ORIGINAL ARTICLE

Neural and Genetic Correlates of Antidepressant Response to Sleep Deprivation

A Functional Magnetic Resonance Imaging Study of Moral Valence Decision in Bipolar Depression

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Context: Total sleep deprivation combined with light therapy causes rapid amelioration of bipolar depression. A polymorphism in the promoter for the serotonin transporter influences both antidepressant response and the structure and function of specific brain areas.

Objective: To determine whether antidepressant therapy or the genotype of the serotonin transporter influence the pattern of neural response to a task targeting the depressive biases in information processing (moral valence decision).

Design: Before-and-after trial studying the biologic correlates of response to treatment.

Setting: University hospital.

Patients: Twenty inpatients with bipolar depression.

Intervention: Repeated total sleep deprivation combined with light therapy for 1 week.

Main Outcome Measures: Brain blood oxygen level–dependent functional magnetic resonance imaging using a 3.0-T scanner before and after treatment. Self-ratings and observer ratings of mood (visual analog scale 3 times daily and Hamilton Depression Rating Scale) before and after treatment.

Results: We found significant interactions of treatment (before and after), response to treatment (Hamilton Depression Rating Scale score <8), and moral valence of the stimuli (positive or negative) in the anterior cingulate cortex, dorsolateral prefrontal cortex, insula, and parietal cortex. In these areas, responders changed their blood oxygen level–dependent responses to emotional stimuli in a pattern opposite of that in nonresponders. Genotype of the promoter for the serotonin transporter predicted response to treatment and influenced baseline neural responses in the anterior cingulate cortex and the dorsolateral prefrontal cortex.

Conclusion: Multiple factors that affect or are affected at the individual level by major depressive episodes in the course of bipolar disorder significantly interact in influencing brain cortical activity in specific areas.

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THE COMBINATION OF CLINICAL chronotherapeutic antidepressant techniques such as repeated total sleep deprivation (TSD) and light therapy (LT) has been shown to cause rapid and sustained antidepressant effects in bipolar depression that occur in a matter of hours or days. Though the exact mechanism of action of TSD and LT is still unknown and likely involves changes in the regulation of biologic rhythms, their clinical effect seems to be influenced by the same biologic variables that influence response to pharmacologic antidepressant therapies.[1] The combination of TSD and LT enables the study of the biologic correlates of antidepressant response at close time points and in the absence of the possible confounding factors linked to prolonged drug therapies.[2]

Cognitive distortions are a regular core symptom of bipolar depression and lead to mood-congruent biases in information processing that influence evaluative processes, social judgment, decision making, attention, and memory.[3] When administered a simple go/no-go task with emotional stimuli, patients with bipolar depression demonstrate facilitation of performance when responding to stimuli with a negative emotional tone,[4] showing that

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congruence between mood state of the subject and tone of the stimuli leads to better cognitive performance.

Blood oxygen level–dependent (BOLD) functional magnetic resonance imaging (fMRI) enables the study of the neural correlates of depressed mood and depression recovery. Blood oxygen level–dependent fMRI makes it possible to localize neural responses to stimuli in brain areas by detecting an indirect shift in oxyhemoglobin and deoxyhemoglobin in local vasculature as a function of increased blood flow associated with neuronal activity. By administering blocks of stimuli with a positive or negative emotional tone, several research groups were able to define regions of interest (ROIs) associated with the processing of affective stimuli and the cognitive generation of affect in depressed patients. The ROIs consistently activated included the anterior cingulate cortex (ACC), medial prefrontal cortex (PFC), dorsolateral PFC, and insula.[5,6]

In these same regions, a go/no-go task with blocks of negative and positive emotional words could elicit different neural responses in depressed patients and in healthy control subjects, which suggests a critical role of the medial PFC and ACC in mediating mood-congruent response biases in depression.[7] In one study, neural responses to blocks of negative and positive affective stimuli in the ACC and insula were associated with response to venlafaxine antidepressant therapy.[8] These findings are consistent with positron emission tomography studies, which consistently linked changes in anterior cingulate metabolism with response to sleep deprivation and serotonergic antidepressant agents such as fluoxetine hydrochloride, paroxetine hydrochloride, and sertraline hydrochloride.[9,10]

The search for genetic factors predisposing to antidepressant response in affective disorders showed that a polymorphism in the promoter region of the serotonin transporter (SERTPR) predicted efficacy for a range of treatments,[11] including TSD alone[12] or in combination with LT,[13] in patients affected by bipolar depression. The same SERTPR has been shown to influence core features of affective illness, such as age at onset and recurrence of mood episodes,[14] and, in healthy subjects, the neural response of limbic structures to emotional stimuli[15] and the functional coupling of the ACC and medial PFC with the amygdala.[16,17] In the present study, we studied the effect of TSD combined with LT treatment and SERTPR on event-related neural responses to a go/no-go task with emotional words in a homogeneous sample of patients affected by bipolar depression.

