote history of substance abuse as the long-term effects of substance abuse on brain volume are poorly understood.

An example of this latter case may be evident in the left lateral orbital C, where an area of reduced GM was found in the medicated BD sample relative to the HC sample, but where GM did not significantly differ between the unmedicated BD sample and either the HC or the medicated BD samples. Notably, Adler et al. (2005) reported that GM in this region of the left lateral orbital cortex was smaller in BD cases who had experienced multiple manic episodes compared to those with only a single episode, suggesting the possibility that repeated illness resulted in progressive GM loss. Although all of our cases had suffered multiple episodes, the longer duration of bipolar illness in the medicated sample versus the unmedicated sample (Table 1) appears consistent with the hypothesis that GM decreases progressively in this region in proportion with greater exposure to bipolar illness. Future neuromorphometric studies of BD are needed which can more directly establish effects of mood stabilizing therapies and illness progression in longitudinal studies that serially image BD cases before and following treatment and across multiple illness episodes.

A more difficult issue to address with respect to interpreting differences between our results and those of some other VBM studies relates to potential technical differences in image quality. For many studies, field strength employed was 1.5 T, resulting in

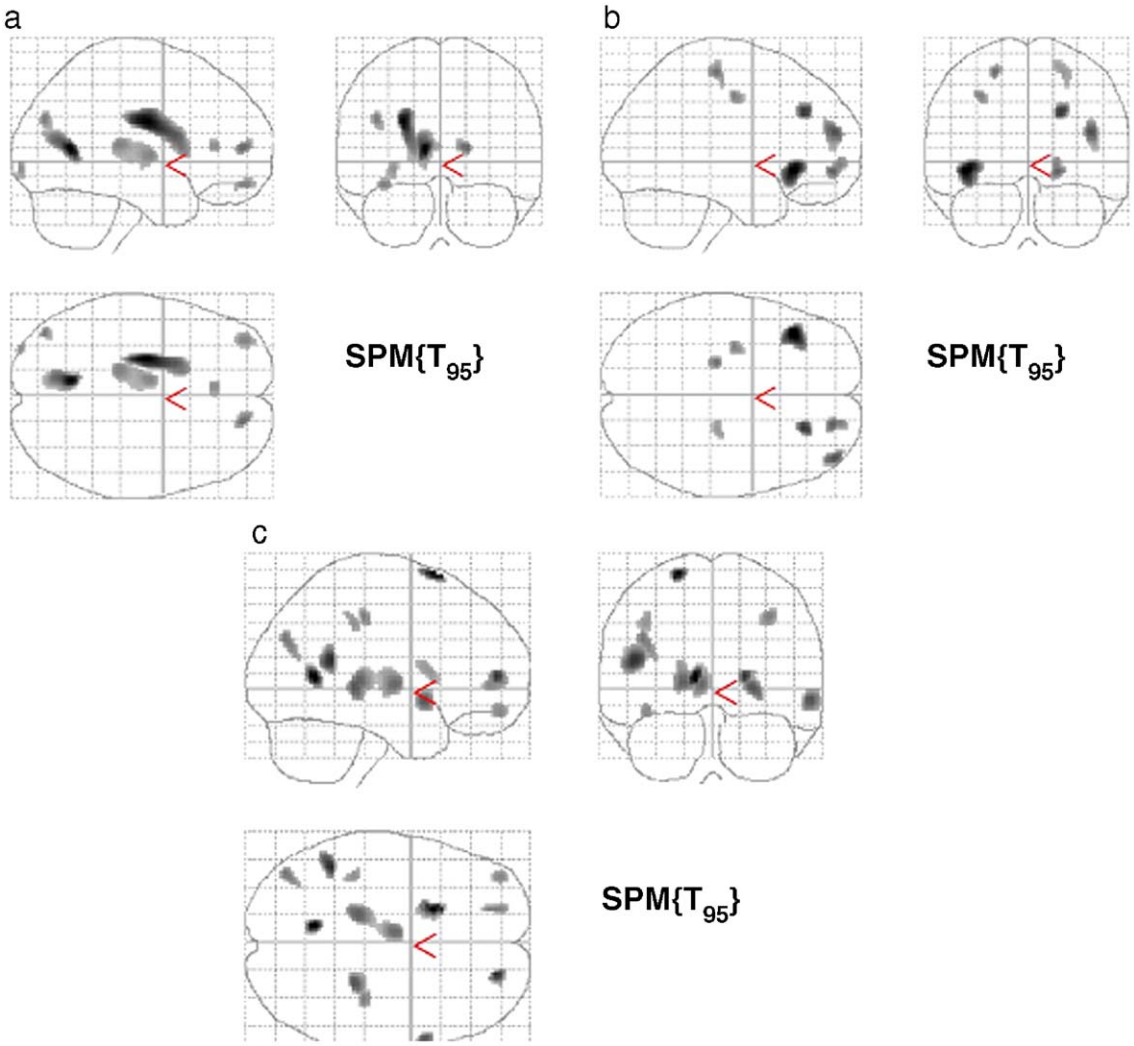

Fig. 3. Maximum intensity projections ("glass brain") of voxel t values computed using SPM2 to show areas where gray matter volume is greater in: (a) the medicated BD relative to the unmedicated BD samples, (b) the healthy control relative to the medicated BD samples and (c) the healthy control versus the unmedicated BD samples. Images are thresholded to show clusters of >25 voxels for which P(uncorrected) < 0.005.

one-half of the NMR signal available from 3 T systems. The increase in resolution made possible by increased signal-to-noise may result in more accurate segmentation of gray and white matter, although scanning at high field also may increase the likelihood of image artifacts. Moreover, our use of an affine normalization technique provided relatively greater validity for investigating cortical abnormalities but was less accurate at spatially normalization subcortical regions (e.g., basal ganglia, thalamus), or mesiotemporal lobe structures (Salmond et al., 2002b), potentially resulting in false negatives in these regions.

Neuroanatomical considerations

Posterior cingulate cortex

In Maddock's review of 51 studies of emotion (Maddock, 1999), the PCC was second only to the inferior PFC as the region most likely to show increases in hemodynamic activity during emotional conditions (Maddock, 1999). For example, in healthy humans, physiological activity increased in the PCC during provocation of anxiety, exposure to pain or other aversive stimuli and elicitation of grief during bereavement (Sewards and Sewards, 2002; Tolle et al., 1999; Gundel et al., 2003; Kimbrell et al., 1999; Maddock et al., 2003). In currently depressed subjects with either BD or major depressive disorder (MDD; "unipolar depression"), the resting blood flow and/or metabolism in the PCC were abnormally increased to an extent that was positively correlated with depression