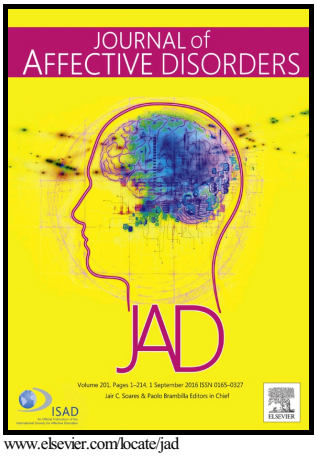


www.elsevier.com/locate/jad

Changes in brain connectivity during a sham-controlled, transcranial magnetic stimulation trial for depression

Stephan F. Taylor, S. Shaun Ho, Tessa Abagis, Mike Angstadt, Daniel F. Maixner, Robert C. Welsh, Luis Hernandez-Garcia

PII: S0165-0327(17)32586-7
DOI: https://doi.org/10.1016/j.jad.2018.02.019
Reference: JAD9565

To appear in: *Journal of Affective Disorders*

Received date: 13 December 2017
Revised date: 13 February 2018
Accepted date: 16 February 2018

Cite this article as: Stephan F. Taylor, S. Shaun Ho, Tessa Abagis, Mike Angstadt, Daniel F. Maixner, Robert C. Welsh and Luis Hernandez-Garcia, Changes in brain connectivity during a sham-controlled, transcranial magnetic stimulation trial for depression, *Journal of Affective Disorders,* https://doi.org/10.1016/j.jad.2018.02.019

Changes in brain connectivity during a sham-controlled, transcranial magnetic stimulation trial for depression

Stephan F. Taylor[*,§], S. Shaun Ho [¶], Tessa Abagis[#], Mike Angstadt[*], Daniel F. Maixner[*], Robert C. Welsh [†], Luis Hernandez-Garcia[¥]

Running title: Brain connectivity changes with rTMS

Keywords: subgenual cingulate gyrus; functional magnetic resonance imaging; default mode network; anterior insula; placebo; dorsolateral prefrontal cortex

Departments of Psychiatry[*], Psychology[#], fMRI Laboratory[¥], University of Michigan, Ann Arbor, Michigan; Department of Psychiatry, Stony Brook University[¶]; Department of Psychiatry, University of Utah[†]

§To whom correspondence should be addressed:
Department of Psychiatry
Rachel Upjohn Building
4250 Plymouth Rd
Ann Arbor MI 48109-2700
Phone: (734) 936-4955 Fax: (734) 936-7868
Email: sftaylor@umich.edu

This work has been previously presented at the annual meeting of the American College of Neuropsychopharmacology, Hollywood, FL, December 2016

Clinicaltrials.gov NCT01900314

Abstract

Background: The subgenual anterior cingulate cortex (sgACC) has been implicated in major depressive disorder (MDD), and this study evaluated sgACC connectivity before and after repetitive transcranial magnetic stimulation (rTMS) treatment.

Methods: Thirty-two MDD patients entered a sham-controlled, double-blinded, randomized trial of rTMS to the left dorsolateral prefrontal cortex (dlFPC). Subjects underwent resting state functional magnetic resonance imaging before and after 20 sessions of high frequency rTMS. Seed voxels identified the affective network (AN; sgACC, amygdala), default mode network (DMN; posterior cingulate cortex [PCC]), and fronto-parietal network (FPN; dlPFC stimulation site).

Results: There was no significant effect of active rTMS over sham on the primary outcome measure (Montgomery-Asberg Depression Scale rating), with both groups improving over time, and no specific effect of rTMS (sham vs active) on connectivity. However, among patients who showed significant improvement, sgACC connectivity decreased for sham (to AN, trend to DMN) and active rTMS responders (to AN, DMN, FPN), but not in non-responders, who tended to maintain connectivity. Including subjects who started with sham but then received open-label active treatment, baseline connectivity from the PCC to the anterior insula was greater in non-responders compared to responders (n=27, excluding 5 sham responders).

Limitations: The sample size was small; the stimulation target was non-standard, and the lack of a significant clinical effect of rTMS limits conclusions about negative findings.

Conclusions: sgACC connectivity reduces along with depressive symptoms, not specific to rTMS therapy. Altered connectivity of DMN with anterior insula may reflect a type of patient less likely to respond to an intervention.

Introduction

Repetitive transcranial magnetic stimulation (rTMS) is an effective treatment for depression (Berlim et al., 2014b; Lam et al., 2008; Schutter, 2009; Slotema et al., 2010), but little is known about how rTMS affects the brain of depressed patients. One possibility is that rTMS changes intrinsic connectivity networks (ICN), which exhibit abnormal brain connectivity in depression (Kaiser et al., 2015). The subgenual anterior cingulate cortex (sgACC) is a key node implicated in depression (Drevets, 2000; Hamani et al., 2011), and several groups have found abnormally increased connectivity between sgACC and the default mode network (DMN) in depression (Berman et al., 2011; Hamilton et al., 2011; Zhu et al., 2012), possibly causing negative affective biasing of internal dialogue, leading to ruminations (Hamilton et al., 2011). This theory suggests that treatment of depression might entail reducing the influence of sgACC on the DMN (Hamilton et al., 2015). rTMS treatment has been associated with reduced metabolism (Baeken et al., 2015) and reduced

blood flow to the sgACC (Kito et al., 2008). Studies of other brain stimulation modalities have implicated the sgACC, such as deep brain stimulation decreasing sgACC activity (Berlim et al., 2014a; Hamani et al., 2011) and electroconvulsive therapy reducing the amplitude of low frequency fluctuations of fMRI signal (Argyelan et al., 2016). Thus, several lines of research suggest that sgACC may mediate the effects of rTMS.

Although repetitive transcranial magnetic stimulation (rTMS) is typically applied to the prefrontal cortex, Fox and colleagues have reported that, amongst published rTMS studies, those that used coil locations closest to the point of maximal *negative correlation* of the dorsolateral prefrontal cortex (dlPFC) with the sgACC reported the greatest reductions in depressive symptoms (Fox et al., 2012; Weigand et al., 2017). These data suggest a possible mechanism by which stimulation of a superior cortical region could affect a deep structure like the sgACC, far from the locus of stimulation. In partial support of this hypothesis, a recent report found reduced sgACC connectivity to the DMN after dorsolateral rTMS (Liston et al., 2014). Another study using dorsomedial rTMS also showed reduced sgACC connectivity with the caudate and dorsal anterior cingulate (Salomons et al., 2014). While this work suggests possible mechanisms of the effect of rTMS to treat depression, these studies lacked a sham-control condition that would allow one to separate specific effects of rTMS from non-specific effects of a treatment intervention.

Another question about the relationship between rTMS and ICNs is whether baseline connectivity can predict the response to rTMS therapy, an important question in the search for biomarkers that could guide therapy. Connectivity of the sgACC has been implicated as a potential response biomarker. Greater connectivity with superior medial prefrontal cortex (Baeken et al., 2014; Salomons et al., 2014), DMN (Liston et al., 2014) and

dlPFC (Salomons et al., 2014) has been associated with better response to rTMS. Other connectivity patterns correlated with a positive therapeutic response to rTMS include lower cortico-thalamic, cortico-striatal and cortico-limbic connectivity (Salomons et al., 2014), higher connectivity in reward-related areas (Downar et al., 2014) and greater connectivity of medial PFC, dlPFC, posterior cingulate cortex (PCC), orbitofrontal cortex and amygdala (Drysdale et al., 2017).

To address these questions, the present study employed a randomized, sham-controlled design, focusing on the sgACC, first by testing the hypothesis that stimulation would alter connectivity between the target site and sgACC (making it more negatively correlated), and second by testing whether connectivity between sgACC and the DMN was reduced with treatment. We defined the following ICNs (Yeo et al., 2011) of primary interest: DMN, fronto-parietal (containing the site of stimulation) and limbic/affective (containing sgACC and amygdala). We also included the ventral attention/salience network because of its involvement in depression (Goodkind et al., 2015; Kaiser et al., 2015). Our study design employed two phases: a double-blinded phase, and then an open-label phase, in which subjects randomized to sham treatment in the first phase went on to receive active treatment, permitting us to explore baseline connectivity markers correlated with treatment response. Although we found no specific effect of rTMS on connectivity in the comparison of active to sham stimulation, we did find that improvement in depressive symptoms was associated with reduced sgACC connectivity.

