Reduced Muscarinic Type 2 Receptor Binding in Subjects With Bipolar Disorder

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Context: A variety of indirect evidence has implicated the central muscarinic-cholinergic system, and more specifically the type 2 muscarinic (M2) receptor, in the pathogenesis of depressive symptoms arising in major depressive disorder and bipolar disorder.

Objective: To assess the binding potential of muscarinic2 receptors in vivo during depression in subjects with major depressive disorder or bipolar disorder.

Design: The M2 receptor binding was compared between unmedicated subjects with major depressive disorder or bipolar disorder during depression vs healthy controls, using positron emission tomography and [18F]FP-TZTP (fluorodopa F 18 [3-(3-[3-fluoroproply]thio)-1,2,5-thiadiazol-4-yl]-1,2,5,6-tetrahydro-1-methylpyridine), a selective M2 receptor radioligand.

Setting: Outpatients at the National Institutes of Health.

Participants: Unmedicated subjects with current depression meeting DSM-IV criteria for either major depressive disorder (n=17) or bipolar disorder (n=16) and 23 healthy control subjects.

Main Outcome Measures: The primary outcome parameter was [18F]FP-TZTP distribution volume, which is proportional to the product of receptor density and affinity and, in the case of [18F]FP-TZTP, is known to be sensitive to endogenous acetylcholine concentrations. The relationship between illness severity, as rated using the Montgomery-Asberg Depression and Hamilton Anxiety Rating scales, and distribution volume also was assessed.

Results: The mean anterior cingulate cortex distribution volume differed across groups (F55=3.4; P=0.04), and this difference was accounted for by significantly lower binding in bipolar disorder compared with both major depressive disorder and control groups.

Conclusions: The mean M2 receptor binding in subjects with bipolar disorder was reduced relative to both healthy controls and subjects with major depressive disorder, to an extent that correlated with depressive symptoms. The reduction in the bipolar disorder group could be accounted for either by a reduction in M2 receptor density or affinity or an elevation in endogenous acetylcholine levels. To our knowledge, these data provide the first direct evidence that altered M2 receptor function contributes to mood dysregulation in bipolar disorder.

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PATHOLOGICAL DISTURBANCES of mood may follow a “bipolar” course (termed bipolar disorder [BD] or, formerly, manic-depressive illness), in which normal mood alternates with both depression and mania, or a “unipolar” course (termed major depressive disorder [MDD]), in which only depression occurs. These conditions are common and disabling, to an extent that MDD is the leading cause of disability for years of life lived worldwide. Nevertheless, little is known about the pathophysiology of these conditions.

The muscarinic-cholinergic system has been implicated in BD and MDD by findings that increasing cholinergic transmission via administration of muscarinic receptor agonists or acetylcholinesterase inhibitors exacerbates depressive symptoms in both illnesses and reduces manic symptoms in BD. Moreover, neurophysiological responses to muscarinic receptor–agonist challenge are exaggerated both in subjects with current depression and subjects with remitted MDD or BD relative to controls. The muscarinic-cholinergic system generally has been
shown to play roles in evaluating and learning the salience of sensory stimuli, suggesting that disturbances of muscarinic function may alter the perception of emotionally valenced events.4

Although the mechanism underlying the abnormal sensitivity to muscarinic agonists seen in mood disorders remains unclear, several types of evidence specifically implicate the type 2 muscarinic (M2) receptor in mood regulation5-7 in general and in MDD in particular. Type 2 muscarinic receptors have been identified presynaptically and postsynaptically, and along with the type 4 muscarinic receptor (M4), constitute the predominant muscarinic inhibitory autoreceptor subtypes (ie, receptor stimulation decreases acetylcholine [ACh] release). In healthy humans, administration of the M2 antagonist procaine, which putatively increases intrasynaptic ACh concentrations, elicits a spectrum of robust emotional responses, ranging from sadness, fear, and severe anxiety, to euphoria, that resembles the spectrum of emotional symptoms manifested in BD. These responses were associated with physiological activation of limbic structures, primarily the anterior cingulate cortex (ACC) (Brodman area 24), a region densely innervated by cholinergic neurons originating from the basal forebrain that also is implicated in the pathophysiology of MDD and BD by neuroimaging and neuropathological evidence. In genetic studies of MDD, several single nucleotide polymorphisms of the M2 gene have been associated with increased risk for developing major depressive episodes.6,7 Nevertheless, to our knowledge, the central M2 receptor system has not been previously assessed in vivo in mood disorders.