METHODS

PATIENT SAMPLE

We studied 20 right-handed patients (7 men and 13 women) having a diagnosis of bipolar disorder type I, depressive episodes without psychotic features. Clinical and demographic characteristics (mean±SD) included the following: age, 52.15±14.50 years; age at onset, 38.3±14.8 years; number of previous depressive episodes, 7.05±6.78; number of previous manic episodes, 3.37±2.87; and duration of current episode, 23.63±28.89 weeks. Inclusion criteria were a baseline Hamilton Depression Rating Scale (HDRS) score of 18 or higher; absence of other diagnoses on axis I; absence of mental retardation on axis II; absence of pregnancy, history of epilepsy, or major medical and neurologic disorders; no treatment with long-acting neuroleptic drugs in the last 3 months before admission; no treatment with neuroleptic drugs in the last month before admission; and absence of a history of drug or alcohol dependency or abuse within the last 6 months.

Physical examinations, laboratory tests, and electrocardiograms were performed at admission. After complete description of the study to the subjects, written informed consent was obtained. Genotyping of SERTPR was performed as previously described[18] by personnel blinded to the clinical course of illness and to the effects of treatment.

TREATMENT AND CLINICAL ASSESSMENT

All patients were administered 3 consecutive TSD cycles (days 1-6); each cycle was composed of a period of 36 hours awake. On days 1, 3, and 5, patients were totally sleep deprived from 7:00 AM to 7:00 PM of the following day. They were then allowed to sleep during the night of days 2, 4, and 6. Total sleep deprivation was carried out in a room with 80-lux ambient light; patients were administered LT (exposure for 30 minutes to a 400-lux green light) at 3:00 AM during the TSD night and in the morning after recovery sleep, and half an hour after waking, between approximately 8:00 and 9:00 AM.

All patients were taking lithium salts and continued taking them at the same dosage. No other antidepressant drug therapy was combined during the study.

Perceived mood levels were assessed by a self-administered 10-cm visual analog scale 3 times daily, at 8:00 AM and at 2:00 and 8:00 PM (days 1-7). Patients were instructed to rate their mood between "very sad" (on the left) and "very happy" (on the right), with a median "normal" point.

Objective mood ratings were obtained before and after the TSD plus LT treatment (days 1, 2, 3, and 7) in the morning with a modified version of the 21-item HDRS from which items that could not be meaningfully rated because of the TSD procedure and the time frame were excluded (ie, weight changes and insomnia: items 4, 5, 6, and 16) (HDRS-NOW). The categorical criterion of full response to treatment (remission) was an HDRS score less than 8 at day 7.

IMAGE ACQUISITION

Functional images were acquired before and after the TSD plus LT treatment (days 1 and 7) in the early afternoon. Gradient-echo echo-planar images were acquired using a 3.0-T scanner (Gyroscan Intera; Philips Medical Systems, Eindhoven, the Netherlands) using a 6-channel sensitivity encoding (SENSE; Philips Medical Systems) head coil. For each functional run, 200 T2*-weighted axial sections parallel to the anterior commissure-posterior commissure plane were acquired using an echo-planar imaging pulse sequence (repetition time, 2200 ms; echo time, 35 ms; flip angle, 90°; field of view, 230 mm; number of sections, 18; section thickness, 4 mm; and matrix size, 80×80 pixels reconstructed up to 128×128 pixels). Two dummy scans before fMRI acquisition allowed us to obtain longitudinal magnetization equilibrium. Total time acquisition was 7 minutes 29 seconds per trial. On the same occasion and using the same magnet, 22 turbo spin-echo T2 axial sections (repetition time, 3000 ms; echo time, 85 ms; flip angle, 90°; turbo factor, 15; and 5-mm-thick axial sections with a 512×512-pixel matrix and a 230×230 mm^2 field of view) were acquired parallel to the AC-PC plane to rule out brain lesions.

Table 1. Changes in Clinical Measures During Treatment*

Day	Responders (n = 9)		Nonresponders (n = 11)		Entire Sample (N = 20)	
	HDRS	VAS	HDRS	VAS	HDRS	VAS
1	21.00 ± 3.16	41.11 ± 19.05	22.09 ± 3.88	25.03 ± 18.90	21.6 ± 3.53	32.27 ± 20.20
2	12.22 ± 5.29	51.31 ± 17.55	16.45 ± 4.61	32.58 ± 25.59	14.55 ± 5.26	41.00 ± 23.79
3	9.22 ± 4.02†	41.00 ± 18.37	17.73 ± 4.71†	24.94 ± 18.80	13.90 ± 6.11	32.17 ± 19.89
4	. . .	44.87 ± 18.50‡	. . .	25.55 ± 17.45‡	. . .	34.24 ± 20.04
5	. . .	54.00 ± 20.91‡	. . .	29.12 ± 20.17‡	. . .	40.32 ± 23.65
6	. . .	53.26 ± 22.77‡	. . .	30.36 ± 19.50‡	. . .	40.67 ± 23.56
7	2.22 ± 2.86†	59.96 ± 15.80†	16.36 ± 5.08†	26.76 ± 18.09†	10.00 ± 8.31	41.70 ± 23.76

Abbreviations: HDRS, Hamilton Depression Rating Scale; VAS, visual analog scale, . . ., data not collected.
*Values are given as mean ± SD. Responders showed significantly better scores compared with nonresponders from day 3 (ie, after the first total sleep deprivation plus light therapy cycle) and thereafter.
†$P<.01$, *t* test (responders vs nonresponders).
‡$P<.05$, *t* test (responders vs nonresponders).