Methods

Subjects

Forty outpatient subjects with major depressive disorder, diagnosed with DSM-IV criteria using the M.I.N.I. (Sheehan et al., 1998), were enrolled and randomized in the protocol between October 2013 and October 2015. Subjects were recruited through advertisements in the community and referrals from clinicians at the University of Michigan Department of Psychiatry, where the study took place. Subjects were between 22 and 65 years of age, failed at least one antidepressant medication trial, had at least moderate depressive severity (Montgomery-Asberg Depression Rating Scale [MADRS] ≥ 18; Montgomery and Asberg, 1979) and ≤ 5 years in the current episode. Exclusion criteria included bipolar disorder (I/II), obsessive-compulsive disorder, post-traumatic stress disorder, any psychosis, serious suicidal ideation/behavior, previous rTMS, previous ECT and contra-indications to rTMS or MRI. Subjects were assessed with the Antidepressant History Treatment Form (ATHF; Sackeim, 2001) to assess level of treatment resistance. Medications had to be maintained on a stable dosage for 1 month prior to and during the double-blinded phase. All subjects gave written consent to participate according to a protocol approved by the University of Michigan Institutional Review Board.

Design and rTMS protocol

The protocol was a sham-controlled, randomized (1:1), double-blinded study. Patients were assigned to treatment arm, using block randomization, stratified by gender. Randomization was performed by a member of the study team (S.H.) not involved in

treatment assessments. Clinicians assessing symptom change over the course of the study (S.F.T, D.F.M.) were blinded as to treatment assignment. Assessment of TMS side effects was performed by a research nurse, who did not discuss treatment side effects with the clinician performing the assessments. After 20 sessions of therapy and the second MRI scan was completed, patients and treating clinicians were queried as to which treatment arm they thought the patient was on. Subjects who received sham stimulation had the option to receive active rTMS in the second, open-label phase of the study.

After initial screening and assessment, subjects entered Phase 1, consisting of the MRI session, followed by motor threshold determination obtained using visual identification of thumb twitch and initiation of treatment. In this blinded phase, subjects had 20 sessions of rTMS therapy or sham treatment, 5 days per week. rTMS treatments were delivered at 10 Hz frequency at 120% of motor threshold and 3000 pulses/session to the left dlPFC, determined by individualized neuronavigation (see Supplementary materials). Not all subjects were able to tolerate 120% at the initial session, but all subjects reached 120% after 1 week of treatments. Stimulation used the NeuroStar XPLOR system in research mode. One coil was active, while the sham coil was identical in shape and weight to the active coil, but did not deliver any magnetic energy. A speaker on the coil gantry delivered a 10 Hz clicking sound mimicking acoustic characteristics of the active coil, but no superficial stimulation emanated from the sham coil. A third coil delivered active stimulation, and it was used to obtain the motor threshold, thus preserving the blind -- all patients saw a new coil switched in after motor threshold was obtained. At the end of Phase 1, a second MRI occurred, followed by open-label treatment in Phase 2: either 5 taper

sessions over 2 weeks for those in the active arm during Phase 1, or 20 sessions plus 5 taper sessions for those receiving sham stimulation in Phase 1.

Clinical assessments and analysis

The primary clinical endpoint was MADRS score after 20 sessions. Secondary endpoints included Hamilton Rating Scale for Depression (17 item; HRSD17; Hamilton, 1960), Quick Inventory of Depressive Symptoms, Self-rated (QIDS-SR; Rush et al., 2003) and Generalized Anxiety Disorder Assessment (GAD-7; Spitzer et al., 2006). Function was assessed with the Work and Social Adjustment Scale (WSAS; Mundt et al., 2002) and the Global Assessment of Function (GAF; Hall, 1995). Assessments occurred weekly for the 20 sessions of rTMS treatment, and at the end of the last taper session. The primary endpoint was MADRS score after 20 sessions, using a linear mixed effects model incorporating treatment group, time (weekly assessments), group by time interaction, with screening MADRS as a co-variate and patient as a random effect. Categorical treatment response was defined as a 50% change from baseline MADRS, and remission was defined as MADRS < 10. Alpha was set to 0.05 and analyses were conducted in SPSS (version 23).

Additional assessments occurred in Phase 2. Patients who had active treatment in blinded, Phase 1 were assessed after they completed their 5 taper treatments. Patients who received sham treatment during Phase 1 and went on to active, open-label treatment, were assessed at the completion of their 20 treatments + 5 taper treatments (identified as treatments 21 through 45, numbered to include the 20 sham treatments). These

assessments were used in the analysis to identify connectivity predictors of response to rTMS, maximizing the power of the sample by including all patients who received rTMS.

Functional MRI acquisition and analysis

MRI scanning occurred on a 3T GE 750 Discovery scanner (General Electric Healthcare, Chicago, IL). Blood oxygenation level dependent (BOLD) functional images were acquired with a T2*-weighted, reverse spiral acquisition (gradient recalled echo, TR = 2000 msec, TE=30 msec, FA=90 degrees, field of view=22 cm, 43 slices, 3.0mm thick/0mm skip, equivalent to 64 x 64 voxel grid – yielding approximately isotropic voxels), with excellent sensitivity to signal in ventral medial frontal regions (Yang et al., 2002). Two hundred and forty acquisitions were acquired in the resting state with eyes open and fixated on a large 'plus' sign projected on a monitor. Four initial volumes were discarded to allow for equilibration of scanner signal. A high resolution T1 scan (3D SPGR) was obtained for anatomic normalization, as well as a T1 spin echo acquisition in the same prescription as the T2* BOLD scan.

Data processing began with standard pre-processing steps, including slice-time correction, realignment and warping of functional images to the MNI152 template (Statistical Parametric Mapping SPM8 package, Wellcome Institute of Cognitive Neurology, London). Adjusted time courses at the subject level were derived from sequential regressions of the time series using regressors to remove effects of movement, physiological artifacts and activity outside a band pass filter of 0.01 – 0.1 Hz.

Seed voxels were placed to test hypotheses and focus on the following networks: the sgACC (Fox et al., 2012) along with bilateral amygdalae (Tzourio-Mazoyer et al., 2002) for the affective network (AN); target of rTMS stimulation, individualized then warped to MNI coordinates for the fronto-parietal network (FPN); posterior cingulate cortex (PCC; (Leech et al., 2011) for the DMN and dorsal anterior cingulate cortex (dACC; (Goodkind et al., 2015) for the salience network (SN). Adjusted time courses were averaged for each seed over a 6 mm spherical radius to match spatial characteristics of the image, except for the target (8 mm to accommodate anatomic variability) and the amygdala (volume from automated anatomic labeling (Tzourio-Mazoyer et al., 2002). The average time course was correlated with time courses from all other voxels in the brain, Fisher r-to-Z transformed, and taken to second level, between-group analyses. A statistical model was constructed to test for the primary effect of *rTMS on changing connectivity* from scan 1 to scan 2, but because no effect was found, a secondary model tested for the effect of symptom change (dichotomized into responder versus non-responder) on changing connectivity, focusing on sgACC as the primary seed. An additional planned analysis tested for *baseline connectivity as a predictor* of symptom change, dichotomized into responder versus non-responder and modeled in the inverse to accommodate the SPM framework, i.e. symptom change predicting connectivity. This analysis included rTMS treatments during the taper phase and subjects who started with sham treatment, but went on to receive active rTMS. Regression models were run with baseline MADRS and subject movement (mean frame displacement [FD]; (Power et al., 2012) as co-variates of no interest. Whole brain, voxel-wise images were statistically thresholded to control type 1 errors at $p<0.05$ using random field theory applied to peak voxel values and cluster sizes (cluster threshold = $p<0.001$;

(Worsley et al., 1997) In addition to the whole-brain analysis, a seed-to-network analysis was performed by averaging Z-scores over each of 7 networks covering the entire cortex (Yeo et al., 2011) for each seed. The primary focus of this analysis was sgACC connectivity, first with DMN, and then with the fronto-parietal network and within the affective network. These analyses paralleled the voxel-wise analyses, using ANCOVAs, in SPSS (version 23).

See Supplementary material for additional details of fMRI analysis.