The development of an M2 receptor, selective, positron emission tomographic (PET) radioligand, [18F]FP-TZTP (fluorodopa F 18 [3-(3-[3-fluoropropyliothio]-1,2,5-thiadiazol-4-yl]-1,2,5,6-tetrahydro-1-methylpyridine), recently made noninvasive investigations of the central M2 receptor system feasible in humans. In the human brain, [18F]FP-TZTP binding was displaced 90% by administration of procaine.3 In vitro analyses determined that [18F]FP-TZTP had inhibition constants (Ki, a measure inversely related to affinity) of 2.2 nMol/L for M2 receptors and 7.4 nMol/L for type 1 muscarinic (M1) receptors, with much weaker affinities for type 3 muscarinic (M3) or σ-1 receptors (Ki = 79.7 and 62.1 nM, respectively). Consistent with these values, the cerebral [18F]FP-TZTP binding was markedly reduced (51%-61%) in M2 receptor knockout mice in all brain regions tested but was unchanged in M3 and M4 receptor knockout mice and showed only slight reductions in M1 receptor knockout mice (that reached statistical significance only in the amygdala and hippocampus, which contain very high M2 receptor concentrations) compared with wild-type mice.9

The cerebral binding of [18F]FP-TZTP also showed sensitivity to changes in intrasynaptic ACh concentrations.10 Because [18F]FP-TZTP is an M2 receptor agonist, this ligand predominately binds to M2 receptors currently existing in the high-affinity state (ie, the population of receptors bound to cell membranes and coupled to G proteins so that stimulation can illicit a functional response). This property putatively renders [18F]FP-TZTP sensitive to intrasynaptic ACh concentrations because this radioligand’s binding can be reduced either by direct competition for receptor binding from endogenous ACh or via agonist-mediated receptor internalization (a process by which receptor stimulation results in the conversion of receptors from the high- to low-affinity state).11 In monkeys, for example, administration of the acetylcholinesterase inhibitor physostigmine (200 mg/kg per minute) decreased cortical-specific [18F]FP-TZTP binding by 25% to 35%.10 Differences in [18F]FP-TZTP binding between diagnostic groups may therefore reflect differences in M2 receptor density, affinity, and/or endogenous ACh concentrations.

The current study compared [18F]FP-TZTP binding between MDD, BD, and control samples. Based on evidence that physostigmine-induced increases in intrasynaptic ACh concentrations can worsen depressive symptoms, we hypothesized that subjects during depression would have reduced ACC [18F]FP-TZTP binding relative to controls. Additionally, because the cholinergic system plays major roles in evaluating and learning the salience of emotional stimuli,3 relationships between [18F]FP-TZTP binding and depression severity, anxiety severity, and performance on an emotionally valenced word-rating task were assessed.

METHODS

PARTICIPANTS

Subjects with current depression meeting the conventional DSM-IV criteria for BD (n = 16) or recurrent MDD (n = 17) and 23 psychiatrically healthy controls were studied. Exclusion criteria included exposure to psychotropic drugs, including nicotine or medications with anticholinergic activity, within the 3 weeks prior to scanning (8 weeks for fluoxetine); pregnancy; major medical or neurological illnesses; lifetime history of substance dependence; or substance abuse within 1 year. Illness severity was assessed using the Montgomery-Asberg Depression (MADRS),12 the Hamilton Anxiety Rating (HAM-A),13 and the Young Mania Rating (YMRS) scales.14 Eleven subjects with BD, 14 subjects with MDD, and 13 control subjects subjectively rated the salience of previously validated, emotionally valenced words on a 7-point scale (very negative, negative, somewhat positive, positive, and very positive).

