Prefrontal Thinning Affects Functional Connectivity and Regional Homogeneity of the Anterior Cingulate Cortex in Depression

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Major depressive disorder (MDD) is associated with structural and functional alterations in the prefrontal cortex (PFC) and anterior cingulate cortex (ACC). Enhanced ACC activity at rest (measured using various imaging methodologies) is found in treatment-responsive patients and is hypothesized to bolster treatment response by fostering adaptive rumination. However, whether structural changes influence functional coupling between fronto-cingulate regions and ACC regional homogeneity (ReHo) and whether these functional changes are related to levels of adaptive rumination and treatment response is still unclear. Cortical thickness and ReHo maps were calculated in 21 unmedicated depressed patients and 35 healthy controls. Regions with reduced cortical thickness defined the seeds for the subsequent functional connectivity (FC) analyses. Patients completed the Response Style Questionnaire, which provided a measure of adaptive rumination associated with better response to psychotherapy. Compared with controls, depressed patients showed thinning of the right anterior PFC, increased prefrontal connectivity with the supragenual ACC (suACC), and higher ReHo in the suACC. The suACC clusters of increased ReHo and FC spatially overlapped. In depressed patients, suACC ReHo scores positively correlated with PFC thickness and with FC strength. Moreover, stronger fronto-cingulate connectivity was related to higher levels of adaptive rumination. Greater suACC ReHo and connectivity with the right anterior PFC seem to foster adaptive forms of self-referential processing associated with better response to psychotherapy, whereas prefrontal thinning impairs the ability of depressed patients to engage the suACC during a major depressive episode. Bolstering the function of the suACC may represent a potential target for treatment.
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INTRODUCTION

Major depressive disorder (MDD) is a severe psychiatric illness with a lifetime risk of 10–20% in the general population (Kessler *et al*, 2003). Recent meta-analyses have reported consistent hyper-activity of the ventromedial prefrontal cortex/anterior cingulate cortex (VMPFC/ACC) at rest (Kuhn and Gallinat, 2013) and reduced grey matter volume in the ACC and the dorsolateral and dorsomedial

prefrontal cortex (DLPFC and DMPFC, respectively) in depressed patients (Bora *et al*, 2012; Koolschijn *et al*, 2009; Lai, 2013). Recently, the DLPFC has been shown to have a causal role in regulating medial PFC/ACC connectivity. Using transcranial magnetic stimulation in healthy subjects, Chen *et al*, (2013a) demonstrated that suppression of the right DLPFC induced negative connectivity of the default mode network (DMN), and this effect is primarily mediated through the medial PFC/ACC component of the DMN. Moreover, thinning of the DMPFC in medicated depressed patients has been associated with increased functional connectivity (FC) with the DLPFC and the ventrolateral PFC/insula as well as decreased FC within parietal and temporal regions (van Tol *et al*, 2013). These results suggest that structural alterations in the DLPFC, the medial PFC, and/or the ACC in MDD may impact their functional coupling.

A better understanding of the structural–functional alterations related to the DLPFC and the medial PFC/ACC is particularly relevant to MDD, because these regions may have a central role not only in the development of a major depressive episode but also in response to treatment. Specifically, greater ACC volume is related to better clinical outcome (Chen et al, 2007; Frodl et al, 2008), and higher ACC activity at rest (measured using various imaging methodologies) is consistently associated with better treatment response. As the ACC is part of the DMN, a network engaged at rest and during self-referential processing tasks (Buckner et al, 2008), these findings have led to the hypothesis that increased ACC activity may foster adaptive self-referential processing that help promote recovery (Pizzagalli, 2011).

The use of resting-state functional magnetic resonance imaging (RS-fMRI) is a valuable strategy to relate structural alterations to functional abnormalities in specific brain networks (van Tol et al, 2013). FC provides information about the functional interactions between brain regions, but RS-fMRI can also be used to investigate spontaneous regional brain function at rest. Regional homogeneity (ReHo) measures the degree of regional synchronization of fMRI time courses. ReHo changes reflect changes in the temporal aspects of neural activity, providing information about local alterations in brain function (Chao-Gan and Yu-Feng, 2010). ReHo alterations in depressed patients have been identified in various brain regions, including the ACC (eg, Yao et al, 2009), and have been shown to correlate with symptom's severity (Wu et al, 2011). These findings show that, similarly to other imaging methodologies, functional alterations of the ACC can also been identified using ReHo in MDD. Moreover, ReHo changes in the DMPFC have been found in response to pharmacological treatment in MDD (Lai and Wu, 2012; Wang et al, 2014). However, whether alterations in ACC ReHo prior to treatment are related to adaptive rumination or treatment response has not been reported yet.