COGNITIVE ACTIVATION PARADIGM

At each of the 4 image acquisition sessions, patients were shown 30 positive and 30 negative morally tuned adjectives (eg, brave or vile), for a total of 60 words. Each word was shown visually for 1 second. The cognitive activation paradigm was based on a classic go/no-go task. Patients were asked to either push a button for positive targets and ignore negative distractions, or to do the opposite.

The experimental setup was composed of 2 positive target sessions and 2 negative target sessions, randomly presented. White words were randomly shown on a black screen and presented to the participant through a mirror positioned above the head coil. Emotionally tuned stimuli were interspersed by 1, 2, or 3 repetition times, with a 4:2:1 schedule, during which subjects were presented a cross-hair fixation. A 0.1-second temporal jittering was inserted to randomly present every word within the repetition times.

DATA PROCESSING

Images were computed, overlaid on anatomical images, and analyzed using Statistical Parametric Mapping–2 software (Wellcome Department of Imaging Neuroscience, Institute of Neurology and the National Hospital for Neurology and Neurosurgery; London, England).[19] Masking of white matter activations and conversion from the Montreal Neurologic Institute coordinates to the Talairach space were performed with PickAtlas software (Wake Forest School of Medicine, Winston-Salem, NC).[20]

We performed section timing on all acquired volumes to correct images for time acquisition between the first and last sections and realigned the scans to correct for head movement. Data were then normalized to a standard echo-planar imaging template volume based on the Montreal Neurological Institute reference brain[21] in the space of Talairach and Tournoux,[22] and smoothed using a 10-mm full-width at half-maximum isotropic gaussian kernel. The evoked hemodynamic responses were modeled as a delta function convolved with a hemodynamic response function and its temporal derivative within the context of the general linear model. All events were time locked to the onset of emotionally tuned words.

fMRI Data Analysis

At the individual level, we first compared (*t* test; threshold $P<.001$) both no-go and go trials to fixation, thereby isolating regions that were engaged by the task during both trial types. We then compared no-go and go images, thus creating double-subtraction images ([no-go > fixation] > [go > fixation]) at the single subject level (*t* test; threshold $P<.001$) that were used at the random-effects level.

The resulting 4 double-subtraction images for each subject (positive or negative and before or after treatment) were then entered into a second-level random-effect 3-way analysis of variance with moral valence of the stimuli, time, and response to treatment (final HDRS score <8) as factors. Second-level analyses were thresholded at $P<.05$ and limited to gray matter areas.

The primary analysis of interest was the 3-way interaction, which allowed us to identify the areas where treatment and response interacted in influencing the moral valence decision task; that is, the areas where the antidepressant therapy and its effects influenced the BOLD response to the task. In these same areas where the effect of the 3-way interaction was significant, we examined the 2-way interaction between SERTPR and moral valence of the stimuli at baseline. Since SERTPR has been shown to predict antidepressant response, this procedure allowed us to evaluate its baseline effects in the same areas influenced by treatment and response to treatment.

RESULTS

CLINICAL EFFECTS OF TREATMENT AND GENOTYPE

Changes in clinical outcome measures are given in **Table 1**. Treatment caused an overall significant decrease in HDRS scores (Friedmann analysis of variance: $\chi^2_3=23.18$; $P<.001$). Nine patients achieved the strict remission criterion of HDRS score less than 8 at day 7 and could be rated as full responders to treatment.

The *SERTPR* allele frequencies were in Hardy-Weinberg equilibrium and not different from those in the general population (long/long 5/20, 25%; long/short 10/20, 50%; and short/short 5/20, 25%). The *SERTPR* genotype significantly influenced the effects of treatment (**Figure 1**). At end point but not at baseline, homozygotes for the short allele had significantly worse HDRS scores (1-way analysis of variance: $F_{2,17}=4.50$; $P=.03$).