severity and anxiety ratings (Drevets, 2004; Bench et al., 1993; Mah et al., 2004) and negatively correlated with treatment resistance (Kimbrell et al., 2002). Consistent with these data, in currently non-depressed subjects with a history of MDD, the transient depressive relapse induced by tryptophan depletion is associated with increased glucose metabolism in the PCC (Neumeister et al., 2004). Finally, treatment with antidepressant drugs generally resulted in a reduction of CBF or metabolism in the PCC (e.g., Drevets et al., 2002; Buchsbaum et al., 1997), although in one study, the direction of metabolic change differed depending upon treatment response (Mayberg et al., 2000).

The PCC sends extensive efferent connections to the ACC and other components of the visceromotor network (Price et al., 2004; Vogt, 1993). For example, the substantial nature of the projections from the hippocampal subiculum to the PCC (Mesulam, 2000) gave rise to hypotheses that the PCC plays a major role in

mediating interactions between emotion and memory (Maddock et al., 2003). Our finding that GM in the PCC is decreased in unmedicated BD subjects relative to controls is compatible with a previous report in "poor outcome" BD subjects (Doris et al., 2004) but extends this finding to include less severely ill BD subjects.

Lateral orbital cortex

The left lateral orbital cortex region where we found abnormally reduced GM density and volume in medicated BD subjects was located on the ventral inferior frontal gyrus and is part of a region where hemodynamic activity increases during a variety of emotional conditions (Drevets, 2004; Maddock, 1999). The VBM study of Lyoo et al. (2004) reported reduced GM density in the homologous area of the right hemisphere ($x = 39$, $y = 26$, $z = -11$) in a BD sample that contained mostly medicated subjects. In the vicinity of the left lateral orbital cortex region implicated herein, volumetric MRI and post mortem neuropathological studies of MDD have shown that unipolar depressives also have reduced GM (reviewed in Drevets, 2004). Lai et al. also found evidence for reduced orbital frontal cortex volume in elderly depressive subjects (Lai et al., 2000), while Bowen et al. specifically implicated the lateral orbital cortex (as opposed to the medial orbital gyrus) in a group of elderly subjects with mood disorders (Bowen et al., 1989). In addition, the left lateral orbital cortex where we found reduced GM was shown by Ketter et al. (2001) to contain abnormally elevated glucose metabolism in mildly, but not moderately to severely, depressed BD patients compared to healthy controls. These findings were similar to those obtained in PET studies of unipolar depression, where glucose metabolism and CBF in the left lateral orbital cortex/IFG region were increased during the depressed phase to an extent that correlated inversely with depression severity (Drevets, 2004; Neumeister et al., 2004; Drevets et al., 1992, 2002). This inverse relationship between left lateral orbital cortex activity and depression severity is consistent with other types of evidence indicating that this cortex functions to modulate or inhibit emotional expression and experience (reviewed in Drevets, 2004) and that lesions of this cortex increase the risk for developing the major depressive syndrome (MacFall et al., 2001). The loss of GM in this region may thus confer a diathesis toward developing depression in BD.