The goals of this study were to better understand how grey matter alterations in fronto-cingulate regions affect their functional coupling and how these changes are related to altered ACC ReHo at rest and adaptive rumination in MDD. As adaptive rumination is considered to foster treatment response, these results may increase our understanding of how structural and functional changes in fronto-cingulate regions influence treatment response in MDD. To this aim, we collected imaging data and rumination scores in unmedicated depressed patients prior to the onset of 22 weekly sessions of cognitive behavioral therapy (CBT). The Response Style Questionnaire provided two measures of rumination: symptom-focused and self-focused (Kühner et al, 2007). The latter is associated with adaptive coping strategies and lower depression severity over time (Burwell and Shirk, 2007; Treynor et al, 2003). Thus we investigated (1) whether higher self-focused rumination was associated with better CBT response in MDD and represented a form of adaptive rumination; (2) whether structural changes in the DLPFC, the medial PFC, and the ACC were present in MDD and were associated with altered FC and increased ACC ReHo; and (3) whether these functional alterations were associated with self-focused rumination.

MATERIALS AND METHODS

Participants

Twenty-six unmedicated patients with MDD and 39 healthy controls without a psychiatric, neurological, or medical illness completed the study. All participants were Caucasian and right-handed. The groups were not significantly different in gender distribution, age, or years of education (Table 1 and Supplementary Table S1). Two patients had stopped medications 6 weeks prior to the study. All other patients had been free from medications for at least 2 years. The exclusion criteria for both groups included age <18 or >65 years, current or past psychosis or mania, major medical or neurological illness, current drug or alcohol abuse, and MRI contraindications (assessed by an MRI safety questionnaire). Inclusion in the MDD group was contingent on a diagnosis of current MDD based on a Structured Clinical Interview for DSM-IV Axis-I Disorders (SCID-I) semi-structured interview. Ten subjects with MDD had comorbid anxiety disorders: three subjects had social phobia (one of whom also had an eating disorder), one subject had specific phobia, three subjects had generalized anxiety disorder, and three subjects had panic disorder. Control subjects did not have a lifetime history of MDD, had no history of MDD in first-degree relatives, and were currently free of all Axis-I disorders based on a SCID-I interview.

A manualized version of CBT was offered as a research study separate from routine care in the outpatient clinic of the University's psychology department. Imaging data were collected prior to the onset of their 22 weekly individual therapy treatment sessions.

The study was approved by the University of Zurich's Institutional Review Board, and all subjects provided written informed consent for participation in the study and were paid a modest compensation.

Psychometric Measures

Depression severity in patients was assessed with the Beck Depression Inventory-II (BDI-II; Beck et al, 1996) and the Inventory for Depressive Symptomatology (Helmreich et al, 2011) prior to the initiation of psychotherapy and after the last psychotherapy session. The patients also completed the Response Style Questionnaire (Kühner et al, 2007, see Supplementary Information). All participants completed the Hopelessness scale (Krampen, 1994), the State-Trait Anxiety Inventory (Laux et al, 1981) and the Snaith–Hamilton-Pleasure Scale (Franz et al, 1998).

Magnetic Resonance Image Acquisition

Images were acquired on a Philips Achieva 3-Tesla whole-body MRI unit equipped with an eight-channel head coil that used a sensitivity-encoded single-shot echo-planar sequence (acceleration factor $R = 2$). A T1-weighted gradient echo sequence (turbo field echo) with a spatial resolution of $0.94 \times 0.94 \times 1.00 \, \text{mm}^3$ (matrix: 240×240 pixels; 160 slices), field of view $= 240 \times 240 \, \text{mm}^2$, TE $= 3.7$ ms, TR $= 8.06$ ms, and flip angle $= 8°$ was applied. For the acquisition of the functional time series, the subjects were told to lie still in the scanner with their eyes closed and let their minds

Table 1 Demographic and Symptom Severity for Control and Depressed Participants Included in the Resting-State Analysis

