Published in final edited form as:

J Psychiatr Res. 2013 April ; 47(4): 494–504. doi:10.1016/j.jpsychires.2012.11.010.

Effects of intensive cognitive-behavioral therapy on cingulate neurochemistry in obsessive–compulsive disorder

Joseph O'Neill^{a,*}, Eda Gorbis^{b,c}, Jamie D. Feusner^b, Jenny C. Yip^c, Susanna Chang^a, Karron M. Maidment^b, Jennifer G. Levitt^a, Noriko Salamon^d, John M. Ringman^e, and Sanjaya Saxena^f

^aDivision of Child and Adolescent Psychiatry, UCLA Semel Institute for Neurosciences, Los Angeles, CA 90024, USA

^bDivision of Adult Psychiatry, UCLA Semel Institute for Neurosciences, Los Angeles, CA 90024, USA

^cWestwood Institute for Anxiety Disorders, Los Angeles, CA 90024, USA

^dUCLA Department of Radiological Sciences, Los Angeles, CA 90024, USA

^eUCLA Department of Neurology, Easton Center for Alzheimer's Disease Research, Los Angeles, CA 90024, USA

^fUCSD Department of Psychiatry and Department of Veterans Affairs Medical Center, La Jolla, CA 92161, USA

Abstract

The neurophysiological bases of cognitive-behavioral therapy (CBT) for obsessive–compulsive disorder (OCD) are incompletely understood. Previous studies, though sparse, implicate metabolic changes in pregenual anterior cingulate cortex (pACC) and anterior middle cingulate cortex (aMCC) as neural correlates of response to CBT. The goal of this pilot study was to determine the relationship between levels of the neurochemically interlinked metabolites glutamate + glutamine (Glx) and *N*-acetyl-aspartate + *N*-acetyl-aspartyl-glutamate (tNAA) in pACC and aMCC to pretreatment OCD diagnostic status and OCD response to CBT. Proton magnetic resonance spectroscopic imaging (¹H MRSI) was acquired from pACC and aMCC in 10 OCD patients at baseline, 8 of whom had a repeat scan after 4 weeks of intensive CBT. pACC was also scanned (baseline only) in 8 age-matched healthy controls. OCD symptoms improved markedly in 8/8 patients after CBT. In right pACC, tNAA was significantly lower in OCD patients than controls at baseline and then increased significantly after CBT. Baseline tNAA also correlated with post-CBT change in OCD symptom severity. In left aMCC, Glx decreased significantly after intensive CBT. These findings add to evidence implicating the pACC and aMCC as loci of the metabolic effects of CBT in OCD, particularly effects on glutamatergic and *N*-acetyl compounds. Moreover, these metabolic responses occurred after just 4 weeks of intensive CBT, compared to 3 months for standard weekly CBT. Baseline levels of tNAA in the pACC may be associated with response to CBT for OCD. Lateralization of metabolite effects of CBT, previously observed in subcortical nuclei and white matter, may also occur in cingulate cortex. Tentative mechanisms for these effects are discussed. Comorbid depressive symptoms in OCD patients may have contributed to metabolite effects, although baseline and post-CBT change in depression ratings varied with choline-compounds and *myo*-inositol rather than Glx or tNAA.

Keywords

Magnetic resonance spectroscopy; Obsessive—compulsive disorder; Cognitive-behavioral therapy; Cingulate cortex; Glutamate; NAA

1. Introduction

Cognitive-behavioral therapy (CBT) for obsessive—compulsive disorder (OCD) regularly yields therapeutic responses that rival or exceed those of drug treatments, are achieved more rapidly, and persist after discontinuation of therapy (Foa et al., 2005). Previous [^{18}F]-fluorodeoxyglucose positron emission tomography (^{18}FDG -PET) studies of adult OCD patients (Baxter et al., 1987, 1988; Kwon et al., 2003; Nordahl et al., 1989; Perani et al., 1995; Sawle et al., 1991; Saxena et al., 2001; Swedo et al., 1989) identified regional abnormalities in brain metabolism that may respond to CBT. These studies found above-normal pretreatment glucose metabolic rates (GMR) in caudate, thalamus, orbitofrontal cortex and anterior cingulate cortex, which are brain structures that form functional neural circuits thought to be hyperactive in OCD (reviewed in Saxena et al., 2001). Several studies (Baxter et al., 1992; Freyer et al., 2011; Nabeyama et al., 2008; Nakatani et al., 2003; Schwartz et al., 1996; Yamanishi et al., 2009), including ours (Saxena et al., 2009a), found significant changes in glucose metabolism or blood flow in these structures after CBT for OCD. Hence, the therapeutic effects of CBT appear to be consistently associated with changes in regional brain energetic metabolism.

Aspects of regional brain energy metabolism are illuminated by proton magnetic resonance spectroscopy (^1H MRS), a neuroimaging modality that is safer and better tolerated than PET. Metabolites assayed in living human brain at clinical field strength (0.5–3 T) include the two most abundant CNS amino acids: *N*-acetyl-aspartate (NAA) and glutamate (Glu). Under clinical conditions, NAA is nearly always measured together with spectrally-overlapping *N*-acetyl-aspartyl-glutamate (NAAG) and Glu is frequently measured together with overlapping glutamine (Gln); NAA + NAAG is abbreviated “tNAA” (total NAA) and Glu + Gln is abbreviated “Glx” thereby. ^1H MRS has linked GMR to tNAA (O'Neill et al., 2000) and to Glx (Pfund et al., 2000), while ^{13}C MRS has linked GMR to NAA proper (Moreno et al., 2001) and to Glu proper (Sibson et al., 1998). Hence, it is conceivable that elevated GMR in OCD leads to abnormal regional tNAA and/or Glx levels. Likewise, CBT-induced GMR changes may induce or accompany changes in tNAA and/or Glx.