Results

Subjects and Clinical Outcomes

Of the 40 subjects enrolled (from 44 screened), 34 completed the first MRI and 32 completed both scans (Supplementary material, Figure S1). Sixteen subjects completed each arm. The two groups did not differ on demographic characteristics, although, in spite of randomization, subjects assigned to the active group had more severe symptoms (Table 1). All subjects, except for 2, were on antidepressant medication (Table S1). Fifty percent of subjects correctly guessed they were receiving sham stimulation, but 80% of subjects receiving active stimulation correctly guessing their assignment. Subjects tolerated the treatments well and there were no serious adverse events.

After 20 treatments at the end of Phase 1, there was a significant effect of time, but no significant group by time interaction, i.e. no differential effect of active compared with sham rTMS ($p = 0.19$). This was true for the primary and secondary endpoints, although effects for all of the depression scales were in the expected direction (Table S2). Response

and remission rates were 44% and 25%, respectively for the active arm, and 31% and 31%, respectively, for the sham arm (χ^2 = 0.53, p=0.46, χ^2=0.15, p=0.69, respectively). At the end of Phase 1, 5 subjects in the sham arm exhibited significant responses (MADRS range 1-8), and 2 of those subjects elected not to continue with active treatment, while the remaining 14 subjects went on to receive active, open-label treatment in Phase 2.

In the analysis of movement during scanning, the groups did not differ on mean FD or frames scrubbed (Supplementary materials, Figure S2).

Connectivity change over time

There were no significant effects (group X time interactions) surviving correction for multiple comparisons across the whole brain in the voxel-wise analysis for any of the 6 seeds. Even examining the sgACC region at an uncorrected threshold of p<0.01, there was no support for an effect of rTMS on target-to-sgACC connectivity. For the seed-to-network analysis, we also found no significant effects for the 6 seeds with any networks (all p's > 0.2).

In contrast to the null effects of rTMS versus sham, sgACC connectivity decreased for several networks in responders (n=12, which included sham responders), compared to non-responders (n=20) during Phase 1. Specifically, Phase 1 responders decreased connectivity between the sgACC with the left inferior parietal lobule (IPL) and the left orbital frontal cortex (OFC; Figure 1). In the seed-to-network analysis, responders exhibited reductions in sgACC connectivity with the DMN, as well as the FPN and AN, after starting with similar connectivity as the non-responders at scan 1 (Figure 2A). As can be

seen in Figure S3B, the decrease in connectivity appears to affect sgACC connectivity with all networks, but was only significant for DMN, FPN and AN. The analysis was conducted without the 5 sham responders, and the network changes in sgACC connectivity over time were still significant. In addition, when the 7 active rTMS responders were excluded, the sham responders also exhibited significant reductions in the AN, a trend-level change for the DMN, but not FPN, in comparison to the non-responders (Figure 2B & C).

Uncorrected, exploratory analyses were conducted for the other seeds with other networks (Figure S3). The only effects at $p<0.05$ occurred for bilateral amygdalae with the AN, showing a slightly different pattern as the responders started with higher connectivity from the amygdala to the AN, which was reduced below the non-responders in the second MRI scan (Figure S4). No other seeds showed significant effects of symptom change in Phase 1 (Figure S3E-G).

Analysis of potential confounding variables for this contrast of responders versus non-responders showed no difference in age, gender distribution, SES, functioning or anxiety levels (Table S3). As would be expected, the responders had fewer trials of antidepressant therapy for the current episode, fewer current psychotropic medications during the study (Table S1) and lower baseline MADRS scores. There was also a differential effect on movement, wherein the responders showed an increase in movement from scan 1 to scan 2. However, the effect of movement on connectivity could not explain our results. See supplementary material for discussion.

Symptom change and baseline connectivity

We examined the relationship between baseline connectivity and symptom response, combining Phase 1 patients receiving active treatment with Phase 2 patients who elected to receive active rTMS after being randomized to sham treatment. Three sham responders who entered Phase 2 treatment were left out of this analysis (since they had already 'responded'), yielding a group of 27 subjects, with response and remission rates at the end of the taper (25 treatments, total) of 48% (13 of 27) and 33% (9 of 27), respectively (Figure S5).

In the voxel-wise search, PCC connectivity with the right anterior insula (aIns) and right inferior frontal gyrus (IFG) was greater for non-responders than responders (Figure 3). The responders had negative PCC connectivity with both of these regions (which were highly correlated, r=0.97), whereas the non-responders had positive connectivity. The aIns focus corresponds to the normal pattern of negative connectivity with the PCC in this region (Figure 3B). No other seeds exhibited a significant relationship with symptom improvement, either in the whole-brain, voxel-wise analysis, or the seed-to-voxel analysis.

The two groups of patients did not differ in age, gender distribution, SES, functioning or symptom measures. However, responders had less treatment resistance (fewer trials of antidepressant medication, Table S4) and fewer psychotropic medications during the study (Table S1). Although the non-responders exhibited more movement, this difference could not have driven the group differences because more movement correlated with less positive connectivity. See Supplementary material for a discussion of movement.

Discussion

This study sought to determine connectivity changes with rTMS therapy, and how connectivity may predict the response to rTMS. While we did not find a significant effect of rTMS stimulation, relative to sham, on ICN's, we did find that sgACC connectivity, within and between networks, reduced with symptom improvement. We also found that DMN connectivity with the aIns and IFG predicted symptom improvement. Overall, our findings provide clues about connectivity networks and symptom reduction in depression with important implications for rTMS.

The affective network and depression

Our study adds new findings to the growing literature demonstrating the role of the sgACC in depression, particularly in the context of treatment response. The decrease in sgACC connectivity over time is consistent with two prior reports of reduced sgACC connectivity in rTMS with the caudate nucleus and dorsal anterior cingulate (Salomons et al., 2014) and with the DMN (Liston et al., 2014) after rTMS treatment. Our findings expand upon this prior work, demonstrating that reductions of sgACC connectivity might be generalized and not specific to selected networks, since sgACC connectivity was reduced with the AN, as well as FPN and DMN. In our corrected, whole brain search, we found reduced connectivity with the left IPL and left OFC, neither of which corresponds to the published reports. However, as the seed-to-network analysis shows, sgACC connectivity was reduced in the networks that contain these regions (the DMN and FPN, respectively). Limited experimental power to detect differences (of these 3 studies, the present, at 32

subjects, was the largest sample size) may have led to variability in the foci that achieved significance. Cell bodies in the sgACC project to medial frontal, lateral frontal and temporal cortex (Hamani et al., 2011; Price and Drevets, 2012), and they may contribute to negative biasing in the attention to external stimuli mediated by executive control systems of the FPN and negative, ruminative self-talk mediated by the DMN (Hamilton et al., 2015; Price and Drevets, 2012). If the reduction in correlation strength identified in our findings represents influence of the sgACC on these regions, it may be the neural correlate of reduced depressive symptoms.

Our study had the advantage of a sham-treatment arm, in contrast to the rTMS studies described above, permitting us to examine specific effects of rTMS in parallel with non-specific effects, such as placebo responses. Non-specific effects could include both expectation-related improvement in mood (e.g. placebo response) as well as benefits from daily trips for rTMS sessions, social interaction during treatment or naturalistic resolution of symptoms. We did not find any specific effects of rTMS, suggesting that observed sgACC connectivity change may be non-specific. sgACC connectivity with AN and DMN was reduced over the course of treatment for both the sham responders and the active rTMS responders, when each group was separately analyzed, in spite of the reduced power in these sub-group analyses. A similar pattern occurred in the right amygdala. With small numbers and large variance, these results must be regarded with caution, but the pattern strongly suggests that AN connectivity change, both within the network (given the amygdala results) and between the sgACC and DMN, occurs for both actual and sham responders.

Our findings are consistent with the conclusion that sgACC connectivity is a 'final common pathway' of rTMS treatment. It may not be specific to the site of rTMS stimulation, since sgACC connectivity was reduced in a study that stimulated a slightly different dlPFC location (Salomons et al., 2014), and another study that stimulated the dorsal medial prefrontal cortex (Liston et al., 2014). sgACC connectivity change occurs in treatment interventions besides rTMS. For example, sgACC connectivity to the DMN was reduced after successful CBT (Straub et al., 2017), and connectivity of sgACC with regions in the AN was reduced in patients undergoing ECT (Argyelan et al., 2016). Studies of placebo response in depressed patients have also reported reductions in cerebral metabolism in the sgACC (Mayberg et al., 2002), and opioid activation of sgACC has been associated with placebo responsiveness in depressed patients (Pecina et al., 2015). Thus, a reasonable interpretation of our rTMS results is that sgACC connectivity changes represent a mechanism of disparate interventions treating depression.