	Depressed patients $N=21$	Healthy controls $N=35$	p-Values
Gender (%)	10 females (48%)	20 females (57%)	$p>0.4$
Age in years (mean ± SD)	36.6 ± 12.3	32.7 ± 11.5	$p>0.2$
Age in men (mean ± SD), years	41.4 ± 12.9	35.0 ± 12.7	$p>0.2$
Age in women (mean ± SD), years	31.5 ± 9.4	31.1 ± 10.4	$p>0.9$
Years of education (mean ± SD)	16.6 ± 2.8	15.8 ± 2.6	$p>0.2$
BDI (mean ± SD)	26.4 ± 9.3	—	
IDS (mean ± SD)	33.9 ± 9.5	—	
Single MD episode (%)	6 (29%)	—	
Medication naive (%)	16 (76%)	—	
Comorbid anxiety disorder (%)	8 (38%)	—	
SHAPS (mean ± SD)	4.2 ± 3.1	0.4 ± 0.9	$t(54) = -6.9; p<0.001$
H-Scale (mean ± SD)	75.9 ± 13.9	41.7 ± 10.1	$t(54) = -10.6; p<0.001$
STAI-Trait (mean ± SD)	57.3 ± 9.6	31.2 ± 7.2	$t(54) = -11.5; p<0.001$
RSQ Self-focused (mean ± SD)	19.1 ± 4.5		
RSQ Symptom-focused (mean ± SD)	19.6 ± 4.6		
Mean displacement (mean ± SD)	0.05 ± 0.04	0.04 ± 0.03	$p>0.1$

Abbreviations: BDI, Beck depression inventory; H-Scale, Hopelessness Scale; IDS, Inventory of Depressive Symptomatology; RSQ, Response Style Questionnaire; SHAPS, Snaith–Hamilton-Pleasure Scale; STAI, Spielberger State-Trait Anxiety Inventory.
p-Values for between-group t-test or chi-squared analyses are presented in the final column.

wander (Logothetis et al, 2009; Northoff et al, 2010 but see also Raichle et al, 2001); 300 functional images were collected in a 10-min run. Thirty-six contiguous axial slices were placed along the anterior–posterior commissure plane covering the entire brain. The following parameters were used: TR = 2000 ms, TE = 30 ms, flip angle = 75°, ascending acquisition order, 80×80 voxel matrix, and voxel size = $3 \times 3 \times 4 \, \text{mm}^3$.

Surface-Based Morphometry Preprocessing

Structural T1-weighted images were analyzed using Free-Surfer (version 5.1.0, http://surfer.nmr.mgh.harvard.edu/fswiki) to create anatomical surface models and to perform statistical analyses (Dale et al, 1999; Fischl et al, 1999). For each subject, the processing stream included the removal of non-brain tissue, transformation to Talairach space, and segmentation of gray matter (GM)–white matter (WM) tissue. The thickness measurements across the cortex were computed by identifying the point on the GM–WM boundary surface that was closest to a given point on the estimated pial surface (and vice versa) and averaging these two values (Fischl and Dale, 2000). To map each subject to a common space, the surface that represented the GM–WM border was registered to an average cortical surface atlas using a nonlinear procedure that optimally aligned sulcal and gyral features across subjects (Fischl et al, 1999).

Resting-State Data Preprocessing

RS-fMRI data were analyzed using the Data Processing Assistant for Resting-State fMRI Advanced edition version 2.2 (http://www.rfmri.org/). The first 10 volumes were removed, and the remaining 290 volumes were slice-time corrected. T1-weighted images were co-registered to the mean functional MRI data for each subject and segmented into GM, WM, and cerebrospinal fluid (CSF) probability maps using a Diffeomorphic Anatomical Registration Through Exponentiated Lie algebra approach (Ashburner, 2007). All GM, WM, and CSF images were resampled to $3 \times 3 \times 3 \, \text{mm}^3$ and spatially normalized to the MNI space using DARTEL deformation parameters. The deformation field was applied to the functional data. The global mean, WM, and CSF signals were included in the model to remove the effects of physiological noise (Yan et al, 2013). The Friston 24-parameter model (ie, 6 head motion parameters, 6 head motion parameters one time point before, and the 12 corresponding squared items) was used to regress out head motion effects (Friston et al, 1996). Only for the FC analysis, data were smoothed using a 4-mm full-width-at-half-maximum (FWHM) Gaussian kernel. All functional data were filtered by a Fast-Fourier-Transform/band-pass filter (0.01–0.08 Hz) to eliminate low-frequency-fluctuations, and correlation coefficients were transformed to z-scores to better satisfy normality. Four controls and five patients were excluded from the group analyses because of motion $\geqslant 1.5 \, \text{mm}$ translations and $1.5°$ rotations based on a single temporal frame (Liao et al, 2010; Liu et al, 2013).