MRS studies of adult (reviewed in O'Neill and Schwartz, 2005; Saxena et al., 2009b; Brennan et al., 2012) and pediatric (reviewed in MacMaster et al., 2008) OCD have, in fact, found abnormal pretreatment levels of tNAA, Glu or Glx, or other MRS metabolites or their ratios to creatine + phosphocreatine (Cr) in cingulate cortex, basal ganglia, thalamus, or their interconnecting white matter (adult: Bartha et al., 1998; Ebert et al., 1997; Jang et al., 2006; Kitamura et al., 2006; Mohamed et al., 2007; Starck et al., 2008; Sumitani et al., 2007; Whiteside et al., 2006; Yücel et al., 2007, 2008; Zurowski et al., 2007; pediatric: Fitzgerald et al., 2000; Mirza et al., 2006; Rosenberg et al., 2000, 2001, 2004; Smith et al., 2003). Some of these findings involved one or more subregions of the cingulate cortex, which we shall designate using the standard nomenclature of Vogt (2009; see also O'Neill et al., 2009). In right pregenual anterior cingulate cortex (pACC), tNAA/Cr was below normal in untreated adult OCD and correlated negatively with OCD symptom severity (Ebert et al., 1997) measured by the Yale–Brown Obsessive–Compulsive Scale (Y-BOCS; Goodman et al., 1989). Again in pretreatment adult OCD, Zurowski et al. (2007) found above-normal Glu in midline (left + right) pACC. In contrast, Glx was below normal in midline pACC in pediatric OCD (Rosenberg et al., 2004), an effect associated with the GRIN2B gene coding

for the *N*-methyl-D-aspartate (NMDA) Glu receptor (Arnold et al., 2009). This represents key evidence favoring the glutamatergic hypothesis of pediatric OCD (Rosenberg and Keshavan, 1998). NAA and Glu are linked by the intraneuronal synthesis ($\text{NAA} + \text{Glu} \rightarrow \text{NAAG}$; Cangro et al., 1987) and the extracellular decomposition ($\text{NAAG} \rightarrow \text{NAA} + \text{Glu}$; Robinson et al., 1987) of NAAG, whereby Glu from the latter reaction is believed to be produced faster than presynaptic vesicular release of free Glu (Rojas et al., 2002). Glu and Gln, the two contributors to the Glx peak, regularly interconvert and are exchanged between neurons and astrocytes (Danbolt, 2001; Hertz and Zielke, 2004; Petroff et al., 2000). Hence, glutamatergic disturbances in OCD may manifest as abnormalities not only in Glx but also in tNAA, as seen in MRS data, including from cingulate cortex.

There have been fewer MRS studies of treatment response in OCD. Jang et al. (2006) found that tNAA/Cr in frontal white matter increased after treatment with serotonin reuptake inhibitors (SRIs). Caudate Glx dropped in response to the SRI paroxetine (Bolton et al., 2001; Moore et al., 1998; Rosenberg et al., 2000) in children with OCD. In one study (Mohamed et al., 2007), OCD patients who were non-responders to SRIs had lower tNAA/Cr in right “basal ganglia” (apparently caudate, globus pallidus, or internal capsule) than responders and healthy controls, as well as higher Cho/Cr in right thalamus than responders. The one published study of CBT for pediatric OCD (Benazon et al., 2003) found no effects on any MRS metabolite, possibly because the study examined only left caudate, leaving out all other brain regions. In adult OCD patients, in contrast, Whiteside et al. (2012) saw tNAA in left caudate increase after CBT and Zurowski et al. (2007) observed declines in midline pACC Glu and in choline-compounds (Cho) in right “ventral striatum” after long-term (3-month) weekly CBT. The same group (Zurowski et al., 2012) acquiring from a voxel containing right orbital frontal cortex and white matter, showed that lower baseline myo-inositol (mI) predicted greater drop in Y-BOCS score following 3-month CBT. Thus, MRS studies of OCD treatment, particularly CBT, are scarce, but there is evidence that treatment may alter regional levels of MRS metabolites, including in cingulate cortex.

The present pilot study used the magnetic resonance spectroscopic imaging (MRSI) variant of ^1H MRS to further explore tNAA and Glx in cingulate cortex in patients before and after brief intensive CBT. Among other aims, we sought to identify neuro-metabolite correlates of the significant increase in GMR we observed in right “dorsal anterior cingulate cortex” of OCD patients after intensive CBT (Saxena et al., 2009a); an increase that correlated significantly with pre- to post-treatment decrease in Y-BOCS scores. In Vogt's (2009) more systematic nomenclature, the dorsal anterior cingulate cortex consists of approximately half pACC and half anterior middle cingulate cortex (aMCC). The pACC is associated with anxiety and other negative emotions (Bush et al., 2000; Devinsky et al., 1995; Whalen et al., 1998), while the cingulate motor area within the aMCC is involved in internal selection of voluntary movements (Picard and Strick, 1996). CBT both reduces anxieties and strengthens volitional control and, hence, could elicit metabolite effects in either or both subregions. The MRSI used in this study had higher spatial resolution than earlier single-voxel MRS studies, enabling us to sample pACC and aMCC separately and bilaterally. This allowed us to examine possible lateralized effects of CBT on OCD brain metabolism, such as have been seen in subcortical nuclei (Baxter et al., 1992; Nakatani et al., 2003; Schwartz et al., 1996). Other aims included evaluation of pretreatment abnormalities in tNAA and Glx levels in OCD, and of relationships of these abnormalities to baseline symptom severity and treatment response. Thus, we sought to identify neuroanatomic loci and neurochemical bases of and the effects of CBT in OCD, as well as objective baseline metabolic measures associated with treatment response. We hypothesized such effects would be present in pACC and aMCC.

2. Methods

2.1. Subjects

Ten patients with DSM-IV OCD (all outpatient; 5 male; mean $\pm$ std. age 36.2 ± 8.9 years; Table 1) underwent baseline MRSI scans. Eight of these patients (4 male; 37.8 ± 8.9 years) underwent a second scan after 4 weeks of intensive daily CBT. Diagnoses were made by clinical interview and confirmed using the Structured Clinical Interview for DSM-IV (SCID; First et al., 1996). For inclusion into the study, OCD patients needed to have a pretreatment Y-BOCS score ≥ 16 , indicating at least moderate OCD symptom severity. All subjects were in good physical health. Subjects with major medical conditions, current or recent substance abuse, or any other concurrent Axis I diagnosis except major depressive disorder and dysthymia were excluded. Depressive symptoms were evaluated using the Hamilton Depression Rating Scale (HamD; Hamilton, 1960). Patients with depressive symptoms were only admitted if the depression was considered “secondary to OCD” (5 patients, Table 1). Secondary meant that OCD and not depression was the primary psychiatric diagnosis, that the patient's OCD was a probable source of the depression, and that onset of OCD symptoms preceded onset of depression symptoms in time. History of Axis I diagnosis in first-degree relatives was not assessed and was not exclusionary. Concurrent psychoactive medication was permitted, but all medication doses were unchanged for at least 12 weeks prior to starting CBT and were not changed during the study. Five patients were being treated with SRIs, three with benzodiazepines, and one each with an atypical neuroleptic (though not for tic-related OCD), an atypical antidepressant, or a stimulant (subject with history of ADHD), while five patients were receiving no psychotropic medication (Table 1; numbers do not sum to 10 due to polypharmacy in some cases). The Y-BOCS and HamD were administered 1 week or less before starting, and again after completing CBT, by a trained rater not involved in treatment. Additionally, the Clinical Global Impression-Severity (CGI-S; Guy, 1976) was administered at baseline and the Clinical Global Impression-Improvement (CGI-I; Guy, 1976) was administered post-CBT. The study was approved by the UCLA Human Subjects Committee and written informed consent was obtained from all subjects.