Superior temporal gyrus

The area of reduced GM density in the STG had not been identified by previous neuromorphometric MRI studies that relied upon manual segmentation of the entire STG (see Introduction), highlighting the sensitivity of VBM for revealing focal differences that could not be specifically localized a priori within a large area of cortex. The specific STG area where GM was reduced in BD was previously implicated as a region where physiological activity changed during bipolar-type mood fluctuations in Parkinson's disease subjects in response to levodopa treatment and during depressive relapse in primary MDD patients in response to tryptophan depletion (Neumeister et al., 2004; Black et al., 2005). For example, the CBF response to levodopa challenge differed between Parkinson's subjects who showed prominent mood changes (mania, depression, anxiety) versus patients who showed only motor changes in the left STG ($x = -52$, $y = -54$, $z = 16$), left lateral orbital cortex (-50, 14, -2) and multiple areas of the PCC and medial PFC (Black et al., 2005). In a more anterior region of the STG ($x = -52$, $y = -18$, $z = 0$), metabolism was shown to correlate with Spielberger State Anxiety Inventory scores in a depressed sample containing 27 BD and 25 MDD subjects (Osuch et al., 2000).

Implications for the pathophysiology of bipolar disorder

The regions where reductions in GM were found between the BD and healthy control samples are conceptually linked by areas where physiological activity increases during emotional processing in healthy humans and where regional glucose metabolism is abnormally elevated during the depressed phase relative to the remitted phase of MDD and BD. In addition, these regions share extensive reciprocal anatomical connections with each other and with other regions reported to have abnormal reductions in GM in BD in the subgenual and pregenual ACC, ventral striatum, hippocampal subiculum and amygdala. Post mortem studies of these latter areas in mood disorders have shown that the histopathological correlates of these volume reductions include loss of synaptic contacts, increased neuronal density and reductions in glial cells and glial markers (especially of oligodendroglia) (Drevets, 2004). These findings at least partly resemble the neuroplastic changes produced in homologous structures of rodents during repeated stress, in which interactions between prolonged glucocorticoid secretion and NMDA receptor stimulation result in dendritic atrophy (McEwen and Chattarji, 2004). Both MDD and BD have been associated with increased glucocorticoid hormone (cortisol) secretion. Moreover, because the glucose metabolic signal measured by PET is dominated by glutamatergic transmission (reviewed in Drevets, 2004), the focal elevation of glucose metabolism in BD suggests that glutamatergic transmission is increased in these projections of the visceromotor circuitry during the depressed phase. The spatial correspondence between the focal elevations in metabolism and the focal reductions in GM in BD suggests the hypothesis that the latter changes reflect the dendritic atrophy induced by repeated stress or depression within the visceromotor circuits that activate during stress and depression.

The observation that the reductions in GM in BD appear left-lateralized both in the current study and in most previous studies may be compatible with the hypothesis that elevated excitatory transmission plays a role in the pathogenesis of neuromorphometric changes in BD. For example, in the subgenual ACC, abnormal reductions in GM volume were found on the left but not the right sides in BD (Drevets et al., 1997; Hirayasu et al., 1999). Similarly, in the hippocampal subiculum, which shares extensive predominantly ipsilateral connections with the subgenual ACC, volume was reduced on the left, but not the right side in BD (Nugent et al., 2004). The accumbens area shares predominantly ipsilateral connections with both the subgenual ACC and the subiculum, and the post mortem study of Baumann et al. (1999) specifically showed that the GM volume was reduced in the left accumbens area. In the STG, Chen et al. (2004) found that GM volume was significantly reduced on the left but not the right side in children and adolescents with BD. Finally, in the lateral orbital cortex, post mortem neuropathological studies of MDD found reductions in cell counts on the left side (Rajkowska et al., 1999), and MRI-based studies of putative cerebrovascular lesions in late-onset depression specifically associated an increased risk for developing the major depressive syndrome with lesions involving the left orbital cortex (MacFall et al., 2001). The abovementioned evidence that glucose metabolism increases on the left side in these regions in the depressed versus the non-depressed phase of mood disorders suggests that glutamatergic transmission may be increased in the visceromotor circuit predominantly in the left

hemisphere, potentially accounting for the left-lateralized anatomical distribution of the GM reductions in BD.