The pattern of connectivity in both amygdalae was similar to that found for the sgACC in an exploratory analysis. The only significant effect (uncorrected $p<0.05$) occurred in the AN, of which the amygdala is a constituent. The amygdala and the sgACC have strong anatomic connectivity as a part of a medial prefrontal network (Ongur and Price, 2000). Hyperactivity of the amygdala has been reported in depressed patients (Hamilton et al., 2012; Palmer et al., 2014; Price and Drevets, 2012), and dysregulated connectivity between the amygdala and sgACC has been suggested as a common pathway underlying depressed mood (Etkin and Schatzberg, 2011; Murphy et al., 2016). Others have shown evidence of abnormal connectivity of the amygdala with prefrontal cortex, related to symptom severity (Satterthwaite et al., 2015; Wang et al., 2016). Decreasing amygdala connectivity with

DMN has been reported in depressed patients undergoing CBT (Straub et al., 2017) and decreasing activation to emotional stimuli (Delaveau et al., 2010). Thus, decreasing amygdala connectivity in our sample, in concert with decreasing connectivity of the sgACC, may reflect a normalization of increased connectivity within the affective network and reduced emotional influence on the rest of the brain. Although no healthy comparison group was matched to our depressed subjects, comparison of our results with the literature supports the idea that the reduction in connectivity may have been a normalization of aberrant affective influence.

Response prediction in PCC-aIns/IFG coupling

Searching for baseline connectivity predictors of treatment response, we found PCC seed connectivity with two regions in close proximity -- the right IFG, actually a part of the DMN, and the right aIns, a constituent of the salience/ventral attention network. Although we did not have a matched group of control subjects, comparison to a normative database revealed that patients showing the more normal pattern of a negative correlation between PCC and aIns were more likely to improve their symptoms. Similarly, the increased PCC-IFG connectivity might also reflect poorer response for patients with more connectivity in the DMN. Several studies show that the DMN is hyper-connected in depression (Dichter et al., 2015; Hamilton et al., 2012; Kaiser et al., 2015), and increased connectivity between DMN and task positive networks (Kaiser et al., 2015) and within the aIns (Manoliu et al., 2013) has also been noted. This increased connectivity of DMN, a network involved in stimulus-independent thought, may reflect the hijacking of cognitive processes mediated

by fronto-parietal and salience networks (also known as 'task positive' networks), leading to poor concentration, depressive rumination and negative biasing of perception (Hamilton et al., 2011). The aIns may mediate the interaction of DMN and task-positive networks, whereby tasks requiring outwardly-directed attention usually activate the task-positive networks and deactivate the DMN (Gusnard and Raichle, 2001), something that depressed patients have trouble doing (Grimm et al., 2009; Kronhaus et al., 2006; Sheline et al., 2009). Increased baseline activity in the aIns is associated with poor clinical response (Fu et al., 2013), and antidepressant treatment is associated with decreased activity in the aIns (Delaveau et al., 2010). A recent publication using vagus nerve stimulation found reduced connectivity between the DMN and aIns with treatment (Fang et al., 2016). There is also evidence that aIns connectivity abnormalities – specifically the right aIns -- are widespread across disorders, as other groups have noted abnormal connections in social anxiety disorder (Klumpp et al., 2012) and obsessive-compulsive disorder (Stern et al., 2011). Recent meta-analyses of structural and functional neuroimaging studies converge on the aIns, along with the dACC, as a set of core of brain regions affected across most psychiatric disorders (Downar et al., 2016). Lastly, it is important to note that the relationship between PCC connectivity may not have been specific to rTMS, as the symptom reduction would also included placebo expectations, spontaneous remission and general effects of study participation. Thus, while PCC-aIns and PCC-IFG connectivity may mark a group of patients with greater insular-DMN pathology who are less likely to respond to rTMS, the effect could be, like sgACC connectivity, non-specific.

Limitations

We did not find a significant clinical effect of active rTMS compared to sham treatment, and this fact must limit any conclusions about the apparent lack of an effect of rTMS on ICN's. Depression scores changed in the expected direction and the effect size for active versus sham treatment was 0.53 (Cohen's-d), very similar to reported effect sizes of 0.39 – 0.5 for rTMS (Berlim et al., 2014b; Lam et al., 2008; Schutter, 2009; Slotema et al., 2010), but our experimental power was low (43% for an independent samples t-test and alpha of 0.05). With only 20 sessions and 60,000 pulses in the blinded phase, a design copied from older randomized studied (George et al., 2010; O'Reardon et al., 2007), our subjects received fewer pulses than the standard clinical practice (Carpenter et al., 2012; Taylor et al., 2017), and data suggests that many patients respond after 20 sessions (Yip et al., 2017). Target localization to the left dlPFC, based on a theoretical idea about the relationship between dlPFC and negative correlations with sgACC, was not standard. These factors could have contributed to a weaker clinical response, while the fact that our subjects had only moderate depression severity, may have led to a higher placebo response rate as others have reported for TMS treatment (Li et al., 2014). More patients receiving active treatment than those receiving sham treatment correctly guessed their treatment assignment, suggesting that the sham was not a perfect control for the study. Thus, it would be inappropriate to conclude from our data that rTMS, itself, has no effect on connectivity.

Several other limitations should be kept in mind. Almost all of our patients were on medication, and since we only required one month of stable medication dose, there may have been more symptom instability than if we had required, for example, two months of stable dosing. In the two analyses contrasting responders with non-responders, the non-

responders had more prior treatment failures and more psychotropic medications during the study. Treatment resistance has been linked to a poor response to rTMS (George et al., 2010; Lisanby et al., 2009), but see (Schutter, 2009). We did not attempt to remove the effect of treatment resistance by statistical co-variance, as the connectivity finding may be part of treatment resistance. It is possible that medication could have moderated symptom improvement observed over the course of rTMS therapy, in line with our interpretation of reductions in sgACC connectivity as a final common pathway. For purposes of predicting response to treatment, the presence of medications makes our results more generalizable, since most patients in regular clinical rTMS are also taking psychotropic medications (Carpenter et al., 2012; Taylor et al., 2017). Nevertheless, additional studies with larger sample sizes will be necessary to parse separate effects of current medications regimens and treatment resistance on connectivity.

Connectivity measures are unstable in small samples, and even though our study, with 32 subjects, is of average size relative to the published literature, negative findings must be regarded with caution. The 7-network parcellation used in this study may not optimally identify appropriate networks for rTMS therapy. While the seed-based approach to connectivity is simple, it is well suited to test specific hypotheses as we did in this study. Nevertheless, future work with larger sample sizes that can support data-driven approaches, like graph theoretic (Rubinov and Bullmore, 2013) or multivariate techniques (Norman et al., 2006), are warranted.

Conclusions

In conclusion, we have conducted a randomized, controlled study of rTMS in moderately depressed patients and demonstrated that symptom improvement was associated with decreasing connectivity of the sgACC within the AN and with DMN and FP networks. These data reinforce a growing literature about the importance of this node in the treatment of depression, supporting the conclusion that changing connectivity may be a final common pathway of disparate treatment interventions, including the placebo response. Our preliminary finding off PCC-aIns connectivity associated with treatment response aligns with a growing literature on the role of the DMN in depression, but more data are needed to better understand how depressive symptoms involve aberrant interactions between DMN and ventral attention/salience node.

Declarations of Interest:

The authors report the following disclosures relevant to the content of this article: Dr. Taylor has received research support from Neuronetics and St.Jude Medical (now Abbott). Dr. Maixner has received research support from St. Jude Medical (now Abbott). All other authors have nothing to disclose.

Author statement

Contributions:

Study conception and design: Taylor, Ho, Hernandez-Garcia, Maixner

Acquisition of data: Taylor, Ho, Abagis, Maixner

Analysis and interpretation of data: Ho, Taylor, Angstadt, Welsh

Drafting of manuscript: Taylor

Critical Revisions: All authors

Approval of final manuscript: All authors

Funding Support: This study was supported by NIH R21 MH098174 and Clinical Sciences Translational Award UL1TR000433. Research support was also provided by Neuronetics, which did not have a role in the design of the study.

Role of the sponsors: The sponsors provided funding only, except for the Clinical Sciences Translational Award, which provided assistance in the set-up of the clinical database.