OCD MRSI data from the pACC were compared to data (baseline only) acquired contemporaneously, under identical conditions on the same scanner, from a group of 8 healthy controls (2 male; 38.3 ± 9.2 years) from another study (Ringman et al., 2006). Controls had no history of any psychiatric disorder or substance abuse and no current major medical conditions or psychoactive medications. There were no healthy comparison data for the aMCC. The limited scope of this pilot investigation precluded obtaining aMCC data from controls or data from other mood disorder-relevant brain regions such as the subgenual anterior cingulate cortex (sACC) in patients or controls. Scan-rescan data, however, were available from the more posteriorly located dorsal posterior cingulate cortex (dPCC; midline, i.e., left + right-hemisphere, acquisition) subregion in a group of 5 additional healthy controls from another investigation (O'Neill et al., 2010) being conducted contemporaneously on the same scanner with the same MRSI pulse sequence. These subjects (2 male, 3 female; mean age 36.0 ± 8.7 years, range 28–46 years) were scanned and rescanned under identical conditions at intervals of 23–37 days. The same MRSI post-processing methods were applied as for the present OCD investigation.

2.2. Cognitive-behavioral therapy (CBT)

All OCD patients received 90-min individual CBT sessions, 5 days a week for 4 weeks, with a therapist with expertise in CBT for OCD (EG, JCY). The method combined two manualized treatments: standard exposure and response prevention (ERP) with 4 h/day homework exercises (Kozak and Foa, 1997) and the Four Steps Method (Schwartz, 1996) that includes mindful awareness and cognitive techniques. ERP involved graded exposures

to both imaginal and real situations and stimuli that typically provoked compulsive behaviors or avoidance, accompanied by prevention of compulsions or avoidance. Intensive CBT was conducted for every patient according to a set protocol and sequence (Foa et al., 2005). Sessions 1–3 included a comprehensive behavioral assessment; education for the patient in self-monitoring of obsessions, compulsions and triggers; and a discussion of the rationale and specific goals of CBT for each individual. A hierarchy of feared and avoided situations and stimuli was created for each patient, using a “subjective units of distress” scale. Sessions 4–15 consisted of *in vivo* and imagined ERP exposures of gradually increasing difficulty, as well as review of daily homework assignments. Sessions 16–20 focused on relapse prevention and included continued ERP practice, cognitive restructuring, and assessment of progress. Patients were also taught to recognize internal and external cues that triggered their OCD symptoms (mindful awareness), so that they could anticipate their over-appraisal of fear and anxiety when their obsessions occurred. Response to treatment was defined a priori as a 35% drop in Y-BOCS score and a CGI rating of “much improved” or “very much improved”, the standard response criteria used in clinical trials for OCD (Pallanti et al., 2002).

2.3. MR acquisition

Whole-brain structural MRI and water-suppressed ¹H MRSI were acquired together in 1.5-h sessions at 1.5 T on a Siemens Sonata scanner using a quadrature headcoil within 1 week before starting and then again within 1 week after completing CBT for patients and at baseline only for controls. MRSI was acquired with a point-resolved spectroscopy (PRESS) sequence with repetition-time (TR) of 1500 ms, echo-time (TE) of 30 ms, and 8 excitations from two bilateral 9 mm-thick 2D arrays (“slabs”; see Fig. 1) of 11 × 11 mm² voxels. One slab sampled pACC, the other aMCC. The first slab was oriented parallel to the cantho-meatal line. Its “PRESS box”—the acquisition volume from which usable spectra could be obtained—measured 4×4 voxels in cross-section in every subject. The pACC PRESS box straddled the longitudinal midline and was positioned with its posterior end at the rostrum of the corpus callosum, set just far back enough to prevent the anterior end of the PRESS box from contacting extracranial tissue. The slab was also centered dorsoventrally about the callosal rostrum (Fig. 1). Thus, the posterior voxels of this box sampled pACC, while the anterior voxels sampled mesial superior frontal cortex. The aMCC slab was oriented parallel to the dorsal anterior corpus callosum. Its PRESS box straddled the midline and was centered on supracallosal cingulate. The PRESS volume varied in size (typically 6 voxels mesial-lateral by 10 voxels anterior-posterior) and sampled cingulate cortex longitudinally without touching extrabrain tissue. It extended approximately from the paracingulate portions of the pACC at its rostral end until dPCC or precuneus at its caudal end. MRSI voxel selection was restricted to the anterior (aMCC) portions of the slab. Acquisition was immediately repeated for each slab without water-suppression (1 excitation). A neuroradiologist (NS) reviewed all structural MRI scans; we excluded any subjects with clinically significant abnormalities.

2.4. MR post-processing

MR spectra were fit automatically with the LCModel commercial software package (Provencher, 2001) yielding levels of tNAA, Glx, Cr, Cho, and mI referenced to unsuppressed water and expressed in institutional units (IU). After segregation of the whole-brain MRI into gray matter, white matter, and CSF binary masks (Shattuck et al., 2001), the MRSI Voxel Picker (MVP) package (Seese et al., 2011) was used for MRI/MRSI co-processing. For each MRSI slab, MVP reconstructed the MRI and the binary masks into the space of the corresponding MRSI PRESS volume; computed the volume percent (vol%) gray matter, white matter, and CSF in each MRSI voxel; corrected the LCModel-derived levels of each metabolite for voxel CSF content; automatically rejected spectra not meeting

quality control criteria (linewidth = 0.1 ppm and signal-to-noise ratio = 3); and displayed results on a guided user interface (GUI). Additionally, within spectra, individual metabolite peaks were rejected that were not considered reliable by LCModel (standard deviation of metabolite signal > 20%). Using the MVP GUI, voxels were selected by a blinded operator in left and right pACC and aMCC. Within each region, MVP averaged together the values for all MRSI voxels that satisfied the above criteria, and for which tissue content was ≥ 60 vol% gray matter.