Summary

This study revealed morphometric abnormalities in BD in regions of the medial and orbital PFC, STG and PCC that, together with anatomically related areas of the amygdala, hippocampus, striatum, thalamus, hypothalamus and brain stem, form part of a visceromotor network that regulates the expression and experience of emotion. The separation of the BD subjects on the basis of recent medication status enabled exploration of psychotropic drug effects on these abnormalities that can facilitate the design of longitudinal studies aimed at differentiating the effects of medication from effects of disease severity/recurrence on GM volume. Elucidating the neurobiological bases for the neuromorphometric abnormalities extant in BD is likely to provide invaluable insights into the pathogenesis of BD and may ultimately guide the development of more effective treatments.

Acknowledgments

Thanks to Michele Drevets, Joan Williams and Joana-Taylor Tavares for their work on this project. This research was supported by the Intramural Program of the NIH, NIMH.

References

Adler, C.M., Levine, A.D., Delbello, M.P., Strakowski, S.M., 2005. Changes in gray matter volume in patients with bipolar disorder. Biol. Psychiatry 58 (2), 151–157.

American Psychiatric Association, 2000. Diagnostic and Statistical Manual of Mental Disorders DSM-IV-TR (Text Revision). American Psychiatric Association, Arlington, VA.

Ashburner, J., Friston, K.J., 2000. Voxel-based morphometry—The methods. NeuroImage 11 (6 Pt. 1), 805–821.

Baumann, B., Danos, P., Krell, D., Diekmann, S., Leschinger, A., Stauch, R., Wurthmann, C., Bernstein, H.G., Bogerts, B., 1999. Reduced volume of limbic system-affiliated basal ganglia in mood disorders: preliminary data from a postmortem study. J. Neuropsychiatry Clin. Neurosci. 11 (1), 71–78.

Beasley, C.L., Chana, G., Honavar, M., Landau, S., Everall, I.P., Cotter, D., 2005. Evidence for altered neuronal organisation within the planum temporale in major psychiatric disorders. Schizophr. Res. 73 (1), 69–78.

Bench, C.J., Friston, K.J., Brown, R.G., Frackowiak, R.S., Dolan, R.J., 1993. Regional cerebral blood flow in depression measured by positron emission tomography: the relationship with clinical dimensions. Psychol. Med. 23 (3), 579–590.

Black, K.J., Hershey, T., Hartlein, J.M., Carl, J.L., Perlmutter, J.S., 2005. Levodopa challenge neuroimaging of levodopa-related mood fluctuations in Parkinson's disease. Neuropsychopharmacology 30 (3), 590–601.

Bowen, D.M., Najlerahim, A., Procter, A.W., Francis, P.T., Murphy, E., 1989. Circumscribed changes of the cerebral cortex in neuropsychiatric disorders of later life. Proc. Natl. Acad. Sci. U. S. A. 86 (23), 9504–9508.

Brambilla, P., Harenski, K., Nicoletti, M., Sassi, R.B., Mallinger, A.G., Frank, E., Kupfer, D.J., Keshavan, M.S., Soares, J.C., 2003. MRI investigation of temporal lobe structures in bipolar patients. J. Psychiatr. Res. 37 (4), 287–395.

Buchsbaum, M.S., Wu, J., Siegel, B.V., Hackett, E., Trenary, M., Abel, L., Reynolds, C., 1997. Effect of sertraline on regional metabolic rate in patients with affective disorder. Biol. Psychiatry 41 (1), 15–22.

Calamante, F., Lythgoe, M.F., Pell, G.S., Thomas, D.L., King, M.D., Busza, A.L., Sotak, C.H., Williams, S.R., Ordidge, R.J., Gadian, D.G., 1999. Early changes in water diffusion, perfusion, T1, and T2 during focal cerebral ischemia in the rat studied at 8.5 T. Magn. Reson. Med. 41 (3), 479–485.

Chana, G., Landau, S., Beasley, C., Everall, I.P., Cotter, D., 2003. Two-dimensional assessment of cytoarchitecture in the anterior cingulate cortex in major depressive disorder, bipolar disorder, and schizophrenia: evidence for decreased neuronal somal size and increased neuronal density. Biol. Psychiatry 53 (12), 1086–1098.