Perceived mood levels varied according to the overall depressive syndrome (**Figure 2**). Two-way repeated-

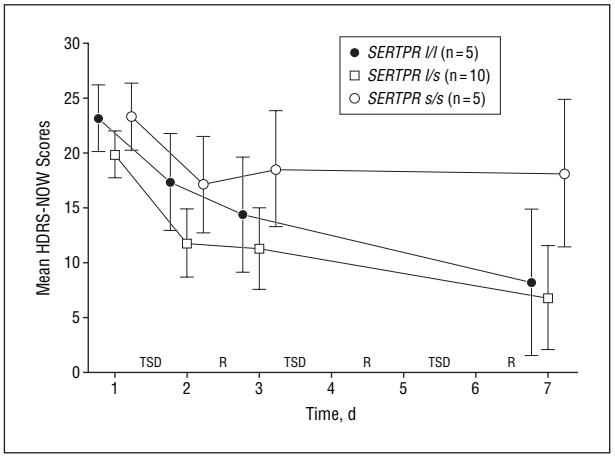

Figure 1. Pattern of change in depression severity (Hamilton Depression Rating Scale [HDRS] scores) during treatment in the 3 genotype groups. HDRS-NOW indicates a modified version of the 21-item HDRS from which items that could not be meaningfully rated because of the total sleep deprivation (TSD) procedure and the time frame were excluded (eg, weight changes and insomnia: items 4 through 6 and 16); *l/l* (long/long), polymorphism showing minimal activation for negative stimuli and maximal activation for positive stimuli; *l/s* (long/short), polymorphism showing intermediate values; R, rest or recovery night; SERTPR, serotonin transporter; *s/s* (short/short), polymorphism showing maximal activation for negative stimuli and minimal activation for positive stimuli; points, means; and whiskers, standard deviations.

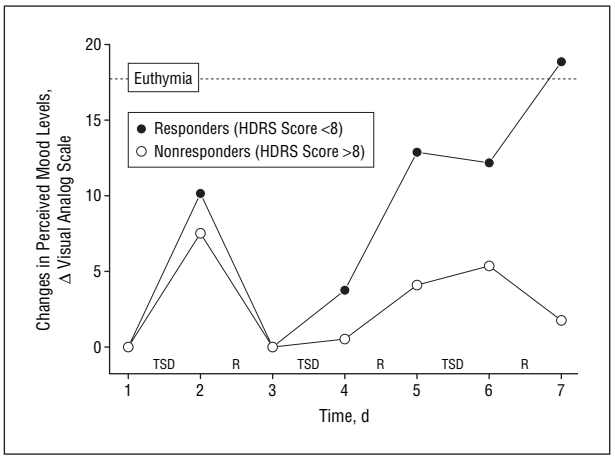

Figure 2. Patterns of change of perceived mood levels in respect to baseline (mean daily visual analog scale scores) in patients who did or did not respond to treatment (Hamilton Depression Rating Scale [HDRS] score <8 at day 7). R indicates rest or recovery night; TSD, total sleep deprivation; and dotted line, improvement corresponding to perceived euthymia.

measures analysis of variance with daily visual analog scale scores as dependent variables and response to treatment (HDRS score <8 at day 7) as an independent factor showed significant effects of treatment ($F_{6,108}$=2.23; P<.05) and response ($F_{1,18}$=10.36; P<.005). The behavioral results of the cognitive task showed that subjects performed the go/no-go task with a high level of accuracy both before and after treatment, with significantly shorter reaction times in responders.

fMRI Data

Treatment caused marked changes in the pattern of activation at the moral decision valence task. The areas where the main effect of valence (negative or positive) was significant at baseline and after treatment are given in **Table 2** and shown in **Figure 3**, and include the a priori ROIs ACC, dorsolateral PFC, and insula (see the introductory section). Figure 3 shows that maximal activations were detected in postrolandic regions at baseline (posterior cingulate cortex and insula) and in prerolandic regions after treatment (ACC and dorsolateral PFC). These changes in brain areas associated with the task could be related to several factors, such as changes in the depressive syndrome or changes in the cognitive strategy used by the subjects (learning effect). To identify the changes in brain activation patterns that were related to the clinical effect of treatment, we introduced the factor "response to treatment" and assessed the significance of its interaction with the effects of time and task.

We then investigated the brain volume for regions that showed a significant interaction of treatment (before and after), response to treatment (HDRS score <8), and moral valence of the stimuli (positive or negative). Gray matter areas where significant effects were detected are given in **Table 3** and shown in **Figure 4**A. This allowed us also to define functional ROIs where the BOLD response was influenced by clinical factors (treatment and response).

To determine whether the predictive value of *SERTPR* genotype could be correlated with a baseline influence on neural responses to the task, we investigated the brain volume for regions that showed a significant interaction of SERTPR polymorphism (*l/l, l/s*, and *s/s*) and moral valence of the stimuli (positive or negative) at baseline, taking into consideration only areas where the above 3-way interaction was significant. Voxels where significant effects were detected are given in **Table 4** and shown in Figure 4B. According to background hypotheses (see the introductory section), 2 of these voxels were located in our a priori ROIs, the right ACC (Brodmann area 24) and right dorsolateral PFC (Brodmann area 46).

Estimated regression coefficients (percent of whole-brain mean T2* BOLD signal) in these 2 voxels are plotted in **Figure 5** and demonstrate that in both areas the pattern of change before and after treatment differed according to the moral valence of the stimuli and to the clinical effect of treatment (response vs nonresponse). In particular, for negative stimuli, responders decreased activation in the ACC and increased activity in the dorsolateral PFC, whereas nonresponders showed the opposite pattern of change in both areas. For positive stimuli, responders decreased activation in both areas, and nonresponders decreased activity in the ACC and increased activity in the dorsolateral PFC.