Acknowledgements: The authors wish to acknowledge the assistance of Mona Goldman for drafting the protocol, Laura Stchur and Iva Grasso for assessing and monitoring patients, Jie Fan for extracting imaging data and Elise Demeter for setting up the image processing and neuronavigation protocol.

References

Argyelan, M., Lencz, T., Kaliora, S., Sarpal, D.K., Weissman, N., Kingsley, P.B., Malhotra, A.K., Petrides, G., 2016. Subgenual cingulate cortical activity predicts the efficacy of electroconvulsive therapy. Transl Psychiatry 6, e789.

Baeken, C., Marinazzo, D., Everaert, H., Wu, G.R., Van Hove, C., Audenaert, K., Goethals, I., De Vos, F., Peremans, K., De Raedt, R., 2015. The Impact of Accelerated HF-rTMS on the Subgenual Anterior Cingulate Cortex in Refractory Unipolar Major Depression: Insights From 18FDG PET Brain Imaging. Brain Stimul 8, 808-815.

Baeken, C., Marinazzo, D., Wu, G.R., Van Schuerbeek, P., De Mey, J., Marchetti, I., Vanderhasselt, M.A., Remue, J., Luypaert, R., De Raedt, R., 2014. Accelerated HF-rTMS in treatment-resistant unipolar depression: Insights from subgenual anterior cingulate functional connectivity. World J Biol Psychiatry 15, 286-297.

Berlim, M.T., McGirr, A., Van den Eynde, F., Fleck, M.P., Giacobbe, P., 2014a. Effectiveness and acceptability of deep brain stimulation (DBS) of the subgenual cingulate cortex for treatment-resistant depression: a systematic review and exploratory meta-analysis. J Affect Disord 159, 31-38.

Berlim, M.T., van den Eynde, F., Tovar-Perdomo, S., Daskalakis, Z.J., 2014b. Response, remission and drop-out rates following high-frequency repetitive transcranial magnetic stimulation (rTMS) for treating major depression: a systematic review and meta-analysis of randomized, double-blind and sham-controlled trials. Psychol Med 44, 225-239.

Berman, M.G., Peltier, S., Nee, D.E., Kross, E., Deldin, P.J., Jonides, J., 2011. Depression, rumination and the default network. Social cognitive and affective neuroscience 6, 548-555.

Carpenter, L.L., Janicak, P.G., Aaronson, S.T., Boyadjis, T., Brock, D.G., Cook, I.A., Dunner, D.L., Lanocha, K., Solvason, H.B., Demitrack, M.A., 2012. Transcranial magnetic stimulation (TMS) for major depression: a multisite, naturalistic, observational study of acute treatment outcomes in clinical practice. Depress Anxiety 29, 587-596.

Delaveau, P., Jabourian, M., Lemogne, C., Guionnet, S., Bergouignan, L., Fossati, P., 2010. Brain effects of antidepressants in major depression: A meta-analysis of emotional processing studies. J Affect Disord.

Dichter, G.S., Gibbs, D., Smoski, M.J., 2015. A systematic review of relations between resting-state functional-MRI and treatment response in major depressive disorder. J Affect Disord 172, 8-17.

Downar, J., Blumberger, D.M., Daskalakis, Z.J., 2016. The Neural Crossroads of Psychiatric Illness: An Emerging Target for Brain Stimulation. Trends Cogn Sci 20, 107-120.

Downar, J., Geraci, J., Salomons, T.V., Dunlop, K., Wheeler, S., McAndrews, M.P., Bakker, N., Blumberger, D.M., Daskalakis, Z.J., Kennedy, S.H., Flint, A.J., Giacobbe, P., 2014. Anhedonia and reward-circuit connectivity distinguish nonresponders from responders to dorsomedial prefrontal repetitive transcranial magnetic stimulation in major depression. Biol Psychiatry 76, 176-185.

Drevets, W.C., 2000. Neuroimaging studies of mood disorders. Biol Psychiatry 48, 813-829.

Drysdale, A.T., Grosenick, L., Downar, J., Dunlop, K., Mansouri, F., Meng, Y., Fetcho, R.N., Zebley, B., Oathes, D.J., Etkin, A., Schatzberg, A.F., Sudheimer, K., Keller, J., Mayberg,

H.S., Gunning, F.M., Alexopoulos, G.S., Fox, M.D., Pascual-Leone, A., Voss, H.U., Casey, B.J., Dubin, M.J., Liston, C., 2017. Resting-state connectivity biomarkers define neurophysiological subtypes of depression. Nat Med 23, 28-38.

Etkin, A., Schatzberg, A.F., 2011. Common abnormalities and disorder-specific compensation during implicit regulation of emotional processing in generalized anxiety and major depressive disorders. The American journal of psychiatry 168, 968-978.

Fang, J., Rong, P., Hong, Y., Fan, Y., Liu, J., Wang, H., Zhang, G., Chen, X., Shi, S., Wang, L., Liu, R., Hwang, J., Li, Z., Tao, J., Wang, Y., Zhu, B., Kong, J., 2016. Transcutaneous Vagus Nerve Stimulation Modulates Default Mode Network in Major Depressive Disorder. Biol Psychiatry 79, 266-273.

Fox, M.D., Buckner, R.L., White, M.P., Greicius, M.D., Pascual-Leone, A., 2012. Efficacy of Transcranial Magnetic Stimulation Targets for Depression Is Related to Intrinsic Functional Connectivity with the Subgenual Cingulate. Biological psychiatry.

Fu, C.H., Steiner, H., Costafreda, S.G., 2013. Predictive neural biomarkers of clinical response in depression: a meta-analysis of functional and structural neuroimaging studies of pharmacological and psychological therapies. Neurobiol Dis 52, 75-83.

George, M.S., Lisanby, S.H., Avery, D., McDonald, W.M., Durkalski, V., Pavlicova, M., Anderson, B., Nahas, Z., Bulow, P., Zarkowski, P., Holtzheimer, P.E., 3rd, Schwartz, T., Sackeim, H.A., 2010. Daily left prefrontal transcranial magnetic stimulation therapy for major depressive disorder: a sham-controlled randomized trial. Arch Gen Psychiatry 67, 507-516.

Goodkind, M., Eickhoff, S.B., Oathes, D.J., Jiang, Y., Chang, A., Jones-Hagata, L.B., Ortega, B.N., Zaiko, Y.V., Roach, E.L., Korgaonkar, M.S., Grieve, S.M., Galatzer-Levy, I., Fox, P.T., Etkin, A., 2015. Identification of a common neurobiological substrate for mental illness. JAMA Psychiatry 72, 305-315.

Grimm, S., Boesiger, P., Beck, J., Schuepbach, D., Bermpohl, F., Walter, M., Ernst, J., Hell, D., Boeker, H., Northoff, G., 2009. Altered negative BOLD responses in the default-mode network during emotion processing in depressed subjects. Neuropsychopharmacology 34, 932-843.

Gusnard, D.A., Raichle, M.E., 2001. Searching for a baseline: functional imaging and the resting human brain. Nat Rev Neurosci 2, 685-694.

Hall, R., 1995. Global Assessment of Functioning modified scale. Psychosomatic 36, 267-275.

Hamani, C., Mayberg, H., Stone, S., Laxton, A., Haber, S., Lozano, A.M., 2011. The subcallosal cingulate gyrus in the context of major depression. Biological psychiatry 69, 301-308.

Hamilton, J.P., Etkin, A., Furman, D.J., Lemus, M.G., Johnson, R.F., Gotlib, I.H., 2012. Functional neuroimaging of major depressive disorder: a meta-analysis and new integration of base line activation and neural response data. The American journal of psychiatry 169, 693-703.

Hamilton, J.P., Farmer, M., Fogelman, P., Gotlib, I.H., 2015. Depressive Rumination, the Default-Mode Network, and the Dark Matter of Clinical Neuroscience. Biol Psychiatry 78, 224-230.

Hamilton, J.P., Furman, D.J., Chang, C., Thomason, M.E., Dennis, E., Gotlib, I.H., 2011. Default-mode and task-positive network activity in major depressive disorder: implications for adaptive and maladaptive rumination. Biological psychiatry 70, 327-333.

Hamilton, M., 1960. A rating scale for depression. J Neurol Neurosurg Psychiatry 23, 56-62.

Kaiser, R.H., Andrews-Hanna, J.R., Wager, T.D., Pizzagalli, D.A., 2015. Large-Scale Network Dysfunction in Major Depressive Disorder: A Meta-analysis of Resting-State Functional Connectivity. JAMA Psychiatry 72, 603-611.