IMAGE ACQUISITION AND PROCESSING

The PET scans were acquired using a GE Healthcare Advance scanner in 3-dimensional mode (reconstructed 3-dimensional spatial resolution = 9 mm full width at half maximum).10 An 8-minute transmission scan was acquired using rotating rods of 68 germanium/68 gallium and then used to perform measured attenuation correction for each subject’s emission image. Two cerebral blood flow (CBF) images then were obtained following intravenous bolus injection of 370 MBq (10 mCi) of [15O]H2O as subjects rested with eyes closed. The CBF scans were separated by 10 minutes. [18F]FP-TZTP was synthesized according to a recently developed automated synthesis procedure.16 Twelve minutes after the final [15O]H2O injection, 352 to 389 MBq (9.5-10.5 mCi) of high-specific activity [18F]FP-TZTP was administered by bolus intravenous injection and a 120-minute dynamic emission scan was acquired as 33 frames of increasing length across 120
minutes (6 × 0.5 minute, 3 × 1 minute, 2 × 2 minutes, and 22 × 5 minutes). The arterial input function for \(^ {18}F\)FP-TZTP was generated by quantifying the plasma concentration of parent \(^ {18}F\)FP-TZTP using a hexane-extraction procedure\(^ {17}\) in 28 serial blood samples drawn at increasing intervals from a radial artery cannulation site (number × frame duration [in minutes]: 6 × 0.25, 5 × 0.5, 2 × 1, 3 × 2, 1 × 3, 5 × 5, 2 × 10, and 4 × 15).

Head motion was minimized during scanning by stabilizing subjects’ heads using a thermoplastic mask that was fixed to the scanner bed. In addition, the PET data were corrected for head motion by aligning all frames to an early high–radioactive count frame (the first 5-minute frame from 10–15 minutes postinjection) using automated image registration.\(^ {18}\) These frame × frame–aligned PET images then were realigned to 3-T magnetic resonance images (MRIs) (General Electric Signa Scanner 3-dimensional magnetization-prepared rapid gradient echo sequence, echo time = 2.982 milliseconds, repetition time = 7.6 milliseconds, inversion time = 725 milliseconds, voxel size = 0.86 × 0.86 × 1.2 mm) for each subject using FLIRT (Oxford Centre for Functional Magnetic Resonance Imaging of the Brain Linear Image Registration Tool) using a mutual information cost function.\(^ {19}\)

The PET data were corrected for partial volume effects before kinetic modeling.\(^ {20}\) Partial volume correction was performed using the Muller-Gartner\(^ {20}\) algorithm as applied by the Adaptive Fuzzy C Means\(^ {21}\) algorithm. This algorithm assumes that cerebrospinal fluid holds zero radioactivity and that the tissue radioactivity of the white matter is both uniform and estimated by fitting the activity in PET pixels with high smoothed white matter membership (>0.99) to a straight line and extrapolating to 1.0.

Distribution volumes (DVs) (DV = \(K_s/k_2\), where \(K_s\) is the rate of delivery of \(^ {18}F\)FP-TZTP or influx constant from plasma to tissue and \(k_2\) is the rate of clearance) and \(K_s\) values were obtained using the plasma-corrected arterial input function and regional tissue pixel’s time radioactivity concentration curves fitted with a 1-tissue compartment model and nonlinear parameter estimation algorithm.\(^ {22}\) Functional DV images corrected for protein binding of the parent radioligand (DV/f1, where \(f_1\) is the plasma free fraction) were generated based on the pixel-wise ratio of \(K_s\) to \(k_2\).

A region of interest (ROI) template was generated on the Montreal Neurological Institute template MRI and applied to all subjects’ registered MRIs using Statistical Parametric Mapping software (SPM2).\(^ {23}\) Each ROI then was positioned by a rater blind to the PET data to improve anatomical specificity (eg, to ensure ROIs were centered within each subject’s structure of interest [Figure 1A]). These regions were then transformed back into each subject’s native magnetic resonance space using automated image registration.\(^ {18}\) A binary mask of the gray matter (Figure 1B) was then used to ensure that only gray matter pixels were included in the analysis. Regions were then transferred to the coregistered PET images (Figure 1C), and the mean DV was obtained for each ROI (Figure 1D) using MEDX (Medical Numerics Inc, Sterling, Va).