In the same voxels, estimated regression coefficients as a function of *SERTPR* genotype are shown in **Figure 6**. In both the ACC and the dorsolateral PFC, baseline activations for negative and positive stimuli varied according to *SERTPR* genotype, with subjects with the *l/l* allelic variant showing minimal activation for negative stimuli and maximal activation for positive stimuli; whereas those with the *s/s* polymorphism exhibited the opposite pattern and heterozygotes with the *l/s* polymorphism demonstrated intermediate values.

Table 2. Gray Matter Areas Where a Significant Effect of the Moral Valence of the Stimuli (Negative-Positive) Was Detected at Baseline and After Treatment*

Region	Side	BA	Signal Peak	*z* Value	*P* Value
Baseline					
Dorsolateral prefrontal cortex					
Middle frontal gyrus	Left	10	–36, 47, 11	2.03	.02
Inferior frontal gyrus	Right	46	55, 28, 13	1.76	.04
Frontal lobe					
Paracentral lobule	Right	5	10, –38, 48	2.07	.02
	Left	5	–12, –32, 48	1.81	.04
Precentral gyrus	Left	4	51, –13, 41	2.13	.02
	Left	43	–61, –5, 11	1.79	.04
Anterior cingulate cortex	Left	32	–2, 19, 34	1.82	.03
	Right	32	8, 25, 32	2.12	.02
Posterior cingulate cortex	Left	24	–2, –13, 39	2.18	.02
	Right	24	4, –11, 41	2.11	.02
	Left	31	–14, –29, 42	1.97	.03
	Right	31	18, –29, 42	2.01	.02
Temporal cortex					
Insula	Left	13	–55, –32, 20	1.69	.045
	Right	13	38, 6, 9	1.70	.045
Transverse temporal gyrus	Right	41	51, –19, 10	2.34	.01
Parietal cortex					
Postcentral gyrus	Left	2	–55, –18, 32	2.29	.01
	Left	40	–55, –22, 16	2.08	.02
	Right	3	24, – 31, 48	1.69	.046
Inferior parietal lobule	Left	40	–65, –34, 26	1.79	.04
After treatment					
Dorsolateral prefrontal cortex					
Middle frontal gyrus	Left	9	–2, 44, 24	2.68	.004
	Right	9	51, 13, 31	2.02	.02
Inferior frontal gyrus	Left	9	–48, 17, 23	1.74	.04
	Right	46	42, 36, 11	1.82	.04
Medial frontal gyrus	Right	8	2, 43, 40	1.94	.03
Anterior cingulate cortex	Left	32	–2, 21, 34	2.52	.006
	Right	32	4, 36, 20	2.23	.02
Posterior cingulate cortex	Left	30	–2, –49, 21	1.76	.04
Temporal cortex					
Insula	Right		44, 16, 1	1.70	.045
Middle temporal gyrus	Right	39	51, –61, 25	1.69	.046
Parietal cortex					
Inferior parietal lobule	Left	40	–57, –42, 44	1.83	.04
Precuneus	Left	31	–6, –47, 30	1.68	.047
Thalamus					
Medial dorsal nucleus	Left	–	–6, –9, 8	1.96	.03
Caudate nucleus, head	Right	NA	14, 16, 1	2.08	.02

Abbreviations: BA, Brodmann area; –, data not collected.
*Data are shown for the maximal activations for each Brodmann area: side (left or right), Talairach coordinates (x, y, z) of voxels with higher *z* values (signal peaks), and level of significance. Glass-brain images of these data are shown in Figure 3.

COMMENT

Chronotherapeutic treatment (TSD plus LT) of bipolar depression resulted in changes in neural response to a moral valence decision task that paralleled clinical amelioration of depressive symptoms. Both clinical and neural responses were influenced by the SERTPR polymorphism.

To our knowledge, this is the first study with the purpose of defining the interactions of multiple factors known to affect or to be affected at the individual level by major depressive episodes in the course of bipolar disorder, including the following: neuropsychological processing of morally tuned information, which is known to bias toward negative[4]; fMRI study of BOLD response to morally tuned information, which is known

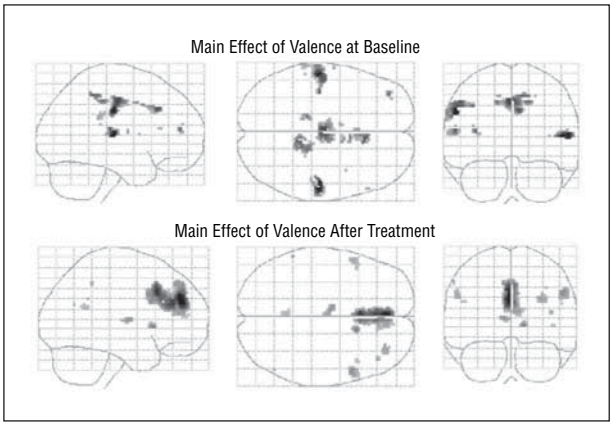

Figure 3. Glass-brain images of gray matter areas in which a significant effect of moral valence (negative or positive) was detected at baseline and after treatment.