Kito, S., Fujita, K., Koga, Y., 2008. Regional cerebral blood flow changes after low-frequency transcranial magnetic stimulation of the right dorsolateral prefrontal cortex in treatment-resistant depression. Neuropsychobiology 58, 29-36.

Klumpp, H., Angstadt, M., Phan, K.L., 2012. Insula reactivity and connectivity to anterior cingulate cortex when processing threat in generalized social anxiety disorder. Biol Psychol 89, 273-276.

Kronhaus, D.M., Lawrence, N.S., Williams, A.M., Frangou, S., Brammer, M.J., Williams, S.C., Andrew, C.M., Phillips, M.L., 2006. Stroop performance in bipolar disorder: further evidence for abnormalities in the ventral prefrontal cortex. Bipolar Disord 8, 28-39.

Lam, R.W., Chan, P., Wilkins-Ho, M., Yatham, L.N., 2008. Repetitive transcranial magnetic stimulation for treatment-resistant depression: a systematic review and metaanalysis. Can J Psychiatry 53, 621-631.

Leech, R., Kamourieh, S., Beckmann, C.F., Sharp, D.J., 2011. Fractionating the default mode network: distinct contributions of the ventral and dorsal posterior cingulate cortex to cognitive control. J Neurosci 31, 3217-3224.

Li, C.T., Chen, M.H., Juan, C.H., Huang, H.H., Chen, L.F., Hsieh, J.C., Tu, P.C., Bai, Y.M., Tsai, S.J., Lee, Y.C., Su, T.P., 2014. Efficacy of prefrontal theta-burst stimulation in refractory depression: a randomized sham-controlled study. Brain 137, 2088-2098.

Lisanby, S.H., Husain, M.M., Rosenquist, P.B., Maixner, D., Gutierrez, R., Krystal, A., Gilmer, W., Marangell, L.B., Aaronson, S., Daskalakis, Z.J., Canterbury, R., Richelson, E., Sackeim, H.A., George, M.S., 2009. Daily Left Prefrontal Repetitive Transcranial Magnetic Stimulation in the Acute Treatment of Major Depression: Clinical Predictors of Outcome in a Multisite, Randomized Controlled Clinical Trial. Neuropsychopharmacology 34, 522-534.

Liston, C., Chen, A.C., Zebley, B.D., Drysdale, A.T., Gordon, R., Leuchter, B., Voss, H.U., Casey, B.J., Etkin, A., Dubin, M.J., 2014. Default Mode Network Mechanisms of Transcranial Magnetic Stimulation in Depression. Biological psychiatry 76, 517-526.

Manoliu, A., Meng, C., Brandl, F., Doll, A., Tahmasian, M., Scherr, M., Schwerthoffer, D., Zimmer, C., Forstl, H., Bauml, J., Riedl, V., Wohlschlager, A.M., Sorg, C., 2013. Insular dysfunction within the salience network is associated with severity of symptoms and aberrant inter-network connectivity in major depressive disorder. Front Hum Neurosci 7, 930.

Mayberg, H.S., Silva, J.A., Brannan, S.K., Tekell, J.L., Mahurin, R.K., McGinnis, S., Jerabek, P.A., 2002. The functional neuroanatomy of the placebo effect. Am J Psychiatry 159, 728-737.

Montgomery, S.A., Asberg, M., 1979. A new depression scale designed to be sensitive to change. Br J Psychiatry 134, 382-389.

Mundt, J.C., Marks, I.M., Shear, M.K., Greist, J.H., 2002. The Work and Social Adjustment Scale: a simple measure of impairment in functioning. Br J Psychiatry 180, 461-464.

Murphy, E.R., Barch, D.M., Pagliaccio, D., Luby, J.L., Belden, A.C., 2016. Functional connectivity of the amygdala and subgenual cingulate during cognitive reappraisal of emotions in children with MDD history is associated with rumination. Dev Cogn Neurosci 18, 89-100.

Norman, K.A., Polyn, S.M., Detre, G.J., Haxby, J.V., 2006. Beyond mind-reading: multi-voxel pattern analysis of fMRI data. Trends Cogn Sci 10, 424-430.

O'Reardon, J.P., Solvason, H.B., Janicak, P.G., Sampson, S., Isenberg, K.E., Nahas, Z., McDonald, W.M., Avery, D., Fitzgerald, P.B., Loo, C., Demitrack, M.A., George, M.S., Sackeim, H.A., 2007. Efficacy and safety of transcranial magnetic stimulation in the acute treatment of major depression: a multisite randomized controlled trial. Biol Psychiatry 62, 1208-1216.

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

Palmer, S.M., Crewther, S.G., Carey, L.M., Team, S.P., 2014. A meta-analysis of changes in brain activity in clinical depression. Front Hum Neurosci 8, 1045.

Pecina, M., Bohnert, A.S., Sikora, M., Avery, E.T., Langenecker, S.A., Mickey, B.J., Zubieta, J.K., 2015. Association Between Placebo-Activated Neural Systems and Antidepressant Responses: Neurochemistry of Placebo Effects in Major Depression. JAMA Psychiatry 72, 1087-1094.

Power, J.D., Barnes, K.A., Snyder, A.Z., Schlaggar, B.L., Petersen, S.E., 2012. Spurious but systematic correlations in functional connectivity MRI networks arise from subject motion. NeuroImage 59, 2142-2154.

Price, J.L., Drevets, W.C., 2012. Neural circuits underlying the pathophysiology of mood disorders. Trends in cognitive sciences 16, 61-71.

Rubinov, M., Bullmore, E., 2013. Fledgling pathoconnectomics of psychiatric disorders. Trends Cogn Sci 17, 641-647.

Rush, A.J., Trivedi, M.H., Ibrahim, H.M., Carmody, T.J., Arnow, B., Klein, D.N., Markowitz, J.C., Ninan, P.T., Kornstein, S., Manber, R., Thase, M.E., Kocsis, J.H., Keller, M.B., 2003. The 16-Item Quick Inventory of Depressive Symptomatology (QIDS), clinician rating (QIDS-C), and self-report (QIDS-SR): a psychometric evaluation in patients with chronic major depression. Biol Psychiatry 54, 573-583.

Sackeim, H.A., 2001. The definition and meaning of treatment-resistant depression. J Clin Psychiatry 62 Suppl 16, 10-17.

Salomons, T.V., Dunlop, K., Kennedy, S.H., Flint, A., Geraci, J., Giacobbe, P., Downar, J., 2014. Resting-state cortico-thalamic-striatal connectivity predicts response to dorsomedial prefrontal rTMS in major depressive disorder. Neuropsychopharmacology 39, 488-498.

Satterthwaite, T.D., Cook, P.A., Bruce, S.E., Conway, C., Mikkelsen, E., Satchell, E., Vandekar, S.N., Durbin, T., Shinohara, R.T., Sheline, Y.I., 2015. Dimensional depression severity in women with major depression and post-traumatic stress disorder correlates with fronto-amygdalar hypoconnectivty. Mol Psychiatry.

Schutter, D.J., 2009. Antidepressant efficacy of high-frequency transcranial magnetic stimulation over the left dorsolateral prefrontal cortex in double-blind sham-controlled designs: a meta-analysis. Psychol Med 39, 65-75.

Sheehan, D.V., Lecrubier, Y., Sheehan, K.H., Amorim, P., Janavs, J., Weiller, E., Hergueta, T., Baker, R., Dunbar, G.C., 1998. The Mini-International Neuropsychiatric Interview (M.I.N.I.): the development and validation of a structured diagnostic psychiatric interview for DSM-IV and ICD-10. J Clin Psychiatry 59 Suppl 20, 22-33;quiz 34-57.

Sheline, Y.I., Barch, D.M., Price, J.L., Rundle, M.M., Vaishnavi, S.N., Snyder, A.Z., Mintun, M.A., Wang, S., Coalson, R.S., Raichle, M.E., 2009. The default mode network and self-referential processes in depression. Proc Natl Acad Sci U S A 106, 1942-1947.

Slotema, C.W., Blom, J.D., Hoek, H.W., Sommer, I.E., 2010. Should we expand the toolbox of psychiatric treatment methods to include Repetitive Transcranial Magnetic Stimulation (rTMS)? a meta-analysis of the efficacy of rTMS in psychiatric disorders. J Clin Psychiatry.