The anterior cingulate ROI (Figure 1A) encompassed the anterior cingulate gyrus (or gyri in cases where the cingulate gyrus was bifurcate) from 3 slices ventral to the ventral aspect of the corpus callosum (subgenual), extending dorsally 13 slices through the ACC anterior to the genu of the corpus callosum (pregenual). The posterior cingulate ROI encompassed the posterior cingulate gyrus from the ventral tip of the splenium of the corpus callosum, extending 9 slices dorsally through the retrosplenial portion of the cingulate gyrus. The dorsal cingulate ROI encompassed the extent of the cingulate gyrus extending dorsal to the superior aspect of the corpus callosum. Amygdalar and hippocampal ROIs were placed as described in

**Figure 1.** A. Magnetic resonance images showing region of interest placement. B, Binary gray matter masks used to measure regional distribution volume (DV). C, [\(^ {18}F\)FP-TZTP fluorodopa F 18-[3-(3-fluoropropyl)thio]-1,2,5-thiadiazol-4-yl]-1,2,5,6-tetrahydro-1-methylpyridine) DV images. D, Regional [\(^ {18}F\)FP-TZTP DV in the primary and secondary structures of interest. Analysis of variance (ANOVA) significance at \(P<.05\); ACC indicates anterior cingulate cortex; DCC, dorsal cingulate cortex; PCC, posterior cingulate cortex; *Bipolar disorder (BD)**; healthy controls and BD < major depressive disorder (MDD) for t test \(P<.05\); †ANOVA significance at \(P_{\text{uncorrected}}<.05\); ‡healthy controls \(P_{\text{uncorrected}}<.05\); §BD < MDD \(P_{\text{uncorrected}}<.05\). Drevets et al.\(^ {23}\) The ventral striatum ROI was placed as an oval (10 × 8 mm) centered in ventral striatum gray matter of the acumbens area situated anterior to the anterior commissure immediately ventral to the ventral tip of the internal capsule and then extending dorsally 2 additional slices (Figure 1A). The orbital cortex was defined in coronal slices to encompass the medial and lateral orbital gyri running between the beginning and end of the olfactory sulcus. The visual cortex ROI was defined by placing circular ROI (20-mm diameter) to encompass the right and left occipital cortex lying along the calcinar sulcus (V1) and extending posteriorly to the occipital pole (V2) through 4 planes (Figure 1A).
**STATISTICAL ANALYSIS**

A priori hypothesis testing using analysis of variance with t tests for specific contrasts between groups was performed on regional DV values in the ACC. Secondary ROIs were assessed to address the specificity of the ACC results. Secondary ROIs included those implicated in emotion regulation generally and mood disorders specifically: posterior (PCC) and dorsal (DCC) cingulate cortices, amygdala, hippocampus, ventral striatum, and lateral orbital cortex. The visual cortex also was included because of its high M2–M1 receptor ratio. Regional and whole brain gray matter ROIs were defined using a gray matter mask segmented from the MRIs and then transferred to the coregistered PET images. Because M2 receptors are expressed in white as well as gray matter, an ROI was similarly defined in deep white matter (defined as white matter voxels situated at least twice the full width at half maximum from any gray matter voxel) in the segmented MRIs. Relationships between [18F]FP-TZTP DV and depression, anxiety, and emotionally valenced word rating were evaluated using Spearman correlations. To reduce the possibility of type II error, voxel × voxel analysis of nonpartial volume correction DV images was performed post hoc using Statistical Parametric Mapping software (SPM2).

![Figure 2](image)

**RESULTS**

The 3 groups were similar for mean ± SD age and sex composition (BD group, age, 32 ± 8 years; 12 of 16 were female; MDD group, age, 36 ± 7 years; 12 of 17 were female; healthy controls, age, 33 ± 6 years; 16 of 23 were female). The mean ± SD age at onset for the MDD group was 20 ± 6 years (range, 10-30 years) and for the BD group, 14 ± 5 years (range, 8-27 years). Seven subjects with BD and 4 subjects with MDD were treatment naive, and the remaining subjects were free of psychotropic medication for a minimum of 3 weeks, with their mean ± SD time not taking medication prior to scanning being 9 ± 5 and 6 ± 7 months, respectively. Five patients had a remote history of substance abuse (none within the 12 months prior to scanning). The mean ± SD depression and anxiety ratings were similar in the BD (MADRS, 27 ± 9; HAM-A, 17 ± 7; YMRS, 8 ± 7) and MDD (MADRS, 24 ± 7; HAM-A, 15 ± 5; YMRS, 5 ± 3) groups and were significantly greater than those of the controls (MADRS, 0.14 ± 0.01; HAM-A, 0.71 ± 1). The mean ± SD duration of illness was similar in the BD (18.5 ± 10.5 years) and MDD (17.5 ± 6.4 years) samples.