Chen, H.H., Nicoletti, M.A., Hatch, J.P., Sassi, R.B., Axelson, D., Brambilla, P., Monkul, E.S., Keshavan, M.S., Ryan, N.D., Birmaher, B., Soares, J.C., 2004. Abnormal left superior temporal gyrus volumes in children and adolescents with bipolar disorder: a magnetic resonance imaging study. Neurosci. Lett. 363 (1), 65–68.

Corson, P.W., Nopoulos, P., Miller, D.D., Arndt, S., Andreasen, N.C., 1999. Change in basal ganglia volume over 2 years in patients with schizophrenia: typical versus atypical neuroleptics. Am. J. Psychiatry 156 (8), 1200–1204.

Doris, A., Belton, E., Ebmeier, K.P., Glabus, M.F., Marshall, I., 2004. Reduction of cingulate gray matter density in poor outcome bipolar illness. Psychiatry Res. 130 (2), 153–159.

Drevets, W.C., 2004. Neuroplasticity in mood disorders. Dialogues Clin. Neurosci. 6 (2), 199–216.

Drevets, W.C., Todd, R.D., 1997. Depression, mania and related disorders. In: Guze, S.B. (Ed.), Adult Psychiatry. Mosby Press, St. Louis, MO, pp. 99–141.

Drevets, W.C., Videen, T.O., Price, J.L., Preskorn, S.H., Carmichael, S.T., Raichle, M.E., 1992. A functional anatomical study of unipolar depression. J. Neurosci. 12 (9), 3628–3641.

Drevets, W.C., Price, J.L., Simpson Jr., J.R., Todd, R.D., Reich, T., Vannier, M., Raichle, M.E., 1997. Subgenual prefrontal cortex abnormalities in mood disorders. Nature 386 (6627), 824–827.

Drevets, W.C., Bogers, W., Raichle, M.E., 2002. Functional anatomical correlates of antidepressant drug treatment assessed using PET measures of regional glucose metabolism. Eur. Neuropsychopharmacol. 12 (6), 527–544.

Drevets, W.C., Gadde, K.M., Krishnan, K.R., 2004. Neuroimaging studies of mood disorders. In: Charney, D.S. (Ed.), Neurobiology of Mental Illness. Oxford University Press, Oxford, UK, pp. 461–490.

Duman, R.S., 2004. Role of neurotrophic factors in the etiology and treatment of mood disorders. Neuromolecular Med. 5 (1), 11–25.

Eastwood, S.L., Harrison, P.J., 2000. Hippocampal synaptic pathology in schizophrenia, bipolar disorder and major depression: a study of complexin mRNAs. Mol. Psychiatry 5 (4), 425–432.

Giedd, J.N., 2004. Structural magnetic resonance imaging of the adolescent brain. Ann. N. Y. Acad. Sci. 1021, 77–85.

Good, C.D., Johnsrude, I.S., Ashburner, J., Henson, R.N., Friston, K.J., Frackowiak, R.S., 2001. A voxel-based morphometric study of ageing in 465 normal adult human brains. NeuroImage 14 (1 Pt. 1), 21–36.

Gundel, H., O'Connor, M.F., Littrell, L., Fort, C., Lane, R.D., 2003. Functional neuroanatomy of grief: an FMRI study. Am. J. Psychiatry 160 (11), 1946–1953.

Gur, R.E., Maany, V., Mozley, P.D., Swanson, C., Bilker, W., Gur, R.C., 1998. Subcortical MRI volumes in neuroleptic-naive and treated patients with schizophrenia. Am. J. Psychiatry 155 (12), 1711–1717.

Hao, Y., Creson, T., Zhang, L., Li, P., Du, F., Yuan, P., Gould, T.D., Manji, H.K., Chen, G., 2004. Mood stabilizer valproate promotes ERK pathway-dependent cortical neuronal growth and neurogenesis. J. Neurosci. 24 (29), 6590–6599.

Hirayasu, Y., Shenton, M.E., Salisbury, D.F., Kwon, J.S., Wible, C.G., Fischer, I.A., Yurgelun-Todd, D., Zarate, C., Kikinis, R., Jolesz, F.A., McCarley, R.W., 1999. Subgenual cingulate cortex volume in first-episode psychosis. Am. J. Psychiatry 156 (7), 1091–1093.

Hirayasu, Y., McCarley, R.W., Salisbury, D.F., Tanaka, S., Kwon, J.S., Frumin, M., Snyderman, D., Yurgelun-Todd, D., Kikinis, R., Jolesz, F.A., Shenton, M.E., 2000. Planum temporale and Heschl gyrus volume reduction in schizophrenia: a magnetic resonance imaging study of first-episode patients. Arch. Gen. Psychiatry 57 (7), 692–699.