2.5. Statistical analyses

Given the small size of the samples, a non-parametric approach to statistics was adopted. This was achieved by rank-transforming regional metabolite level and voxel tissue composition values prior to performing the corresponding parametric tests. Between-group differences in baseline MRSI voxel tissue composition (vol% gray matter, vol% white matter) were analyzed with independent *T*-tests. Effects of CBT on voxel tissue composition were analyzed with paired *T*-tests comparing pre- and post-treatment values. No separate analyses were performed for vol% CSF since it is a linear combination of vol% gray and white matter. To account for multiple comparisons of metabolite levels, omnibus tests were performed on the rank-transformed data. For between-group comparisons of baseline pACC metabolite levels, the omnibus test was a repeated-measures multivariate analysis of covariance (R-MANCOVA) on the measures tNAA and Glx, with Hemisphere (two levels: left, right) as within-subjects factor, diagnosis (two levels: OCD, control) as between-subjects factor, and sex as covariate. Sex was used as a covariate due to the relatively higher numbers of female subjects in the control group. If a significant main effect or interaction involving diagnosis was found, R-MANCOVA was followed-up with omnibus protected *post-hoc* comparisons of the metabolite levels between the two groups in each subregion. The *post-hoc* test was a univariate ANCOVA of tNAA or Glx values, with diagnosis as between-subjects factor and sex as covariate. For pre- to post-CBT comparisons of metabolite levels within the OCD group, the omnibus test was a repeated-measures multivariate analysis of variance (R-MANOVA) on the measures tNAA and Glx with subregion (two levels: pACC, aMCC), hemisphere (two levels: left, right), and CBT (two levels: pre-CBT and post-CBT) as within-subjects factors. If a significant main effect or interaction involving CBT was found, R-MANOVA was followed-up with protected *post-hoc* comparisons of the pre- and post-CBT metabolite levels in each subregion. The *post-hoc* test was a paired *T*-test. Spearman correlations (non-parametric) were performed on the untransformed OCD data, between baseline metabolite levels and baseline Y-BOCS scores, between baseline metabolites and pre- to post-CBT change in Y-BOCS, and between change in Y-BOCS and change in metabolites. Criterion for statistical significance was $p < 0.05$. Analyses were performed using the SPSS package (SPSS, Inc.; Chicago, IL).

Although not major endpoints of this study nor part of its specific aims, Cr, Cho, and mI levels were automatically provided by LCModel in each voxel and are of general interest. Therefore, exploratory independent or paired *T*-tests of rank-transformed data were undertaken to analyze effects of OCD and CBT, respectively, on cingulate levels of these metabolites. Similarly, exploratory analyses were performed of the relationships between baseline HamD scores and baseline metabolite levels (tNAA, Glx, Cr, Cho, mI), between baseline metabolite levels and post-CBT change in HamD, and between change in metabolite levels and change in HamD.

3. Results

3.1. Clinical variables and response to CBT

Across the 10 OCD patients receiving pre-CBT clinical assessment and MRSI scans, Y-BOCS scores (Fig. 2) ranged from 22 to 37 indicating “moderate” to “extreme” severity of OCD symptoms. The mean was 28.4 ± 4.4 . All major OCD symptom domains (checking, symmetry, contamination) were represented, except hoarding, which was excluded by design. As is more typical for patients participating in intensive than weekly CBT, patients were highly motivated and were only enrolled if they agreed to embark upon the intensive all-day treatment regime described in Section 2.2 above. Nine patients (all except Patient 9) underwent post-CBT assessment and all exhibited reduced Y-BOCS (Fig. 2), resulting in a highly significant within-subject effect of CBT ($p < 0.0005$). Eight patients (all but Patients 9 and 10) received a post-CBT MRSI scan. Among these 8 patients, mean post-CBT Y-BOCS was 10.8 ± 4.6 (range 5–20), a mean 60.2% decrease from pretreatment severity. Based on a >35% reduction in Y-BOCS score and a CGI rating of much or very much improved (Pallanti et al., 2002), all patients except Patient 1 and non-completer Patient 9 were classified as responders to intensive CBT.

As is typical in our clinical population seeking intensive CBT for OCD and common in OCD patients generally, symptoms of depression (as evidenced by HamD scores) were common in the OCD sample (Fig. 4). Across the 10 OCD patients receiving pre-CBT assessment, HamD scores ranged from 9 to 29, with a mean of 18.5 ± 6.5 . Nine patients (all except Patient 9) underwent post-CBT assessment and all exhibited reduced HamD (Fig. 4), resulting in a highly significant within-subject effect of CBT ($p < 0.01$). Mean post-CBT HamD was 7.4 ± 6.2 (range 1–22), which represents a mean 60.4% decrease from pretreatment severity.

3.2. MRSI voxel tissue composition

Group-mean tissue compositions of MRSI voxels sampled in each cingulate subregion are listed in Table 2. Independent *T*-tests of rank-transformed data revealed no significant differences between OCD patients and controls in vol% gray or white matter in left or right pACC at baseline (all $p > 0.05$). Within OCD patients, paired *T*-tests of rank-transformed data revealed no significant pre-to post-CBT differences in vol% gray or white matter in left or right pACC or aMCC (all $p > 0.05$). Thus, there was no need to use vol% gray matter or vol% white matter as covariates in analyses of effects of OCD or CBT on neurometabolite levels. Note that these gray matter and white matter values refer to the amounts of gray, respectively, white matter inside the MRSI voxels sampled and not to the overall anatomic gray and white matter volumes of the cingulate subregions.

3.3. Comparisons of OCD to control MRSI tNAA and Glx levels in pACC at baseline

Baseline neurometabolite levels for OCD patients and healthy controls are listed in Table 2. R-MANCOVA for tNAA and Glx across left and right pACC covarying sex showed a significant main effect of diagnosis ($F_{2,13} = 4.5$, $p = 0.033$). *Post-hoc* protected ANCOVA covarying for sex found that baseline tNAA in right pACC was significantly lower in OCD patients than in controls ($F_{1,16} = 15.9$, $p = 0.001$), with the mean difference from control levels being 13.8% (Fig. 3). Pre-CBT OCD tNAA in left pACC and Glx in left and right pACC did not differ significantly from control levels (all $p > 0.05$). Cr, Cho, and mI levels did not differ between groups in any region tested.

3.4. Effects of CBT on tNAA and Glx levels in pACC and aMCC within OCD sample

Pre- and post-CBT neurometabolite levels for OCD patients are listed in Table 2. R-MANOVA for tNAA and Glx across left and right pACC and aMCC showed a significant

subregion-by-hemisphere-by-CBT three-way interaction ($F_{2,4} = 57.0, p = 0.001$). *Post-hoc* protected paired *T*-tests revealed a significant pre- to post-CBT increase in tNAA in right pACC ($F_6 = 2.8, p = 0.032$). The mean increase was 10.2% (Fig. 3). Post-CBT and pre-CBT tNAA did not differ significantly in left pACC or left or right aMCC (all $p > 0.05$). *Post-hoc* protected paired *T*-tests also revealed a significant pre- to post-CBT decrease in Glx in left aMCC ($F_6 = -4.3, p = 0.005$). The mean decrease was 22.0% (Fig. 3). Post-CBT and pre-CBT Glx did not differ significantly in right aMCC or left or right pACC (all $p > 0.05$). There were no significant pre- to post-CBT changes detected in Cr, Cho, or mI levels in OCD patients.