Seed-Based FC

The regions with reduced cortical thickness defined the seeds for subsequent FC analyses. For each ROI, the blood-oxygenation-level-dependent (BOLD) fMRI time course was extracted, and the correlation coefficient between the average time course of the seed region and the time courses of all other voxels in the brain was calculated to generate the seed-based FC map.

Regional Homogeneity

The Kendall's coefficient of concordance of each voxel was calculated with its nearest neighbors (26 voxels) in a voxel-wise analysis (Chao-Gan and Yu-Feng, 2010; Zuo *et al*, 2010) as a ReHo measure.

Statistical Analyses

Demographic and psychometric data were analyzed with unpaired *t*-tests, and gender was analyzed with a Chi-squared test using StatView 5.0.1 (SAS Institute, Inc, Cary, NC). Significance was set at $p < 0.05$ two-tailed alternatives.

As a measure of psychotherapy response independent of initial depression severity, standardized residual scores were calculated from a regression of pretherapy BDI-II scores on posttherapy scores (BDI-II$_{POST}$ (Siegle *et al*, 2006); Supplementary Information). To assess correlations with self-focused rumination independent of depression severity, standardized residual scores were calculated from a regression of pretherapy BDI-II scores on self-focused rumination scores (residuals of self-focused rumination).

To detect local differences in cortical thickness between the patients with MDD and the healthy subjects, a smoothing of 10-mm FWHM kernel was applied. Vertex-wise analyses were computed using a general linear model with an initial cluster forming height threshold of $p < 0.05$ fully corrected for multiple comparisons using 5000 synthetic *z*-score permutations (Monte Carlo simulations) on the cluster extent, while simultaneously controlling for global mean cortical thickness, age, and gender.

Group differences in FC and ReHo were analyzed using a two-sample *t*-test, with age and gender as covariates in SPM8/r4290. For the ReHo analyses, a mask of the cingulate cortex was created using SPM-toolbox/wfupickatlas/aal and including the ACC and midcingulate cortex. In the depressed patients, associations between the FC and residual of self-focused rumination scores were calculated using a multiple regression analysis, with age and gender as covariates. Unless otherwise specified, clusters were identified with a global height threshold of $p < 0.001$ uncorrected and a spatial extent cluster size to achieve a family-wise-error corrected statistical threshold of $p < 0.05$ (Friston *et al*, 1994). Mean *z*-scores within the functional cluster were extracted using MarsBaR from the SPM-toolbox.

Correlations between the functional data and psychometric scores were calculated using Pearson correlation coefficient, whereas correlations with cortical thickness were calculated using the Kendall's tau correlation coefficient (van Tol *et al*, 2013). The Kendall's tau correlation coefficient quantifies the difference between the percentage of concordant and discordant pairs, and it is considered more accurate than the Spearman's rho for small sample sizes of non-normally distributed data. Because of their potential relevance, we report correlation results without correcting for multiple comparisons. Results that would not survive correction are referred as marginal. Correlation coefficients between groups were compared using Fisher's *r*-to-*z* transform (http://vassarstats.net/rdiff.html).

RESULTS

Demographic and Psychometric Data

The demographic and clinical characteristics of the participants are summarized in Table 1 and Supplementary Table S1. There was no significant difference between groups in age, gender, years of education, or frame-wise displacement during the acquisition of RS-fMRI data (Van Dijk *et al*, 2012).

In the depressed patients, the residual scores of self-focused rumination (Res-SeRumination) positively correlated with the residual scores of the BDI-II$_{POST}$ (Res-BDI-II$_{POST}$; $p < 0.04$, $r^2 = 0.23$; Supplementary Figure S1). Two patients did not complete treatment; thustheir Res-BDI-II$_{POST}$ score could not be correlated with Res-SeRumination.

Structural Data

Surface-based morphometry identified one cluster of reduced cortical thickness in patients with MDD compared with the controls in the right rostral middle frontal cortex (Figure 1a, MNI coordinates $x = 36$, $y = 52$, $z = 18$, cluster-wise $p < 0.02$, Max-logP $= 3.5$, size $= 880$ mm^2, number of vertices $= 1094$). No significant group differences were found in the total GM, WM, or CSF volumes (Supplementary Table S3).