The mean ACC DV differed across groups (F55 = 3.4; P = .04), and this difference was accounted for by significantly lower binding in the BD compared with both the MDD and control groups (Figure 1). Comparisons in the secondary ROI showed 12% to 16% reductions in total gray and white matter, DCC, and posterior cingulate, orbital, and visual cortices (Figure 1). Although differences between the BD and control groups in these ROIs would not remain significant after Bonferroni corrections, the probability of detecting differences at Puncorrected <.05 in 6 of 9 regions is substantially higher than chance. The magnitude of the observed differences (percentage change in DV) between groups was similar in both the partial volume corrected (−15.3%) and the uncorrected (−13.9%) image data in the ACC and in the other regions examined.

In contrast, binding in subjects with MDD did not differ significantly from controls. The voxel × voxel analysis performed post hoc revealed significantly lower binding in subjects with BD relative to controls after correction for multiple comparisons using the conservative cluster test in the DCC, ACC, and anterior corpus callosum (Puncorrected <.001) (Figure 2).

The mean ± SD ACC CBF did not differ significantly between subjects with BD (67.0 ± 17.7 mL/min per hектogram) and MDD (63.5 ± 23.7 mL/min per hектogram) and control subjects (66.4 ± 25.4 mL/min per hектogram) nor in any other of the predefined ROIs. Consistent with these data, the mean ± SD delivery rate parameter of [18F]FP-TZTP (K1, milliliters of plasma per minute per milliliter of tissue) did not differ significantly across groups in the ACC (controls, 0.46 ± 0.10; MDD, 0.49 ± 0.12; BD, 0.46 ± 0.09) or in any region examined. The [18F]FP-TZTP uptake is rapid and is rate limited by blood flow. According to blood flow and K1, were correlated (r = 0.42; df = 55; P = .002).

The gray matter volume did not differ between the control, MDD, or BD groups in the ACC (mean ± SD, 10.409 ± 1.560 mL; 10.126 ± 1.094 mL; and 10.333 ± 1.182 mL, respectively), whole brain (mean ± SD, 644.54 ± 77.586 mL, 647.83 ± 58.411 mL, and 628.06 ± 51.628 mL, respectively), or any other region examined.

Depression (r = −0.48; df = 15; P = .03) and anxiety (r = −0.54; df = 15; P = .03) severity correlated negatively with the ACC DV values in the BD sample. Post hoc exploratory analyses also demonstrated (Puncorrected <.05) negative correlations (which would not have remained significant after applying corrections for multiple comparison) between regional DV and depression severity in the hippocampus, amygdala, ventral striatum, and DCC (r = −0.55, −0.47, −0.43, and −0.43, respectively) and between DV and anxiety severity in the
amygdala, hippocampus, DCC, and ventral striatum (r = -0.55, -0.53, -0.51, and -0.50, respectively). Rat-
ings of the salience of emotionally valenced words correlated negatively with the ACC DV values in the BD group (r = -0.65; df = 10; P = .03) and the entire sample (r = -0.42; df = 37; P = .008) but not in the MDD (r = -0.41; df = 13; P = .15) or control groups (r = -0.193; df = 12; P = .53). Post hoc assessments of corresponding relationships involving the secondary ROI showed that regional DV also correlated negatively (Puncorrected < .05) with salience ratings in cerebral gray matter, DCC, PCC, and orbital and visual cortices (r = -0.71, -0.73, -0.67, -0.66, and -0.64, respectively). In contrast, salience ratings by controls and subjects with MDD were not correlated significantly with the DV of any region examined. The mean ± SD salience score for the BD (3.9 ± 0.9; n = 11), MDD (4.0 ± 0.6; n = 14), and control groups (3.9 ± 0.8; n = 13) did not differ significantly.

Further exploratory post hoc analyses aimed at examining the relationship between DV and specific symptoms revealed that the MADRS item for reported sadness (r = -0.60) and the HAM-A items for tension (r = -0.61), depressed mood (r = -0.53), and genital and urinary somatic complaints (r = -0.56) contributed to the observed correlations with symptom severity. The total Inventory for Depressive Symptomatology Clinician-Rated scale (IDS-C) score (BD group, r = -0.44; MDD group, r = -0.39) and the score for the anxiety symptom cluster within the IDS-C (BD group, r = -0.25; MDD group, r = -0.22) did not correlate with ACC DV in either patient group. However, the IDS-C anhedonia symptom cluster correlated with ACC DV in subjects with BD (r = -0.60; df = 15; P = .01) but not in subjects with MDD (r = -0.32; df = 15; P = .38), and the IDS-C depression symptom cluster trended in the same direction as the MADRS in subjects with BD (r = -0.47; df = 15; P = .06) but not in subjects with MDD (r = -0.37; df = 15; P = .16). None of the items or symptom clusters showing correlations with ACC DV differed significantly between the MDD and BD samples (P > .10).