3.5. Correlations between pre-CBT tNAA and Glx levels in pACC and aMCC and pre-CBT Y-BOCS score; correlations with post—pre-CBT change in Y-BOCS score

Within OCD patients, pre-CBT Y-BOCS scores did not correlate significantly with pre-CBT tNAA or Glx in left or right pACC or aMCC (all $p > 0.05$). Pre- to post-CBT change in Y-BOCS correlated significantly and negatively with pre-CBT tNAA in right pACC ($R = -0.86, p = 0.007$; Fig. 3), but not in any other cingulate subregion (all $p > 0.05$). Pre-CBT Glx did not correlate significantly with post-pre-CBT change in Y-BOCS in any cingulate subregion (all $p > 0.05$). There were no significant correlations between pre- to post-CBT change in Y-BOCS and pre- to post change in metabolite levels, nor any significant correlations involving baseline Y-BOCS or change in Y-BOCS and baseline Cr, Cho, or mI.

3.6. Correlations between HamD score and metabolite levels in pACC and aMCC

In exploratory analyses within the OCD sample, HamD score at baseline correlated significantly and positively with mI in left pACC ($R = +0.77, p = 0.026$; Fig. 4) and with Cho in left aMCC ($R = +0.69, p = 0.04$). Baseline Cho in left aMCC also correlated (negatively) with pre- to post-CBT change in HamD ($R = -0.79, p = 0.021$). And in right aMCC pre- to post-CBT change in HamD correlated positively with pre- to post-CBT change in both Cho ($R = +0.76, p = 0.028$) and mI ($R = +0.79, p = 0.021$).

3.7. MRSI scan-rescan in dPCC in additional healthy control cohort

Fig. 5 illustrates tNAA and Glx levels at initial (Scan1) and at 23–37-day follow-up (Scan2) acquisitions from midline dPCC in the 5 additional healthy controls from another study conducted contemporaneously on the same MR scanner using the same PRESS MRSI pulse sequence (O'Neill et al., 2010). Group-mean scan-to-scan change was 1.7% for tNAA and -1.8% for Glx. Thus, there was relatively little variability in the major study metabolite endpoints in this cingulate subregion located posterior to the aMCC and pACC target subregions.

4. Discussion

To our knowledge, this study was the first magnetic resonance spectroscopic imaging (MRSI) investigation of the effects of rapid, intensive CBT on levels of *N*-acetyl and glutamatergic neurometabolites in OCD. Treatment was highly effective, with most patients exhibiting strong symptomatic response or even remission, as measured by Y-BOCS and CGI scores. There were four major findings: 1) Pre-CBT levels of tNAA in right pACC in OCD patients were significantly below those in age-matched healthy controls; 2) Right pACC tNAA levels increased significantly with intensive CBT in OCD patients; 3) Glx levels in left aMCC in OCD patients dropped significantly after CBT; and 4) Higher pre-CBT levels of tNAA in right pACC were correlated with greater pre- to post-CBT decline in OCD symptoms. These findings add to evidence of the involvement of the anterior cingulate cortex in OCD and its response to CBT, and suggest differing, and possibly lateralized,

metabolic effects in these two cingulate subregions. Strong clinical and neurometabolic effects were obtained after only 4 weeks of daily, intensive treatment.

4.1. Clinical response of OCD to brief intensive CBT

On average, the OCD sample was severely impaired (mean pre-CBT Y-BOCS 28.4) and therefore well suited for the above-described intensive CBT methods refined at our center for severe and refractory OCD. Except for one, all OCD patients treated showed a marked decline in Y-BOCS score with brief intensive CBT (Fig. 2). All of these patients were rated as “much improved” or “very much improved” on CGI-I. Such responses are typical for intensive CBT for OCD (Foa et al., 2005). Although some patients received psychoactive medication concurrent with CBT, their lack of notable response in the 12 weeks preceding CBT compared to the vigorous response after 4 weeks of intensive CBT, suggests that symptomatic improvement was due primarily to CBT rather than to medication. Thus, the pre- to post-CBT metabolic changes found in our study are most likely due to successful CBT.

4.2. Baseline cingulate neurometabolite levels in OCD

At baseline, tNAA in right pACC, was significantly lower in OCD patients than in controls. This is consistent with several prior studies of adult OCD that reported below-normal *N*-acetyl metabolites levels or ratios in various cingulate subregions (reviewed in Saxena et al., 2009b). Ebert et al. (1997) found below-normal tNAA/Cr in right pACC, Jang et al. (2006) found below-normal tNAA/Cr in midline aMCC, and Yücel et al. (2007) found below-normal tNAA in left and right pACC. In the present study, Glx in pACC did not differ significantly between pre-CBT OCD patients and controls. This is in contrast to Zurowski et al. (2007) who found above-normal Glu in midline pACC in OCD, and to Yücel et al. (2008) who found below-normal Glx in left and right pACC and in left posterior middle cingulate cortex (pMCC, immediately caudal to aMCC) in female OCD patients only. In pediatric OCD, Rosenberg et al. (2004) similarly found below-normal Glx in midline pACC. Overall, our baseline tNAA findings add to evidence for diminished *N*-acetyl compounds in the cingulate in OCD. The lack of difference in Glx between groups may have been due to the fact that most patients in this study were on psychotropic medications, which have been found to alter Glx levels (Bolton et al., 2001; Moore et al., 1998; Rosenberg et al., 2000).

There are several possible reasons for the disagreements in results amongst these various MRS studies, including ours. First, some studies used unilateral (Ebert et al., 1997) or midline (left + right; Kitamura et al., 2006; Zurowski et al., 2007) MRS acquisition. The former can miss lateralized effects on the side not sampled. The latter may fail to detect lateralized effects because they are “diluted” across the simultaneously sampled left and right cingulate gyri or may detect such effects but be unable to assign them uniquely to one of the two gyri. Second, although, based on examination of figures, we have used Vogt's (2009) nomenclature to designate the cingulate subregions regions sampled in the prior studies, each prior study sampled the cingulate according to its own anatomic criteria. Hence, variability in selection and definition of cingulate subregions may have contributed to variability in MRS results. We suggest that adopting Vogt's (2009) or other standardized nomenclature and definitions for cingulate subregions might reduce this variability in future work. We have introduced a method to parcellate MRI of the human cingulate (O'Neill et al., 2009) that may facilitate such standardization. Moreover, Y-BOCS in some of the other studies averaged 15–20 (mild-to-moderate OCD) (Kitamura et al., 2006; Yücel et al., 2007, 2008) but ranged from 22 to 37 (moderate-to-extreme) in our patients. Thus, variation in severity of OCD may have contributed to differences in results between studies. Finally, differences in other sample characteristics, such as comorbid disorders, medications, and

subject handedness (not explicitly assessed in our study) may have contributed to discrepancies between the various MRS studies of OCD patients.