Table 3. Gray Matter Areas Showing a Significant Interaction of Treatment, Response to Treatment, and Moral Valence of the Stimuli*

Region	Side	BA	Signal Peak	*z* Value	*P* Value
Dorsolateral prefrontal cortex					
Middle frontal gyrus	Left	10	–28, 51, 20	2.20	.02
	Right	10	42, 42, 20	1.95	.03
	Right	46	51, 32, 15	2.52	.006
Inferior frontal gyrus	Left	44	–59, 16, 14	2.66	.004
Cingulate cortex	Left	32	–2, 41, 11	1.66	.048
	Right	24	6, 1, 28	2.03	.02
Temporal cortex					
Insula	Right	13	46, –15, 17	1.72	.04
Supramarginal gyrus	Left	40	–53, –47, 24	2.86	.002
Superior temporal gyrus	Right	41	36, –30, 14	1.79	.04
Frontal cortex					
Precentral gyrus	Left	4	–49, –12, 39	1.90	.03
	Left	6	–36,–2, 35	2.02	.02
	Right	6	63, –10, 35	1.72	.04
	Left	43	–53, –11, 13	1.83	.03
	Left	44	–53, 10, 12	2.70	.003
Parietal cortex					
Postcentral gyrus	Right	2	57, –27, 44	1.87	.03
Precuneus	Right	19	34, –78, 39	2.91	.002
	Right	19	24, –79, 41	2.18	.02
	Left	31	–2, –51, 32	1.88	.03
	Left	39	–35, –62, 34	1.79	.04
Inferior parietal lobule	Left	40	–61, –33, 40	2.14	.02
	Right	40	57, –42, 44	2.75	.003
Caudate nucleus					
Tail	Left	–	–20, –32, 16	2.47	.007
Body	Right	–	10, 3, 15	1.76	.04

Abbreviations: BA, Brodmann area; –, data not collected.
*Data are shown for the maximal activations for each Brodmann area: side (left or right), Talairach coordinates (x, y, z) of voxels with higher *z* values (signal peaks), and level of significance. Glass-brain images of these data are shown in Figure 4A.

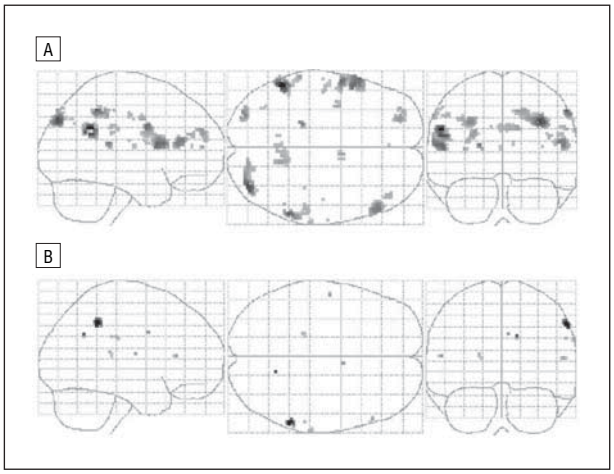

Figure 4. Glass-brain images of gray matter areas in which a significant interaction of valence × treatment × response was detected (A) and, within the same areas, voxels in which a significant interaction of valence × genotype was detected at baseline (B).

to provide sound correlates of this cognitive distortion[5,6]; chronotherapeutic antidepressant therapy (TSD plus LT), which is known to produce rapid and sustained antidepressant effects involving the entire depressive syndrome, and including the information processing biases[1,2]; and the SERTPR polymorphism, which is known both to predict response to multiple antidepressant treatments, including chronotherapeutic therapy of bipolar depression[1,11] and to influence the processing of affective stimuli.[16,17]

The main finding of the present study is that all of these factors showed significant interactions in ROIs pointed out in previous studies in the field: the ACC and the dorsolateral PFC.[7,8,17] Neural responses in the right dorsolateral PFC to negative stimuli have been shown to be linked to voluntary suppression of sadness in healthy subjects,[23] and positron emission tomography scans showed that local metabolism was decreased by induction of transient sadness in healthy subjects[24] and activated by response to treatment in patients with depression.[25] We found that response to antidepressant therapy is paralleled by increased activation of negative stimuli and decreased activation of positive stimuli and that baseline activity in this area varied according to *SERTPR* genotype. This finding supports the hypothesis that the right dorsolateral PFC is involved in the control of sadness in normal and pathologic conditions, with the condition of normality involving activations for negative stimuli.