Kasai, K., Shenton, M.E., Salisbury, D.F., Hirayasu, Y., Lee, C.U., Ciszewski, A.A., Yurgelun-Todd, D., Kikinis, R., Jolesz, F.A., McCarley, R.W., 2003. Progressive decrease of left superior temporal gyrus gray matter volume in patients with first-episode schizophrenia. Am. J. Psychiatry 160 (1), 156–164.

Ketter, T.A., Kimbrell, T.A., George, M.S., Dunn, R.T., Speer, A.M., Benson, B.E., Willis, M.W., Danielson, A., Frye, M.A., Herscovitch, P., Post, R.M., 2001. Effects of mood and subtype on cerebral glucose metabolism in treatment-resistant bipolar disorder. Biol. Psychiatry 49 (2), 97–109.

Kimbrell, T.A., George, M.S., Parekh, P.I., Ketter, T.A., Podell, D.M., Danielson, A.L., Repella, J.D., Benson, B.E., Willis, M.W., Herscovitch, P., Post, R.M., 1999. Regional brain activity during transient self-induced anxiety and anger in healthy adults. Biol. Psychiatry 46 (4), 454–465.

Kimbrell, T.A., Ketter, T.A., George, M.S., Little, J.T., Benson, B.E., Willis, M.W., Herscovitch, P., Post, R.M., 2002. Regional cerebral glucose utilization in patients with a range of severities of unipolar depression. Biol. Psychiatry 51 (3), 237–252.

Kondo, H., Saleem, K.S., Price, J.L., 2004. The connections of the perirhinal and parahippocampal cortex with the orbital and medial prefrontal networks and the temporal pole in the monkey. Society for Neuroscience 34th Annual Meeting, San Diego, CA.

Lai, T., Payne, M.E., Byrum, C.E., Steffens, D.C., Krishnan, K.R., 2000. Reduction of orbital frontal cortex volume in geriatric depression. Biol. Psychiatry 48 (10), 971–975.

Lochhead, R.A., Parsey, R.V., Oquendo, M.A., Mann, J.J., 2004. Regional brain gray matter volume differences in patients with bipolar disorder as assessed by optimized voxel-based morphometry. Biol. Psychiatry 55 (12), 1154–1162.

Lyoo, I.K., Kim, M.J., Stoll, A.L., Demopulos, C.M., Parow, A.M., Dager, S.R., Friedman, S.D., Dunner, D.L., Renshaw, P.F., 2004. Frontal lobe gray matter density decreases in bipolar I disorder. Biol. Psychiatry 55 (6), 648–651.

MacFall, J.R., Payne, M.E., Provenzale, J.E., Krishnan, K.R., 2001. Medial orbital frontal lesions in late-onset depression. Biol. Psychiatry 49 (9), 803–806.

Maddock, R.J., 1999. The retrosplenial cortex and emotion: new insights from functional neuroimaging of the human brain. Trends Neurosci. 22 (7), 310–316.

Maddock, R.J., Garrett, A.S., Buonocore, M.H., 2003. Posterior cingulate cortex activation by emotional words: fMRI evidence from a valence decision task. Hum. Brain Mapp. 18 (1), 30–41.

Mah, L., Duan, Y.-F., Zarate, C.A., Singh, J., Payne, J.L., Nugent, A.C., Charney, D.S., Drevets, W.C., 2004. Glucose metabolism in the amygdala in bipolar II depression. Biol. Psychiatry 55 (8 Suppl. 1), 1785.

Manji, H.K., Moore, G.J., Chen, G., 1999. Lithium at 50: have the neuroprotective effects of this unique cation been overlooked? Biol. Psychiatry 46 (7), 929–940.

Mayberg, H.S., Brannan, S.K., Tekell, J.L., Silva, J.A., Mahurin, R.K., McGinnis, S., Jerabek, P.A., 2000. Regional metabolic effects of fluoxetine in major depression: serial changes and relationship to clinical response. Biol. Psychiatry 48 (8), 830–843.

McEwen, B.S., Chattarji, S., 2004. Molecular mechanisms of neuroplasticity and pharmacological implications: the example of tianeptine. Eur. Neuropsychopharmacol. 14 (Suppl 5), S497–S502.

Mesulam, M.-M., 2000. Behavioral neuroanatomy: large-scale networks, association cortex, frontal syndromes, the limbic system, and hemispheric specializations. In: Mesulam, M.-M. (Ed.), Principles of Behavioral and Cognitive Neurology. Oxford Univ. Press, London, pp. 1–120.