What could cause below-normal cingulate tNAA in OCD patients? One possibility is that abnormal levels may reflect aberrancies related to the astrocyte membrane-bound enzyme glutamate carboxypeptidase II (GCP-II; Cassidy and Neale, 1993). GCP-II decomposes NAAG into NAA and Glu in the extracellular fluid (Robinson et al., 1987). This is followed at the oligodendrocyte membrane by rapid hydrolysis of NAA by aspartoacylase (Baslow, 2000). Therefore, regional elevation of astrocyte membrane surface coverage, GCP-II surface density, or GCP-II specific reactivity in OCD, if appreciable, lead to reduced NAAG and thence tNAA levels, as seen in our data and elsewhere.

What could cause such neurophysiological lateralization as we found within the cingulate in OCD? Findings of lateralization within the cingulate have precedent, since high-resolution BOLD fMRI (Lütcke and Frahm, 2008) has recently identified lateralization within the pACC and aMCC of OCD-relevant functions such as response inhibition. A very speculative possibility could be differential “GABA-switching”, i.e., shift of GABA action from excitatory to inhibitory, between the two hemispheres due to neurodevelopment (Li and Xu, 2008) or to change in the predominant energetic substrate from glucose to lactate or pyruvate (Holmgren et al., 2010). Variation in GABAergic contribution to inhibition could change net inhibitory or excitatory output of a cingulate region thus determining normal functional lateralization or disrupting it in OCD. Abnormal GABA levels have also been recorded in pACC in OCD (Simpson et al., 2012).

4.3. Response of cingulate neurometabolite levels to CBT

In OCD patients treated with brief intensive CBT, tNAA in right pACC increased and Glx in left aMCC decreased significantly. Few MRS studies of OCD treatment response, particularly one examining the cingulate, exist for comparison. Zurowski et al. (2007) measured a sharp drop in Glu in left + right pACC after CBT. Jang et al. (2006) saw tNAA/Cr increase after citalopram treatment in both left + right aMCC, but could not report reliable Glu or Glx results due to MRSI acquisition at TE = 272 ms. Our results agree with those of both prior studies, although they are somewhat different in so far as we observed a pre- to post-CBT increase of tNAA in pACC (rather than aMCC) and a decrease in Glx in aMCC (rather than pACC). These discrepancies may again be attributable to the anatomic selection of cingulate subregions, medication effects, or greater severity of OCD in our patients. Nevertheless, the pre- to post-CBT drop in cingulate glutamatergic metabolites is notable for its frequency: 7/8 patients in our study and 14/17 in Zurowski et al. (2007).

How could CBT induce a drop in cingulate Glu or Glx? The pACC and aMCC exchange Glu with striatum and thalamus, and with other cortices. These structures form circuits that execute “behavioral macros” (complex sets of choreographed actions adapted to particular behavioral scenarios; Baxter et al., 2000). As OCD patients actively expose themselves to anxiogenic stimuli in the course of CBT, fears extinguish and associated pathological macros (e.g., OCD rituals) are weakened while adaptive macros (rational thoughts and habits) are strengthened, likely by modifying synaptic weightings and connectivity in the corresponding neural circuits. Thus, CBT may promote synaptic plasticity involved in formation of new habits or enhancements in volitional control, and these changes in plasticity may result in metabolite changes such as those observed. A role for Glu in these processes is supported by the known enhancement of CBT efficacy and of extinction learning by D-cycloserine (Rothbaum, 2008; Wilhelm et al., 2008; Cleva et al., 2010), a drug that acts at the glycine modulatory site (Emmett et al., 1991) of glutamatergic NMDA receptors. Similarly, physical exercise in rodents promotes glutamatergic changes in the brain (Biedermann et al., 2012).

4.4. Correlation of pre-CBT cingulate neurometabolite levels and CBT response

Within our OCD sample, higher values of pre-CBT tNAA in right pACC correlated with greater pre- to post-CBT drop in Y-BOCS score, i.e., greater improvement in OCD symptoms. In female OCD patients, Yücel et al. (2008) found that Glx in left and right pACC correlated positively with baseline Y-BOCS score, i.e., patients with higher Glx had worse symptoms. Although not part of our original study aims, this observation by Yücel et al. (2008) led us to test correlations of Glx with Y-BOCS within our female subjects only on an exploratory basis. We saw that in right aMCC for our female OCD patients only, there was a similar positive correlation between pre-CBT Glx and pre-CBT Y-BOCS ($r = 0.90$, $p = 0.037$, Spearman). Hence, our findings and the limited literature suggest that cingulate tNAA or Glx are associated with present severity of OCD symptoms or response to treatment. However, further research is needed to replicate these findings and clarify differences in subgroups before such data could be useful for predicting treatment response.

4.5. Limitations

Our sample sizes were small, although key results were reasonably uniform within the sample. Some of our OCD patients received psychopharmacologic treatment for OCD concurrent with CBT. Although the timescale of symptom response strongly suggests that the therapeutic effects observed were due to CBT, nevertheless, medication effects could have influenced pre-CBT neurometabolite levels in our sample. As is common in OCD, depressive symptoms were present in the patient sample, and these symptoms declined after CBT. Thus, regional baseline metabolite levels and their post-treatment changes may have reflected depression instead of or in addition to OCD. Depressive symptoms in all patients, however, were considered secondary to OCD. These symptoms generally responded to CBT, despite the fact that it targeted OCD explicitly and was not designed specifically for depression. Moreover, while OCD symptoms (Y-BOCS scores) correlated with cingulate tNAA levels, depressive symptoms (HamD scores) correlated with cingulate Cho and mI levels; thus, depressive and OCD symptoms and their relief may affect different aspects of neurometabolism. MRSI was acquired at 1.5 T; due to the limited spectral resolution, we thus opted not to attempt to segregate the Glu signal from Gln. Nonetheless, our major finding of pre- to post-CBT reduction of cingulate Glx was consistent with that of the high-field study of Zurowski et al. (2007), in which Glu was reported in isolation. Due to the limited scope of this pilot investigation, our healthy control group underwent MRSI scanning at one time-point only and only in pACC, not in aMCC. Although rescanning of a separate healthy control cohort in a neighboring cingulate subregion across a comparable timespan showed little variation in metabolite levels. Future studies should scan controls at two time-points, separated by a timespan equal to that between the pre- and post-CBT OCD scans, and in all cingulate subregions-of-interest. Bearing these limitations in mind, this ^1H MRSI study adds support to neurobiological models implicating pACC and aMCC in OCD (e.g., Saxena et al., 2009b).

Acknowledgments

Dr. Jeffrey M. Schwartz provided an initial suggestion to investigate two cingulate subregions. We thank Drs. George Bush and Brent A. Vogt for assistance in localizing the subregions on MRI. We are grateful to Dr. John C. Mazziotta for fostering this project in its germinal stage and to Drs. Fawzy Fawzy, Edythe D. London, and James T. McCracken for life support in a near terminal stage.