In healthy subjects, the ACC is widely implicated in the detection of unfavorable outcomes, response errors, response conflict, and decision uncertainty.[26] A wide and consistent literature on positron emission tomography measures of metabolic activity in the perigenual ACC in major depression at baseline and after recovery showed

Table 4. Gray Matter Areas Showing a Significant Interaction of *SERTPR* Genotype and Moral Valence of the Stimuli*

Region	Side	BA	Signal Peak	*z* Value	*P* Value
Dorsolateral prefrontal cortex					
Inferior frontal gyrus	Right	46	55, 28, 10	1.87	.03
Cingulate cortex	Right	24	6, 4, 31	1.92	.03
Frontal cortex					
Precentral gyrus	Left	43	–53, –9, 13	1.90	.03
Parietal cortex					
Precuneus	Right	31	14, –55, 32	2.24	.01
Inferior parietal lobule	Right	40	57, –43, 43	2.53	.006
Caudate nucleus, tail	Left	–	–20, –32, 16	1.78	.04

Abbreviations: BA, Brodmann area; SERTPR, serotonin transporter; –, data not collected.
*The analysis has been limited to the voxels where a significant interaction of treatment, response to treatment, and valence was detected (Table 2). Data are shown for the maximal activations for each Brodmann area: side (left or right), Talairach coordinates (x, y, z) of voxels with higher *z* values (signal peaks), and level of significance. Glass-brain images of these data are shown in Figure 4B.

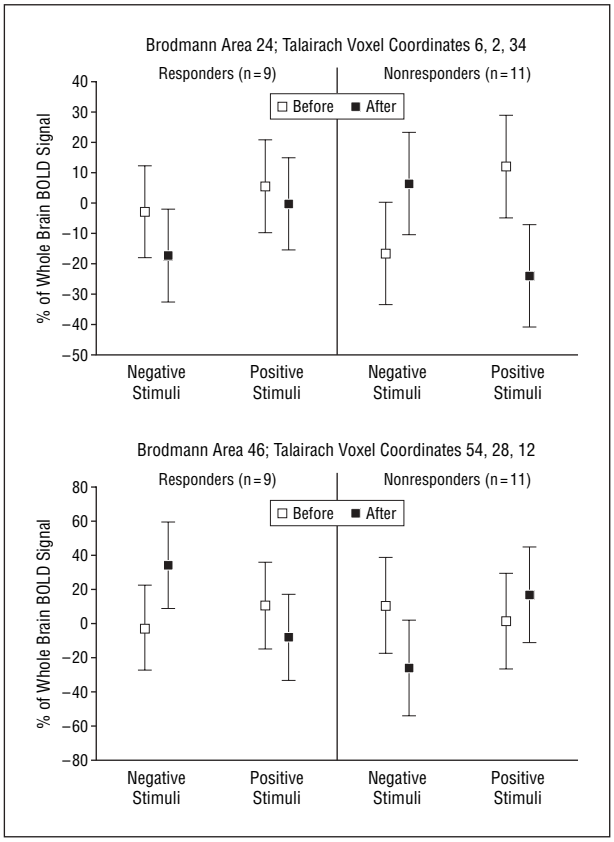

Figure 5. Direction and size effects of the significant interactions of treatment, response to treatment, and moral valence of the stimuli on the event-related blood oxygen level–dependent (BOLD) activations in the right anterior cingulate cortex (Brodmann area 24; Talairach voxel coordinates 6, 2, 34) and right dorsolateral prefrontal cortex (Brodmann area 46; Talairach voxel coordinates 54, 28, 12). Points indicate estimated regression coefficients for the tasks (percent of whole brain mean T2* BOLD signal) before and after treatment; whiskers, standard errors.