Moore, G.J., Bebchuk, J.M., Hasanat, K., Chen, G., Seraji-Bozorgzad, N., Wilds, I.B., Faulk, M.W., Koch, S., Glitz, D.A., Jolkovsky, L., Manji, H.K., 2000a. Lithium increases *N*-acetyl-aspartate in the human brain: in vivo evidence in support of bcl-2's neurotrophic effects? Biol. Psychiatry 48 (1), 1–8.

Moore, G.J., Bebchuk, J.M., Wilds, I.B., Chen, G., Manji, H.K., 2000b. Lithium-induced increase in human brain grey matter. Lancet 356 (9237), 1241–1242.

Neumeister, A., Nugent, A.C., Waldeck, T., Geraci, M., Schwarz, M., Bonne, O., Bain, E.E., Luckenbaugh, D.A., Herscovitch, P., Charney, D.S., Drevets, W.C., 2004. Neural and behavioral responses to tryptophan depletion in unmedicated patients with remitted major depressive disorder and controls. Arch. Gen. Psychiatry 61 (8), 765–773.

Nugent, A.C., Wood, S., Bain, E.E., Mah, L., Cannon, D., Neumeister, A., Marrett, S., Koretsky, A.P., Talagala, L., Price, C.J., Charney, D.S., Drevets, W.C., 2004. High resolution MRI neuromorphometric assessment of the hippocampal subiculum in mood disorders. ISMRM Twelfth Scientific Meeting, Kyoto, Japan.

Ongur, D., Price, J.L., 2000. The organization of networks within the orbital and medial prefrontal cortex of rats, monkeys and humans. Cereb. Cortex 10 (3), 206–219.

Osuch, E.A., Ketter, T.A., Kimbrell, T.A., George, M.S., Benson, B.E., Willis, M.W., Herscovitch, P., Post, R.M., 2000. Regional cerebral metabolism associated with anxiety symptoms in affective disorder patients. Biol. Psychiatry 48 (10), 1020–1023.

Parry, A., Clare, S., Jenkinson, M., Smith, S., Palace, J., Matthews, P.M., 2003. MRI brain T1 relaxation time changes in MS patients increase over time in both the white matter and the cortex. J. Neuroimaging 13 (3), 234–239.

Pearlson, G.D., Barta, P.E., Powers, R.E., Menon, R.R., Richards, S.S., Aylward, E.H., Federman, E.B., Chase, G.A., Petty, R.G., Tien, A.Y., 1997. Ziskind-Somerfeld Research Award Medial and superior temporal gyral volumes and cerebral asymmetry in schizophrenia versus bipolar disorder. Biol. Psychiatry 41 (1), 1–14.

Price, J.L., Saleem, K.S., Baker, J.T., Snyder, L.H., 2004. Evaluation of in-vivo axonal tracing in the macaque. Society for Neuroscience, San Diego, CA.

Quinn, B., Fenstermacher, E., Han, X., Pacheco, J., Czanner, S., van der Kouwe, A., Maguire, P., Raunig, D., Albert, M., Makris, N., Desikan, R., Killiany, R., Dickerson, B., Fischl, B., 2005. Test–retest reliability assessment for longitudinal mRI studies: a comparison of the effects of different T1-weighted protocols, scanner platforms, and field strengths on semi-automated hippocampal volume measures. Human Brain Mapping. (Toronto, Canada).

Rajkowska, G., Miguel-Hidalgo, J.J., Wei, J., Dilley, G., Pittman, S.D., Meltzer, H.Y., Overholser, J.C., Roth, B.L., Stockmeier, C.A., 1999. Morphometric evidence for neuronal and glial prefrontal cell pathology in major depression. Biol. Psychiatry 45 (9), 1085–1098.

Rajkowska, G., Halaris, A., Selemon, L.D., 2001. Reductions in neuronal and glial density characterize the dorsolateral prefrontal cortex in bipolar disorder. Biol. Psychiatry 49 (9), 741–752.

Rosoklija, G., Toomayan, G., Ellis, S.P., Keilp, J., Mann, J.J., Latov, N., Hays, A.P., Dwork, A.J., 2000. Structural abnormalities of subicular dendrites in subjects with schizophrenia and mood disorders: preliminary findings. Arch. Gen. Psychiatry 57 (4), 349–356.

Saleem, K.S., Kondo, H., Price, J.L., 2004. Organization of anatomical streams between the temporal cortex and the orbital–medial prefrontal networks in macaque monkeys. Society for Neuroscience 34th Annual Meeting, San Diego, CA.