Role of the funding source: This work was supported by NIH grants R01 MH069433 (Dr. Saxena), R01 MH085900 (Drs. O'Neill and Feusner), R01 MH081864 (Drs. O'Neill and J.C. Piacentini), K08 AG22228 (Dr. Ringman), and by donations to the Westwood Institute for Anxiety Disorders (Dr. Gorbis). For generous support of OCD and neuroimaging research at UCLA, the authors wish to thank the Brain Mapping Medical Research Organization, Brain Mapping Support Foundation, Pierson-Lovelace Foundation, The Ahmanson Foundation, William M. and Linda R. Dietel Philanthropic Fund at the Northern Piedmont Community Foundation, Tamkin

Foundation, Jennifer Jones-Simon Foundation, Capital Group Companies Charitable Foundation, Robson Family, Northstar Fund, and the National Center for Resources grants RR12169, RR13642 and RR08655.

References

- Arnold PD, MacMaster FP, Richter MA, Hanna GL, Sicard T, Burroughs E, et al. Glutamate receptor gene (GRIN2B) associated with reduced anterior cingulate glutamatergic concentration in obsessive-compulsive disorder. *Psychiatry Research*. 2009; 172(2):136–9. [PubMed: 19324536]
- Bartha R, Stein MB, Williamson PC, Drost DJ, Neufeld RWJ, Carr TJ, et al. A short echo 1H spectroscopy and volumetric MRI study of the corpus striatum in patients with obsessive-compulsive disorder and comparison subjects. *American Journal of Psychiatry*. 1998; 155:1584–91. [PubMed: 9812122]
- Baslow MH. Functions of N-acetyl-L-aspartate and N-acetyl-L-aspartylglutamate in the vertebrate brain. Role in glial cell-specific signaling. *Journal of Neurochemistry*. 2000; 75:453–9. [PubMed: 10899919]
- Baxter, LR.; Ackermann, RF.; Swerdlow, NR.; Brody, A.; Saxena, S.; Schwartz, JM., et al. Specific brain system mediation of OCD responsive to either medication or behavior therapy. In: Goodman, WK.; Rudorfer, MV., editors. *Obsessive-compulsive disorder: Contemporary issues in treatment*. Mahwah, New Jersey: Lawrence Erlbaum Associates, Inc.; 2000. p. 573–609.
- Baxter LR Jr, Phelps ME, Mazziotta JC, Guze BH, Schwartz JM, Selin CE. Local cerebral glucose metabolic rates in obsessive-compulsive disorder – a comparison with rates in unipolar depression and in normal controls. *Archives of General Psychiatry*. 1987; 44:211–8. [PubMed: 3493749]
- Baxter LR, Schwartz JM, Mazziotta JC, Phelps ME, Pahl JJ, Guze BH, et al. Cerebral glucose metabolic rates in non-depressed obsessive-compulsives. *American Journal of Psychiatry*. 1988; 145:1560–3. [PubMed: 3264118]
- Baxter LR Jr, Schwartz JM, Bergman KS, Szuba MP, Guze BH, Mazziotta JC, et al. Caudate glucose metabolic rate changes with both drug and behavior therapy for obsessive-compulsive disorder. *Archives of General Psychiatry*. 1992; 49:681–9. [PubMed: 1514872]
- Benazon NR, Moore GJ, Rosenberg DR. Neurochemical analyses in pediatric obsessive-compulsive disorder in patients treated with cognitive-behavioral therapy. *Journal of the American Academy of Child and Adolescent Psychiatry*. 2003; 42(11):1279–85. [PubMed: 14566164]
- Biedermann S, Fuss J, Zheng L, Sartorius A, Falfán-Melgoza C, Demirkaya T, et al. In vivo voxel based morphometry: detection of increased hippocampal volume and decreased glutamate levels in exercising mice. *NeuroImage*. 2012; 61(4):1206–12. [PubMed: 22521257]
- Bolton J, Moore GJ, MacMillan S, Stewart CM, Rosenberg DR. Case study: caudate glutamatergic changes with paroxetine persist after medication discontinuation in pediatric OCD. *Journal of the American Academy of Child and Adolescent Psychiatry*. 2001; 40:903–6. [PubMed: 11501689]
- Brennan, BP.; Rauch, SL.; Jensen, JE.; Pope, HG, Jr. A critical review of magnetic resonance spectroscopy studies of obsessive-compulsive disorder. *Biological Psychiatry*. 2012. <http://dx.doi.org/10.1016/j.biopsych.2012.06.023>
- Bush G, Luu P, Posner MI. Cognitive and emotional influences in anterior cingulate cortex. *Trends Cognitive Science*. 2000; 4:215–22.
- Cangro CB, Namboodiri MAA, Sklar LA, Corigliano-Murphy A, Neale JH. Immunohistochemistry and biosynthesis of N-acetylaspartylglutamate in spinal sensory ganglia. *Journal of Neurochemistry*. 1987; 49:1579–88. [PubMed: 2889802]
- Cassidy M, Neale JH. N-acetylaspartylglutamate catabolism is achieved by an enzyme on the cell surface of neurons and glia. *Neuropeptides*. 1993; 24:271–8. [PubMed: 8100987]
- Cleva RM, Gass JT, Widholm JJ, Olive MF. Glutamatergic targets for enhancing extinction learning in drug addiction. *Current Neuropharmacology*. 2010; 8(4):394–408. [PubMed: 21629446]
- Danbolt NC. Glutamate uptake. *Progress in Neurobiology*. 2001; 65:1–105. [PubMed: 11369436]
- Devinsky O, Morrell MJ, Vogt BA. Contributions of anterior cingulate cortex to behavior. *Brain*. 1995; 118:279–306. [PubMed: 7895011]