Functional Connectivity

The rostral middle frontal cortex cluster identified in the surface-based morphometric analysis showed increased FC with the supragenual ACC (suACC) in the depressed patients compared with the healthy controls (Table 2, Figure 1b). The FC maps for each group are reported separately in Supplementary Figure S2.

When the correlation between the FC connectivity strength (*z*-score) and the cortical thickness was investigated, a marginal positive correlation was identified in the healthy controls ($r_{Kendall's\ tau} = 0.25$, $p < 0.04$) but not in the depressed patients ($r_{Kendall's\ tau} = 0.21$; Supplementary Figure S3A). The correlation coefficients were not significantly different between groups ($p > 0.8$).

In the depressed patients, the FC strength was positively correlated with Res-SeRumination ($p < 0.005$, $r^2 = 0.36$; Figure 1c) and was also marginally negatively correlated with Res-BDI$_{POST}$ ($p < 0.03$, $r^2 = 0.26$; Supplementary Figure S3B).

Regional Homogeneity

The small-volume-corrected analysis identified increased ReHo of the suACC in the depressed patients compared with the healthy controls ($p < 0.005$, $t = 4.19$, 17 voxels, $x = 3$, $y = 36$, $z = 24$, Figure 2a; at the whole brain level, this finding was approximately significant ($p = 0.052_{FDR-corrected}$), and no other significant cluster was found).

Because the cluster of increased ReHo spatially overlapped with the cluster of increased FC previously discussed, we assessed whether ReHo was associated with cortical thickness and FC strength. We determined that ReHo *z*-scores were positively correlated with the FC
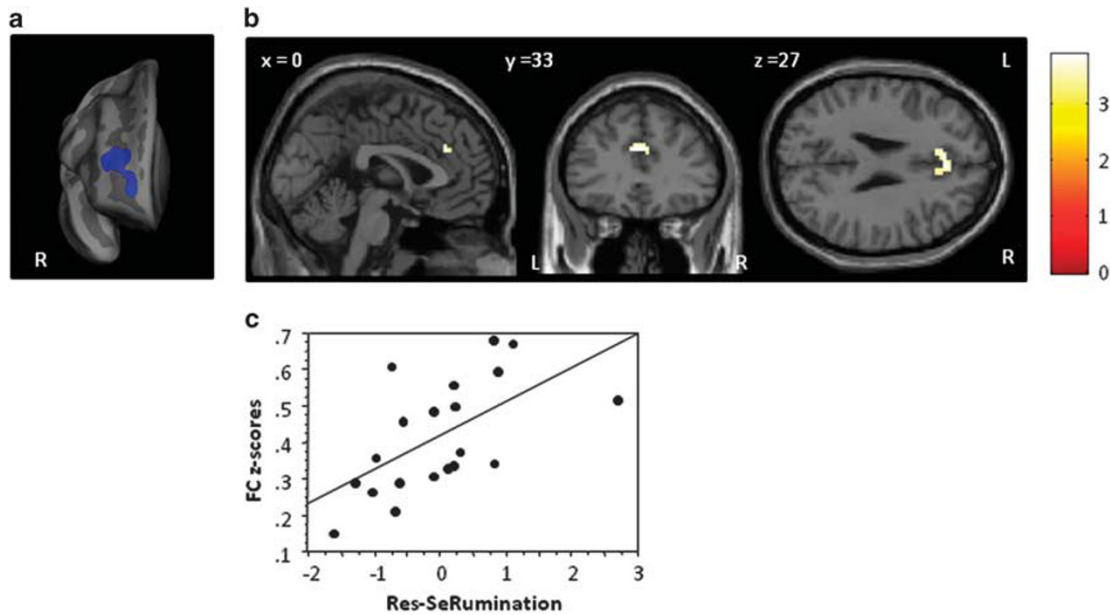
Figure 1 Brain regions showing structural and functional connectivity (FC) differences in patients with major depressive disorder (MDD) and healthy controls. (a) Cortical thinning in patients with MDD compared with healthy controls was identified in the right rostral middle frontal cortex. The cluster is rendered in blue on the inflated surface after correction for multiple comparisons. (b) Depressed patients exhibited increased functional connectivity of the right rostral middle frontal cortex with the supragenual anterior cingulate cortex (suACC) compared with healthy controls. (c) Scatterplot shows the correlation between FC strength of the right rostral middle frontal cortex with the suACC (z-scores) and the standardized residual scores of self-focus rumination in depressed patients (solid line; $r^2 = 0.36$).