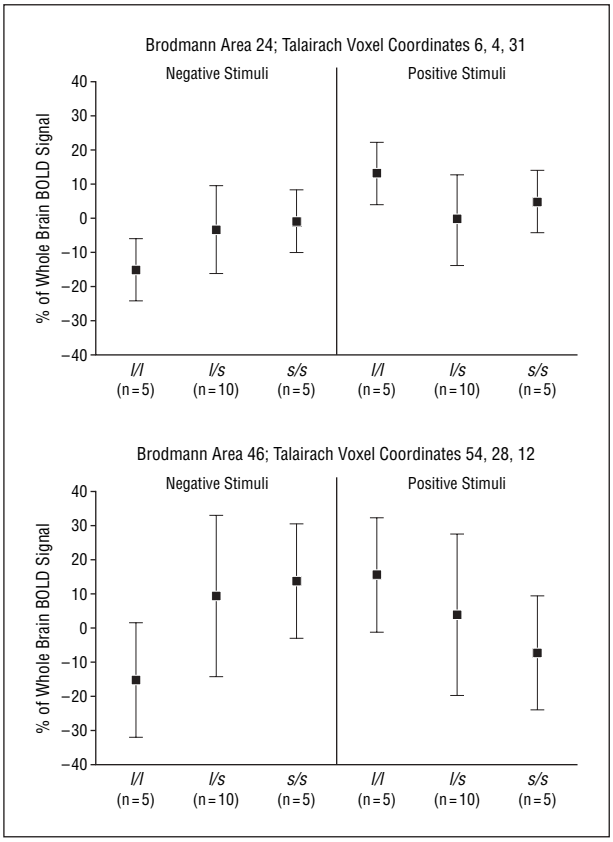

Figure 6. Direction and size effects of the significant interactions of the serotonin transporter polymorphism and moral valence of the stimuli in right anterior cingulate cortex (Brodmann area 24; Talairach voxel coordinates 6, 4, 31) and right dorsolateral prefrontal cortex (Brodmann area 46; Talairach voxel coordinates 54, 28, 12). Points indicate estimated regression coefficients for the tasks (percent of whole-brain mean T2*-weighted blood oxygen level–dependent [BOLD] signal) before and after treatment; whiskers, standard errors, *l/l* (long/long), polymorphism showing minimal activation for negative stimuli and maximal activation for positive stimuli; *l/s* (long/short), polymorphism showing intermediate values; and *s/s* (short/short), polymorphism showing maximal activation for negative stimuli and minimal activation for positive stimuli.

higher metabolic rates at baseline, with a decrease during treatment that was proportional to the clinical amelioration[27] both in response to pharmacologic treatment and to TSD. In particular, 4 different groups with a total of 5 published studies on TSD effects with positron emission tomography or hexamethylpropyleneamine oxime–single-photon emission computed tomography reported that responders had increased relative localized metabolic activity in the general location of the ventral ACC compared with nonresponders and healthy control subjects at baseline.[9] Structural ACC abnormalities

have been detected in patients affected by mood disorders, thus allowing a tentative definition of an anatomical endophenotype of bipolar disorder.[28] In the only study of BOLD neural responses to emotional stimuli in depressed patients before and after antidepressant therapy, neural responses to negative stimuli in the rostral ACC increased with antidepressant response to venlafaxine and their baseline intensity correlated with response.[8] In the present study, we showed that the baseline activation in the dorsal ACC are influenced by SERTPR and that response to treatment is paralleled in the same voxel by decreased activation for negative stimuli. These data confirm the high relevance of activity in this region as a biologic correlate of clinical antidepressant response and also that the ACC is a complex structure with different anatomical regions that show different patterns of activation in response to emotional stimuli.[17,29] Our results support the need for further studies linking these anatomical and functional characteristics with health, depression, and antidepressant response.

The moral valence of the stimuli had different effects on BOLD neural response at baseline and after treatment (Figure 3). Maximal activation was detected at baseline in the postrolandic cingulate cortex and after treatment in the prerolandic ACC. Since the interaction of response to treatment did not influence activity in the posterior cingulate cortex (Table 2), further study is needed to clarify whether these changes in the activation of cingulate cortex reflected changes in the clinical status or changes in the cognitive strategy used by the subjects and were the result of learning effects or aspecific effects of the TSD plus LT treatment. The posterior cingulate cortex showed higher responses to emotional than to neutral stimuli[5] and has been involved in self-referential processing[30] and in the experience of grief.[31]

Not all of the neural responses that were clinically relevant (ie, showed a significant interaction of the response status with factors related to the moral valence decision task) were influenced by the *SERTPR* genotype. The interaction of treatment, response to treatment, and moral valence of the stimuli, but not of the *SERTPR*, was significant in the left anterior dorsolateral PFC (Brodmann area 10). Neural responses in the left dorsolateral PFC have been shown to be linked to self-referential but not other referential processing of emotional words in healthy subjects[30] and to the execution of a moral valence decision task in healthy subjects.[5]

The same interaction of all factors except for SERTPR was significant in the insular cortex (Table 2 and Table 3). Previous findings have shown that, during antidepressant treatment with venlafaxine, increased BOLD responses to negative stimuli in the left insular cortex were observed.[8] Insular cortex activation has been shown to be sensitive to the negative salience of the stimuli,[32] with higher responses during transient induction of sadness,[23] and in a wide range of negative experiences, including the experience of guilt,[33] in healthy and psychiatric populations.[34]

The results of the present study are consistent with proposed models of gene-modulated interactions between the ACC, dorsolateral PFC, limbic system, serotonergic nuclei, and environmental stimuli in the generation and control of depressed mood.[24,29,35,36] We have confirmed that the interaction of these multiple factors is significant in a priori predicted ROIs, and we found that the direction of the interaction varies according to both the individual genotype and the response to treatment.

CONCLUSIONS

We replicated and extended previous findings about the neural correlates of mood-congruent biases in depressed information processing[7] and about the neural correlates of antidepressant response,[8] and linked them with ongoing research on chronotherapeutic treatment of bipolar depression[1] and genetic predictors of antidepressant response.[11] The pioneering nature of our findings does not allow us to draw hypotheses on the possible pathogenetic value of the exploited interactions, but, when replicated in a larger sample, they suggest the possibility of defining sound biologic correlates of the recovery process from bipolar depression. The clinical interest for such correlates is warranted because this could be a first step toward a neurobiologic method of assessment of depressive illness and the response to antidepressant therapy.

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