**COMMENT**

The a priori hypothesis that the [18F]FP-TZTP DV would be reduced in the ACC in depression was confirmed in subjects with BD during depression. In contrast, [18F]FP-TZTP binding did not differ significantly between the MDD and control samples and was significantly lower in the BD group relative to the MDD group. Moreover, in comparisons involving other regions, this abnormality appeared relatively widespread in subjects with BD relative to control subjects and subjects with MDD (Figure 1). In addition to the abnormal binding values in the secondary ROI that were significant before but not after Bonferroni correction, voxel × voxel analysis additionally revealed significantly lower binding in the DCC as well as the ACC that remained significant after applying corrections for multiple comparisons. The a priori hypothesis that depression and anxiety severity would correlate negatively with ACC binding was confirmed in the subjects with BD. The degree of emotionality assessed to the affectively valenced words and the severity of anhedonia symptom ratings also were found to correlate inversely to ACC binding, although these latter analyses were considered exploratory.

Reduced [18F]FP-TZTP DV in BD may reflect increased intrasynaptic ACh concentrations or decreased M2 receptor density or affinity. However, a recent report that ACC M2 receptor density (Bmax) was unaltered in subjects with BD relative to controls postmortem would appear to inform the interpretation of our results by suggesting that the total population of M2 receptors is not decreased in BD. These post-mortem data were obtained using a radiolabelled M2, antagonist, which quantified the density of high- plus low-affinity–state M2 receptors, without sensitivity to endogenous ACh concentrations. The data reported herein, in contrast, were obtained using a radiolabelled M2 agonist, which predominantly quantified M2 receptors in the high-affinity (active) state, thereby providing a measure more sensitive to the functional state of this cholinergic receptor system. The in vivo [18F]FP-TZTP binding data reported herein taken together with the post-mortem data described earlier suggest, therefore, that the pathophysiology of BD involves elevations in intrasynaptic ACh concentrations and/or reductions in M2 receptor affinity (eg, a smaller proportion of receptors extant in the high-affinity relative to the low-affinity state, possibly the result of a compensatory response to elevated ACh release) without an actual reduction in the total number of M2 receptors.

An alternative explanation for the reduction in [18F]FP-TZTP binding in the BD sample may be that neuronal processes expressing M2 receptors are quantitatively reduced in BD. The gray matter of the subgenual portion of the ACC has been shown to be decreased in subjects with BD relative to controls. This explanation appears unlikely for several reasons, however. First, the gray matter reductions in this area were found to a similar extent in MDD cases as in BD cases, yet the [18F]FP-TZTP binding did not differ in this region between the MDD and control groups. Second, the ACC ROI applied in the present study included more dorsal areas of the ACC where no differences in gray matter were found between controls and subjects with BD in the same study. Third, in the current study, the number of gray matter voxels within each ROI did not differ significantly across groups for any region examined.

These observations also are unlikely to be accounted for by differences in blood flow between the BD and control groups because the modeling method effectively removes the effects of CBF. Consistent with this are the observations that neither radioligand delivery (K1) nor CBF differed across groups. In addition, the ACC ROI used encompassed subgenual and pregenual regions where increases and decreases in blood flow have been reported, respectively; thus, the entire region used in the present study would not be expected to have altered blood flow.