Table 2 Functional Connectivity of the Right Rostral Middle Frontal Cortex in Patients with MDD and Healthy Controls

Cluster (voxels)	T (Peak)	p-Values	Region	x	y	z	Hem
HC > MDD							
None							
MDD > HC							
41	3.89	$< 0.03_{\text{— FWE-corrected}}$	Anterior cingulum	-12	27	30	L
			Anterior cingulum	0	33	27	R/L
			Anterior cingulum	12	36	21	R

Abbreviations: Hem, hemisphere; L, left; R, right.

strength between the middle frontal cortex and the suACC in the depressed patients ($p < 0.003$, $r^2 = 0.39$). This correlation was only marginal in the healthy controls ($p < 0.05$, $r^2 = 0.12$; Figure 2b) and was not significantly different from patients ($p > 0.3$).

Moreover, in the depressed patients, the ReHo z-scores also showed a significant correlation with the cortical thickness (Supplementary Figure S3A; $p < 0.006$, $r_{\text{Kendall's tau}} = 0.45$; healthy controls: $r_{\text{Kendall's tau}} = 0.15$) and a marginal correlation with Res-SeRumination ($p < 0.04$, $r^2 = 0.2$; Supplementary Figure S3B). The correlation coefficients between ReHo and thickness were not significantly different between groups ($p > 0.2$).

As differences in anxiety levels between patients and controls during the imaging session could influence the functional results, FC and ReHo analyses were also conducted by including STAI-State as covariate of no interest in the model. However, similar results were found whether STAI-State was included or not (Supplementary Results).

DISCUSSION

Structural changes in the DLPFC and the medial PFC/ACC have been consistently identified in MDD, but it remains unclear whether these structural changes affect functional coupling. A better understanding of structural–functional alterations in these fronto-cingulate regions is important, because higher ACC activity at rest is found in treatment-responsive patients and is considered to foster recovery from a major depressive episode by supporting adaptive forms of rumination (Pizzagalli, 2011).
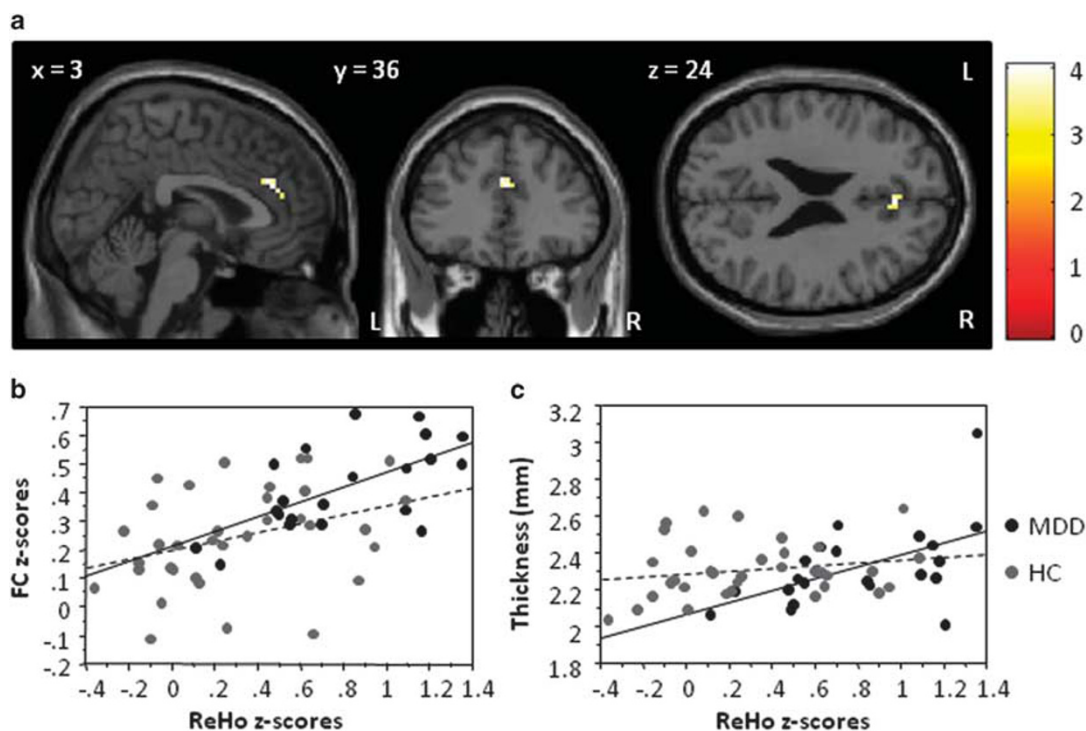
Figure 2 Brain region showing regional homogeneity differences in patients with major depressive disorder (MDD) and healthy controls. (a) Supragenual anterior cingulate cortex cluster of increased regional homogeneity (ReHo) in depressed patients compared with healthy controls (HC). (b) Scatterplots show the correlation between ReHo *z*-scores and functional connectivity (FC) strength. The correlation was significant in patients with MDD (solid line; $r^2 = 0.39$) and in healthy controls (HC: dashed line; $r^2 = 0.12$). (c) Scatterplots show the correlation between ReHo *z*-scores and cortical thickness. The correlation was significant in patients with MDD (solid line; $r_{kendall's\ tau} = 0.45$) but not in HC (dashed line; $r_{kendall's\ tau} = 0.15$).