Spitzer, R.L., Kroenke, K., Williams, J.B., Lowe, B., 2006. A brief measure for assessing generalized anxiety disorder: the GAD-7. Arch Intern Med 166, 1092-1097.

Stern, E.R., Welsh, R.C., Fitzgerald, K.D., Gehring, W.J., Lister, J.J., Himle, J.A., Abelson, J.L., Taylor, S.F., 2011. Hyperactive Error Responses and Altered Connectivity in Ventromedial and Frontoinsular Cortices in Obsessive-Compulsive Disorder. Biol Psychiatry 69, 583-591.

Straub, J., Metzger, C.D., Plener, P.L., Koelch, M.G., Groen, G., Abler, B., 2017. Successful group psychotherapy of depression in adolescents alters fronto-limbic resting-state connectivity. J Affect Disord 209, 135-139.

Taylor, S.F., Bhati, M.T., Dubin, M.J., Hawkins, J.M., Lisanby, S.H., Morales, O., Reti, I.M., Sampson, S., Short, E.B., Spino, C., Watcharotone, K., Wright, J., 2017. A naturalistic, multi-site study of repetitive transcranial magnetic stimulation therapy for depression. J Affect Disord 208, 284-290.

Tzourio-Mazoyer, N., Landeau, B., Papathanassiou, D., Crivello, F., Etard, O., Delcroix, N., Mazoyer, B., Joliot, M., 2002. Automated anatomical labeling of activations in SPM using a macroscopic anatomical parcellation of the MNI MRI single-subject brain. Neuroimage 15, 273-289.

Wang, Y.L., Yang, S.Z., Sun, W.L., Shi, Y.Z., Duan, H.F., 2016. Altered functional interaction hub between affective network and cognitive control network in patients with major depressive disorder. Behav Brain Res 298, 301-309.

Weigand, A., Horn, A., Caballero, R., Cooke, D., Stern, A.P., Taylor, S.F., Press, D., Pascual-Leone, A., Fox, M.D., 2017. Prospective validation that subgenual connectivity predicts antidepressant efficacy of transcranial magnetic stimulation sites. Biological Psychiatry, published online 10 Nov 2017.

Worsley, K.J., Poline, J.B., Friston, K.J., Evans, A.C., 1997. Characterizing the response of PET and fMRI data using multivariate linear models. Neuroimage 6, 305-319.

Yang, Y., Gu, H., Zhan, W., Xu, S., Silbersweig, D., ., Stern, E., 2002. Simultaneous perfusion and BOLD imaging using reverse spiral scanning at 3T: characterization of functional contrast and susceptibility artifacts. Magn Reson Med 48, 278-289.

Yarkoni, T., Poldrack, R.A., Nichols, T.E., Van Essen, D.C., Wager, T.D., 2011. Large-scale automated synthesis of human functional neuroimaging data. Nat Methods 8, 665-670.

Yeo, B.T., Krienen, F.M., Sepulcre, J., Sabuncu, M.R., Lashkari, D., Hollinshead, M., Roffman, J.L., Smoller, J.W., Zollei, L., Polimeni, J.R., Fischl, B., Liu, H., Buckner, R.L., 2011. The organization of the human cerebral cortex estimated by intrinsic functional connectivity. Journal of neurophysiology 106, 1125-1165.

Yip, A.G., George, M.S., Tendler, A., Roth, Y., Zangen, A., Carpenter, L.L., 2017. 61% of unmedicated treatment resistant depression patients who did not respond to acute TMS treatment responded after four weeks of twice weekly deep TMS in the Brainsway pivotal trial. Brain Stimul 10, 847-849.

Zhu, X., Wang, X., Xiao, J., Liao, J., Zhong, M., Wang, W., Yao, S., 2012. Evidence of a dissociation pattern in resting-state default mode network connectivity in first-episode, treatment-naive major depression patients. Biol Psychiatry 71, 611-617.

Table 1: Demographic and clinical characteristics of subjects

	Active rTMS (n = 16)		Sham rTMS (n = 16)			
	Mean	S.D.	Mean	S.D.	t, χ^2	P =
Age, y	46.9	10.7	44.13	11.1	0.71	0.48
Males/females, n	5/11		6/10		0.14	0.71
SES	2.63	0.88	2.38	0.72	0.88	0.39
ATHF -life	5.06	3.17	5.56	2.13	0.52	0.60
ATHF –current	2.56	1.75	2.94	1.77	0.60	0.55
Baseline scores						
MADRS	25.4	5.7	21.9	3.1	2.14	*0.04*
HRSD-17	16.0	3.9	13.1	2.3	2.56	*0.02*
QIDS-SR	16.5	4.2	13.9	3.2	1.94	*0.06*
GAD-7	9.9	5.6	8.1	5.7	0.91	0.37
WSAS	27.4	8.0	26.6	5.8	0.30	0.76
GAF	52.6	4.7	55.6	4.3	1.86	*0.07*

Abbreviations: SES: Socio-Economic Status; ATHF-life, current: Antidepressant Treatment History Form, total number of medication trials and augmentation strategies, lifetime, current; MADRS: Montgomery-Asberg Depression Rating Scale; HRSD-17: Hamilton Rating Scale for Depression, 17-item version; QIDS-SR: Quick Inventory of Depressive Symptoms, Self-Rated; GAD-7: Generalized Anxiety Disorder Assessment; WSAS: Work and Social Adjustment Scale; GAF: Global Assessment of Function.

Figure Legends

Figure 1: Change in sgACC connectivity for responders versus non-responders

(A) Areas of connectivity changes for the sgACC seed are shown, contrasting responders versus non-responders during Phase 1. A significant focus occurred in the left inferior parietal lobule (IPL; -54, -55, 49; k=126, Z=4.37,p=0.005 $FWE_{corr\text{-}cluster}$) and the left orbital frontal cortex (OFC; -30, 50, -11; k=87, Z=3.93, p=0.027 $FWE_{corr\text{-}cluster}$). Foci are displayed at an uncorrected threshold of p<0.001. (B) Extractions for these regions show the change in connectivity from scan 1 to scan 2, for each focus.

Figure 2: Change in connectivity for responders versus non-responders, seed-to-network analysis

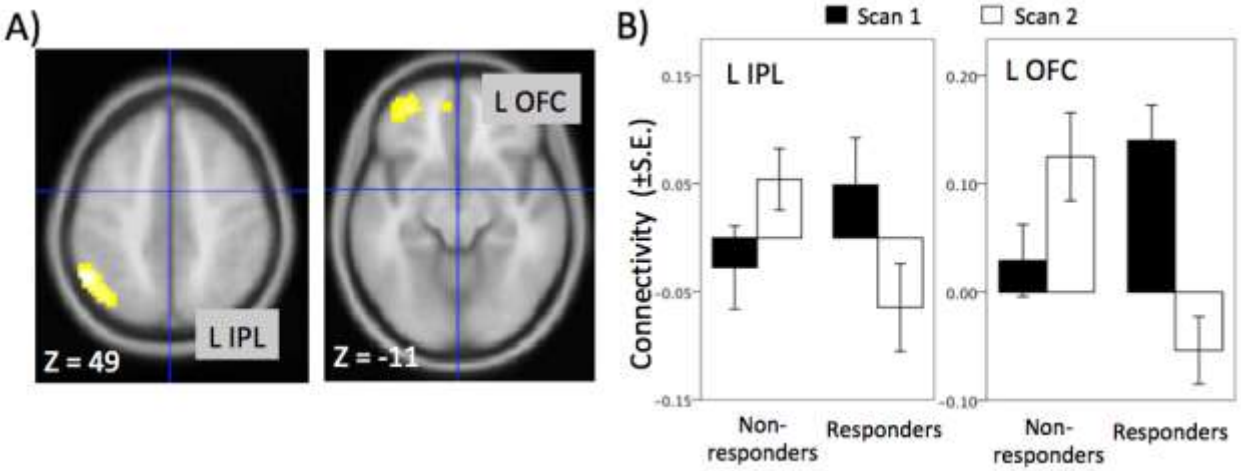
Seed-to-network connectivity is reduced in responders during Phase 1 for the sgACC seed (A) to the affective network (F[1,28] = 13.17, p =0.001), fronto-parietal network F[1,28] = 6.57, p =0.016) and default mode network (DMN; F[1,28] = 6.15, p =0.019). (B) Seed-to-network connectivity changes in the sgACC are demonstrated separately for the active rTMS responders (compared to all non-responders) for AN (F[1,23] = 6.77, p =0.016), FPN (F[1,23] = 11.6, p =0.002) and DMN (F[1,23] = 5.65, p =0.026); and (C) the sham responders (compared to all non-responders) for AN (F[1,21] = 9.33, p =0.006), FPN (F[1,21] = 1.27, p =0.27) and DMN (F[1,21] = 3.29, p =0.084). S1 = Scan 1; S2 = Scan 2.