Salmond, C.H., Ashburner, J., Vargha-Khadem, F., Connelly, A., Gadian, D.G., Friston, K.J., 2002a. Distributional assumptions in voxel-based morphometry. NeuroImage 17 (2), 1027–1030.

Salmond, C.H., Ashburner, J., Vargha-Khadem, F., Connelly, A., Gadian, D.G., Friston, K.J., 2002b. The precision of anatomical normalization in the medial temporal lobe using spatial basis functions. NeuroImage 17 (1), 507–512.

Sassi, R.B., Brambilla, P., Hatch, J.P., Nicoletti, M.A., Mallinger, A.G., Frank, E., Kupfer, D.J., Keshavan, M.S., Soares, J.C., 2004. Reduced left anterior cingulate volumes in untreated bipolar patients. Biol. Psychiatry 56 (7), 467–475.

Sato, K., Taki, Y., Fukuda, H., Kawashima, R., 2003. Neuroanatomical database of normal Japanese brains. Neural Netw. 16 (9), 1301–1310.

Schlaepfer, T.E., Harris, G.J., Tien, A.Y., Peng, L.W., Lee, S., Federman, E.B., Chase, G.A., Barta, P.E., Pearlson, G.D., 1994. Decreased regional cortical gray matter volume in schizophrenia. Am. J. Psychiatry 151 (6), 842–848.

Sewards, T.V., Sewards, M.A., 2002. The medial pain system: neural representations of the motivational aspect of pain. Brain Res. Bull. 59 (3), 163–180.

Sowell, E.R., Thompson, P.M., Holmes, C.J., Batth, R., Jernigan, T.L., Toga, A.W., 1999. Localizing age-related changes in brain structure between childhood and adolescence using statistical parametric mapping. NeuroImage 9 (6 Pt. 1), 587–597.

Strakowski, S.M., Delbello, M.P., Adler, C.M., 2005. The functional neuroanatomy of bipolar disorder: a review of neuroimaging findings. Mol. Psychiatry 10 (1), 105–116.

Talaraich, J., Tournoux, P., 1988. Co-Planar Stereotaxic Atlas of the Human Brain. Georg Thieme Verlag.

Todtenkopf, M.S., Vincent, S.L., Benes, F.M., 2005. A cross-study meta-analysis and three-dimensional comparison of cell counting in the anterior cingulate cortex of schizophrenic and bipolar brain. Schizophr. Res. 73 (1), 79–89.

Tolle, T.R., Kaufmann, T., Siessmeier, T., Lautenbacher, S., Berthele, A., Munz, F., Zieglgansberger, W., Willoch, F., Schwaiger, M., Conrad, B., Bartenstein, P., 1999. Region-specific encoding of sensory and affective components of pain in the human brain: a positron emission tomography correlation analysis. Ann. Neurol. 45 (1), 40–47.

Vogt, B., 1993. In: Vogt, B., Gabriel, M. (Eds.), Neurobiology of Cingulate Cortex and Limbic Thalamus. Birkhauser, Boston, pp. 19–70.

Wilke, M., Kaufmann, C., Grabner, A., Putz, B., Wetter, T.C., Auer, D.P., 2001. Gray matter-changes and correlates of disease severity in schizophrenia: a statistical parametric mapping study. NeuroImage 13 (5), 814–824.

Wilke, M., Kassubek, J., Ziyeh, S., Schulze-Bonhage, A., Huppertz, H.J., 2003a. Automated detection of gray matter malformations using optimized voxel-based morphometry: a systematic approach. NeuroImage 20 (1), 330–343.

Wilke, M., Sohn, J.H., Byars, A.W., Holland, S.K., 2003b. Bright spots: correlations of gray matter volume with IQ in a normal pediatric population. NeuroImage 20 (1), 202–215.

Wilke, M., Kowatch, R.A., DelBello, M.P., Mills, N.P., Holland, S.K., 2004. Voxel-based morphometry in adolescents with bipolar disorder: first results. Psychiatry Res. 131 (1), 57–69.

Worsley, K.J., Marrett, S., Neelin, P., Vandal, A.C., Friston, K.J., Evans, A.C., 1996. A unified statistical approach for determining significant signals in cerebral activation. Hum. Brain Mapp. 4 (1), 58–73.

Zhang, Y., Brady, M., Smith, S., 2001. Segmentation of brain MR images through a hidden Markov random field model and the expectation maximization algorithm. IEEE Trans. Med. Imaging 20 (1), 45–57.