In the current study, we used the self-focused rumination score of the Response Style Questionnaire as a measure of adaptive rumination in unmedicated depressed patients and showed that higher Res-SeRumination was associated with lower depressive symptoms after 22 weekly CBT sessions (independently of depression severity prior to therapy). Although changes in depressive symptoms prior to and after therapy represent a direct measure of treatment response, self-focused rumination is a self-referential process and thus more directly related to the function of the ACC and DMN at rest (Nejad *et al*, 2013; Pizzagalli, 2011). Our results show that MDD was associated with thinning of the right rostral middle frontal cortex and stronger FC with the suACC. Importantly, in depressed patients greater FC was associated with higher Res-SeRumination. MDD was also associated with higher ReHo values in the suACC. As the ACC clusters of increased ReHo spatially overlapped with ACC cluster of increased FC, we assessed whether ReHo values were related to thickness and/or FC strength. We found that, in patients, higher ReHo scores correlated with stronger FC and greater thickness of the right rostral middle frontal cortex. These results suggest that increased suACC FC and ReHo may foster forms of self-referential processing associated with better CBT response and that thinning of the rostral middle frontal cortex may reduce the ability of depressed patients to engage the suACC.

In the present study, MDD was associated with a thinning of the right rostral middle frontal cortex. The peak of the thinning occurred in the right DLPFC, but the cluster spatially extended into the frontopolar cortex. This finding is consistent with a recent meta-analysis that reported reduced DLPFC volume in patients with recurrent episodes (Bora *et al*, 2012). Studies that have investigated cortical thickness in MDD have also reported thinning of several prefrontal regions, including the DLPFC and the medial PFC (eg, Tu *et al*, 2012; van Tol *et al*, 2013). Moreover, reduced right DLPFC volume and widespread thinning across the right lateral cortex have also been identified in healthy subjects with a familial risk for MDD (Amico *et al*, 2011; Peterson *et al*, 2009), which suggests that PFC thinning particularly in the right hemisphere may be present even prior to the onset of the disorder and may represent a vulnerability to MDD, an hypothesis that should be further investigated.

This interpretation is consistent with our FC results, which indicated increased FC of the right anterior PFC with the suACC in patients compared with controls. Although stronger connectivity between these fronto-cingulate regions was not related to greater thickness in depressed patients, there was a marginal relationship in controls, which suggests that the right anterior PFC thickness may influence the functional coupling with the suACC in healthy subjects. Moreover, a recent study reported increased BOLD response in the DLPFC, the suACC, and the midcingulate cortex during a cognitive interference task in healthy subjects with a high familial risk for MDD, which suggests that it may represent a resilient endophenotype of the disorder (Peterson *et al*, 2014). These results are also supported by previous evidence showing that, during

expectancy-induced modulation of emotional picture processing, altered DMPFC and DLPFC response in depressed patients normalized after remission in the DMPFC but not in the DLPFC (Bermpohl et al, 2009). Importantly, in depressed patients greater FC was associated with higher Res-SeRumination and marginally with lower depressive symptoms posttherapy. The suACC also showed higher ReHo values in depressed patients compared with healthy controls; in patients only, higher ReHo values were associated with greater PFC thickness, stronger fronto-cingulate connectivity, and higher Res-SeRumination. These results suggest that greater suACC ReHo and connectivity may foster adaptive rumination and that right PFC thinning may impair the ability of depressed patients to engage the suACC during a major depressive episode.