Figure 3: Baseline connectivity predicting treatment response

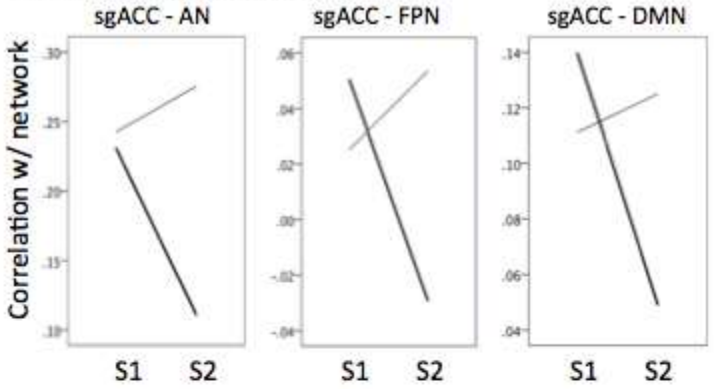
Treatment responders have (A) less connectivity between the PCC seed and the right anterior insula (aIns; 42, 17, -14; k=60, Z=4.84 p=0.035 $FWE_{corr\text{-}peak}$) and right inferior frontal gyrus (IFG, 36, 32, -23, k=117, Z=4.24, p=0.008 $FWE_{corr\text{-}cluster}$). (B) From a database of 1000 healthy subjects (neurosynth.org; (Yarkoni et al., 2011) connectivity to the right aIns is normally negative, (C) which is the pattern of connectivity for responders (green circles) depicted in the scattergram.

Highlights

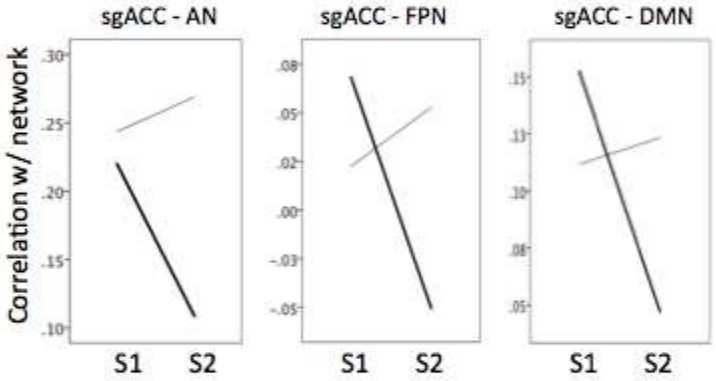
- Subgenual connectivity decreased with response to sham and active treatment
- Connectivity decreased within affective network and with default mode network
- No differential effect of active versus sham TMS on connectivity
- sgACC connectivity change may be a final common pathway in depression treatment
- Baseline default mode connectivity predicted symptom change



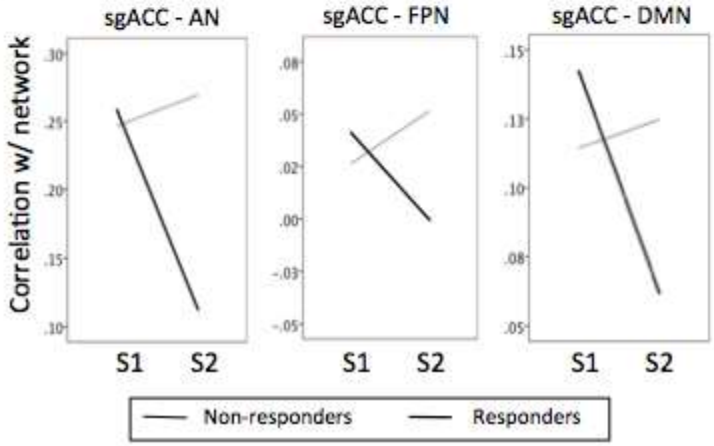
A) All responders vs non-responders

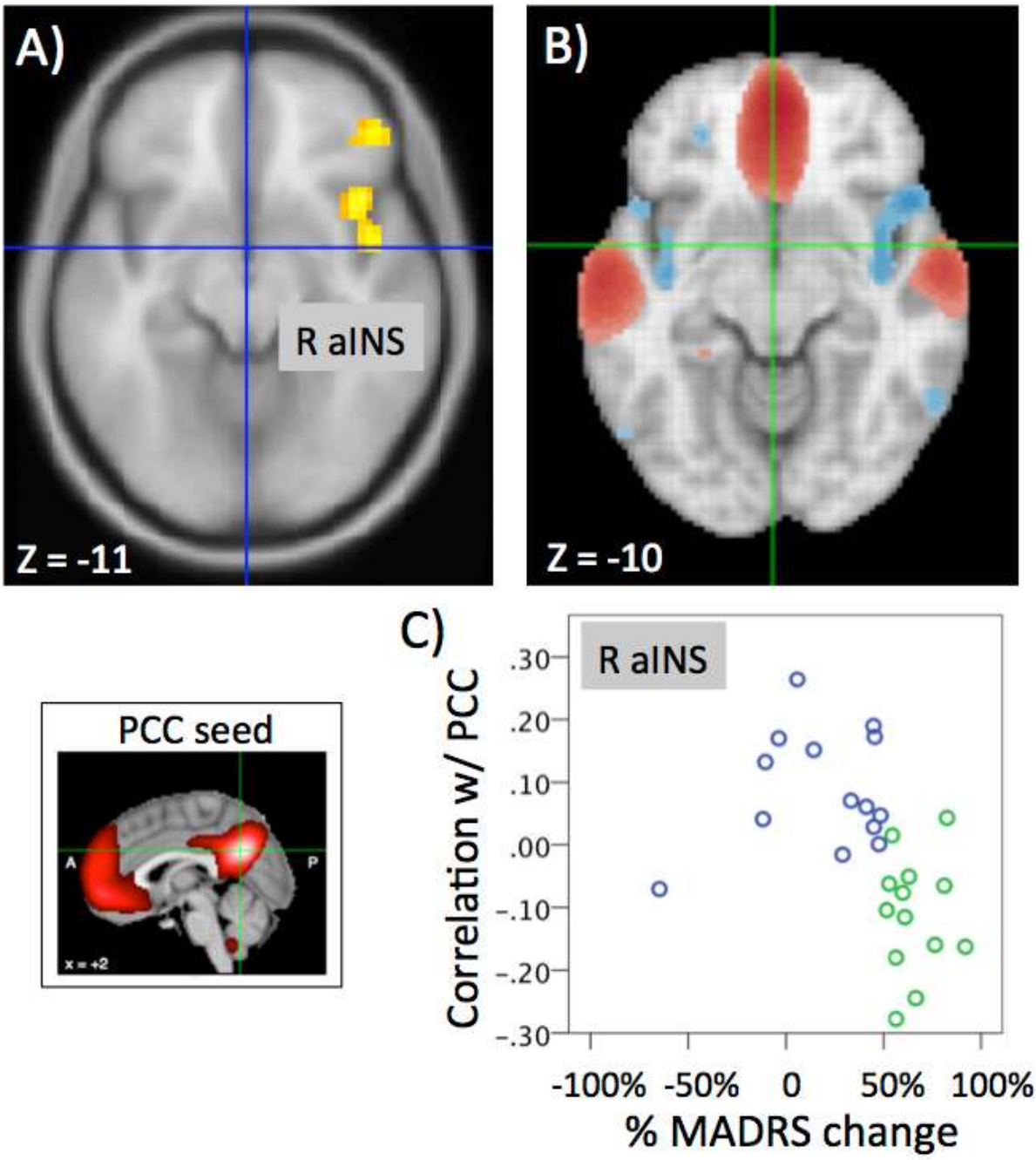


B) rTMS responders vs non-responders



C) Sham responders vs non-responders




A)
R aINS
Z = -11
B)
Z = -10
C)
R aINS
PCC seed
A
P
x = +2
Correlation w/ PCC
.30
.20
.10
.00
-.10
-.20
-.30
-100%
-50%
0
50%
100%
% MADRS change