Notably, either increased ACh concentrations or reduced M2-mediated inhibitory regulation over ACh release (via altered autoreceptor function) could account for the exaggerated neurophysiological responses to cholinomimetic agents observed in BD. For example, mice lack-
ing M₁ receptors developed higher intrasynaptic ACh concentrations compared with wild-type mice during stress and maintained this elevation beyond the cessation of stress.34 Thus, it might be speculated that reduced M₁ receptor function in BD contributes to the exaggerated behavioral and clinical responses to stress seen in BD.35

A limitation of the PET–[¹⁸F]FP-TZTP method was the inability to assess free and nonspecific binding in a receptor-free basis because of the widespread distribution of M₂ receptors.24,25 In support of the specificity of DV measures for receptor-specific binding, however, were the observations that [¹⁸F]FP-TZTP binding correlated with illness severity in BD and that the rank order of regions showing the greatest decrease in [¹⁸F]FP-TZTP DV corresponded well to that of the relative M₂ receptor density and of the M₂/M₁ receptor ratio for each region.24 Moreover, the observation that the subjective ratings of the impact of emotionally valenced words correlated inversely with ACC DV was compatible with the known role of the muscarinic-cholinergic system in attributing salience to experiential stimuli.3

The integrity of the cholinergic neuronal projections from the basal forebrain to the cerebral cortex is necessary for a wide range of attentional functions.36-38 However, disinhibition or excessive reactivity of these projections that pathologically elevate cholinergic function produce profound attentional impairments that are hypothesized to result from overprocessing of stimuli and their associations—irrespective of their cognitive and behavioral significance—and the consequent depletion of processing resources available for other activities.36 Increased cholinergic function thus may contribute to some of the marked attentional impairments that characterize BD.35,39 which also were present in our BD sample (D.M.C. and W.C.D., unpublished data, January 2006), and may account for the exaggerated salience ascribed to emotional word stimuli found herein in BD.

The role of the cholinergic system in processing the salience of stimuli also may relate to the correlation found between the [¹⁸F]FP-TZTP binding and the anhedonia ratings. Cholinergic agonist administration has been dose dependently associated with anhedonia in 1 case study in vivo (RS86)59 and with impairment in memory for reductions in reward magnitude (oxotremorine).61 The degree of correlation between the [¹⁸F]FP-TZTP binding in the ACC and the severity of anhedonia, depression, and anxiety accounts for 36%, 23%, and 29% of the variance in these clinical ratings, respectively.

The abnormalities of [¹⁸F]FP-TZTP binding in the ACC and DCC in BD also are noteworthy because neurophysiological activity increases in these regions during pro- caine administration, in association with mood responses that, as in BD, range from severe dysphoria and anxiety to euphoria.3 The electrical stimulation of the ACC elicits intense fear or pleasure in conscious patients with epilepsy62 and vocalizations in monkeys exposed to stress.63 Moreover, in subjects with BD during depression, glucose metabolism is abnormal in the ACC.6 The observations that the [¹⁸F]FP-TZTP DV in the ACC correlated inversely with depression severity and that physostigmine administration exacerbated depressive symptoms in BD support a hypothesis that abnormal cholinergic function in the ACC contributes to the development of pathological mood symptoms.

Finally, the current study found, to our knowledge, one of the clearest neurobiological distinctions between individuals with unipolar and bipolar depression discovered to date. Major depressive disorder and BD appear to some extent genetically distinct, yet they have proven more similar than different in previous neuroimaging studies.35 Although abnormalities of cholinergic sensitivity have been demonstrated in both MDD and BD, the current results suggest they are accounted for by prominent differences in M₂ receptor binding and/or intrasynaptic ACh concentrations only in BD.

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REFERENCES


Correction

Error in Figure Key. In the Original Article titled “Reduced Muscarinic Type 2 Receptor Binding in Subjects With Bipolar Disorder,” published in the July issue of the ARCHIVES (2006; 63:741-747), there is an error in the key for Figure 1D. The blue bars represent controls, the yellow bars represent subjects with major depressive disorder, and the green bars represent subjects with bipolar disorder. The figure and correct key are shown below.

![Figure 1D: Regional [11F]FP-TZTP DV (fluorodopa F 18 [3-(3-fluoropropyl)thio]-1,2,5-thiadiazol-4-yl]-1,2,5,6-tetrahydropyro-1-methylpyridine) in the primary and secondary structures of interest. Analysis of variance (ANOVA) significance at P<.05. ACC indicates anterior cingulate cortex; DCC, dorsal cingulate cortex; PCC, posterior cingulate cortex; *Bipolar disorder (BD)< healthy controls and BD< major depressive disorder (MDD) for ttest P<.05; †ANOVA significance at P<.05; ‡BD< healthy controls P<.05; §BD< MDD P<.05.](https://archpsyc.jamanetwork.com/pdfaccess.ashx?url=/data/journals/psych/5256/ on 04/03/2017)