The role of the frontopolar cortex and DLPFC in treatment response is supported by recent findings showing that, in depressed patients, decrease in depression severity over 6 months of pharmacological treatment is associated with an increased ability to engage the frontopolar cortex and right DLPFC when regulating negative emotions (Heller et al, 2013). Our results are consistent with previous evidence indicating that the response to pharmacological treatment is related to higher FC between the suACC and the DLPFC in elderly depressed patients (Alexopoulos et al, 2012). Moreover, they provide some support for the hypothesis of Pizzagalli (2011) that increased ACC activity at rest fosters adaptive forms of rumination important for treatment response and further indicates that this process is related to greater thickness and stronger connectivity of the right anterior PFC. Although the suACC cluster found in our study is located more dorsally than the pregenual ACC region reported by Pizzagalli (2011), recent electrophysiological findings comparing responders to antidepressant treatment to non-responders show that greater pretreatment rest activity (slow-wave frequency power) is found in both the suACC and the pregenual ACC (Rentzsch et al, 2014). However, only pretreatment activity in the suACC was associated with symptom improvement. Moreover, the suACC activity predicted treatment response with a higher accuracy than the reduction in symptom severity during the early phase of the therapy (considered an important clinical predictor of treatment response (Henkel et al, 2009)). Nevertheless, it is important to consider that these ACC regions are anatomically connected, leading to the possibility that reciprocal functional influences may occur. In addition, although we did not find structural alterations in the ACC, reduced ACC volume has been widely reported in depressed patients (Bora et al, 2012; Koolschijn et al, 2009; Lai, 2013; van Tol et al, 2010). Recently, structural changes in the suACC have been proposed as a trait marker of MDD (Li et al, 2014; van Eijndhoven et al, 2013). Moreover, greater ACC volume has been associated with better clinical outcome (Chen et al, 2007; Frodl et al, 2008), supporting the hypothesis that the ACC function may be a useful predictor of treatment response in MDD.

Repetitive transcranial magnetic stimulation (rTMS) of the DLPFC has been shown to have therapeutic effects for treatment-resistant MDD (Chen et al, 2013b). The neurobiological mechanisms underlying these effects are poorly understood; however, rTMS has been shown to influence cerebral blood flow in the suACC and pregenual ACC (Paus et al, 2001). In addition to the DLPFC, other brain regions have been proposed as a potential therapeutic rTMS targets for MDD, including the DMPFC and the frontopolar cortex (Downar and Daskalakis, 2013). Recent evidence shows that greater FC of the subgenual ACC with the suACC and the DLPFC prior to treatment predicts better response to rTMS of the DMPFC (Salomons et al, 2014). As FC does not provide a measure of directionality, the reported findings suggest that bolstering the function of the suACC may improve response to a variety of treatments in MDD by fostering connectivity with fronto-cingulate regions.

Several limitations must be considered. First, we employed the Res-SeRumination, but previous studies have employed the self-reflection subscale of the Ruminative Responses Scale, which may provide a more specific measure of adaptive rumination (Hamilton et al, 2011). Second, changes in depressive symptoms prior to and after therapy represent a direct measure of treatment response; however, self-focused rumination is a self-referential process directly related to brain function at rest and thus may be more easily associated with RS-fMRI data (Nejad et al, 2013). Third, we did not find altered cingulate thickness in MDD. However, reduced ACC volume is consistently reported in depressed patients (Bora et al, 2012; Koolschijn et al, 2009; Lai, 2013; van Tol et al, 2010). Moreover, greater ACC volume is associated with better clinical outcome (Chen et al, 2007; Frodl et al, 2008), suggesting that individual differences in the ACC structure may contribute to our ReHo and FC findings (Kuhn et al, 2012). Fourth, although we acknowledge that the altered FC may be the result of the cortical-thickness-guided seed selection, the ReHo results provide an additional measure of functional suACC alterations in MDD. Fifth, patients with comorbid anxiety were not excluded; thus our results may be more relevant for a subgroup of patients with high levels of anxiety. Finally, physiological noise can affect the FC and ReHo estimation in RS-fMRI studies.

In conclusion, our results suggest that greater suACC ReHo and connectivity with the anterior PFC are functional changes that foster adaptive rumination. Furthermore, right anterior PFC thinning may impair the ability of depressed patients to engage the suACC during a major depressive episode. We suggest that bolstering the function of the suACC may represent a potential target for developing more effective MDD treatments.

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