- Ebert D, Speck O, König A, Berger M, Hennig J, Hohagen F. ^1H -magnetic resonance spectroscopy in obsessive-compulsive disorder: evidence for neuronal loss in the cingulate gyrus and the right striatum. *Psychiatry Research: Neuroimaging*. 1997; 74:173–6.
- Emmett M, Mick S, Cler J. Actions of d-cycloserine at the *N*-methyl-d-aspartate-associated glycine receptor site in vivo. *Neuropharmacology*. 1991; 30:1167–71. [PubMed: 1663594]
- First, MB.; Spitzer, RL.; Gibbon, M.; Williams, JBW. Structured clinical interview for DSM-IV patient edition (SCID-I/P). New York: New York State Psychiatric Institute, Biometrics Research Department; 1996.
- Fitzgerald KD, Moore GJ, Paulson LA, Stewart CM, Rosenberg DR. Proton spectroscopic imaging of the thalamus in treatment-naïve pediatric obsessive-compulsive disorder. *Biological Psychiatry*. 2000; 47:174–82. [PubMed: 10682215]
- Foa EB, Liebowitz MR, Kozak MJ, Davies S, Campeas R, Franklin ME, et al. Randomized, placebo-controlled trial of exposure and ritual prevention, clomipramine, and their combination in the treatment of obsessive-compulsive disorder. *American Journal of Psychiatry*. 2005; 162:151–61. [PubMed: 15625214]
- Freyer T, Klöppel S, Tüscher O, Kordon A, Zurowski B, Kuelz A-K, et al. Frontostriatal activation in patients with obsessive-compulsive disorder before and after cognitive behavioral therapy. *Psychological Medicine*. 2011; 41:207–16. [PubMed: 20236568]
- Goodman WK, Price LH, Rasmussen SA, Mazure C, Delgado P, Heninger GR, et al. The Yale-Brown obsessive compulsive scale I: development, use, and reliability. *Archives of General Psychiatry*. 1989; 46:1006–11. [PubMed: 2684084]
- Guy, W. ECDEU assessment manual for psychopharmacology Rev (DHEW publication no (ADM) 76-338). Rockville, MD: NIMH; p. 1976
- Hamilton M. Diagnosis and rating scale for depression. *British Journal of Psychiatry*. 1960; 3:76–9.
- Hertz L, Zielke HR. Astrocytic control of glutamatergic activity: astrocytes as stars of the show. *Trends in Neurosciences*. 2004; 27(12):735–43. [PubMed: 15541514]
- Holmgren CD, Mukhtarov M, Malkov AE, Popova IY, Bregestovski P, Zilberter Y. Energy substrate availability as a determinant of neuronal resting potential, GABA signaling and spontaneous network activity in the neonatal cortex *in vitro*. *Journal of Neurochemistry*. 2010; 112:900–12. [PubMed: 19943846]
- Jang JH, Kwon JS, Jang DP, Moon WJ, Lee JM, Ha TH, et al. A proton MRSI study of brain N-acetylaspartate level after 12 weeks of citalopram treatment in drug-naïve patients with obsessive compulsive disorder. *American Journal of Psychiatry*. 2006; 163:1202–7. [PubMed: 16816225]
- Kitamura H, Shioiri T, Kimura T, Ohkubo M, Nakada T, Someya T. Parietal white matter abnormalities in obsessive-compulsive disorder: a magnetic resonance spectroscopy study at 3-Tesla. *Acta Psychiatrica Scandinavica*. 2006; 114:101–8. [PubMed: 16836597]
- Kozak, MJ.; Foa, EB. Mastery of obsessive-compulsive disorder: a cognitive-behavioral approach—therapist guide (treatments that work). New York: Graywind Publications; 1997.
- Kwon JS, Kim JJ, Lee DW, Lee DS, Kim MS, Lyoo IK, et al. Neural correlates of clinical symptoms and cognitive dysfunctions in obsessive-compulsive disorder. *Psychiatry Research: Neuroimaging*. 2003; 122:37–47.
- Lütcke H, Frahm J. Lateralized anterior cingulate function during error processing and conflict monitoring as revealed by high-resolution fMRI. *Cerebral Cortex*. 2008; 18:508–15. [PubMed: 17576750]
- Li K, Xu E. The role and the mechanism of γ -aminobutyric acid during central nervous system development. *Neuroscience Bulletin*. 2008; 24(3):195, 200. [PubMed: 18500393]
- MacMaster FP, O'Neill J, Rosenberg DR. Brain imaging in pediatric obsessive compulsive disorder. *Journal of the American Academy of Child and Adolescent Psychiatry*. 2008; 47(11):1262–72. [PubMed: 18827717]
- Mirza Y, O'Neill J, Smith EA, Russell A, Smith J, Banerjee SP, et al. Increased medial thalamic creatine/phosphocreatine found by proton magnetic resonance spectroscopy in children with obsessive-compulsive disorder versus major depression and healthy controls. *Journal of Child Neurology*. 2006; 21(2):106–11. [PubMed: 16566872]

- Mohamed MA, Smith MA, Schlund MW, Nestadt G, Barker PB, Hoehn-Saric R. Proton magnetic resonance spectroscopy in obsessive-compulsive disorder: a pilot investigation comparing treatment responders and non-responders. *Psychiatry Research: Neuroimaging*. 2007; 156:175–9.
- Moore GJ, MacMaster FP, Stewart C, Rosenberg DR. Case study: caudate glutamatergic changes with paroxetine therapy for pediatric obsessive-compulsive disorder. *Journal of the American Academy of Child and Adolescent Psychiatry*. 1998; 37:663–7. [PubMed: 9628087]
- Moreno A, Ross BD, Bluml S. Direct determination of the N-acetyl-L-aspartate synthesis rate in the human brain by (13)C MRS and [1-(13)C]glucose infusion. *Journal of Neurochemistry*. 2001; 77(1):347–50. [PubMed: 11279290]
- Nabeyama M, Nakagawa A, Yoshiura T, Nakao T, Nakatani E, Togao O, et al. Functional MRI study of brain activation alterations in patients with obsessive-compulsive disorder after symptom improvement. *Psychiatry Research: Neuroimaging*. 2008; 163:236–47.
- Nakatani E, Nakagawa A, Ohara Y, Goto S, Uozumi N, Iwakiri M, et al. Effects of behavior therapy on regional cerebral blood flow in obsessive-compulsive disorder. *Psychiatry Research: Neuroimaging*. 2003; 124:113–20.
- Nordahl TE, Benkelfat C, Semple WE. Cerebral glucose metabolic rates in obsessive-compulsive disorder. *Neuropsychopharmacology*. 1989; 2(1):23–8. [PubMed: 2803479]
- O'Neill J, Eberling JL, Schuff N, Jagust WJ, Reed B, Soto G, et al. Correlation of ¹H MRSI and ¹⁸F-DG-PET. *Magnetic Resonance in Medicine*. 2000; 43:244–50. [PubMed: 10680688]
- O'Neill J, Schwartz JM. OCD treatment: the role of volition in OCD therapy: neurocognitive, neuroplastic, and neuroimaging aspects. *Clinical Neuropsychiatry*. 2005; 1(1):10–8.
- O'Neill, J.; Sobel, TL.; Vogt, BA. Localizing cingulate subregions of interest in magnetic resonance images guided by cytological parcellations. In: Vogt, BA., editor. *Cingulate neurobiology and disease*. New York: Oxford University Press; 2009. p. 3-30.
- O'Neill, J.; Thomas, S.; Hudkins, M.; Dean, A.; Tobias, MC.; London, ED. MRSI and a model of metabolic dysfunction in human methamphetamine dependence. *Translational research in methamphetamine addiction conference*; Pray, MT. July 21, 2010;
- Pallanti S, Hollander E, Bienstock C, Koran L, Leckman J, Marazziti D, et al. Treatment non-response in OCD: methodological issues and operational definitions. *International Journal of Neuropsychopharmacology*. 2002; 5:181–91. [PubMed: 12135542]
- Perani D, Colombo C, Bressi S, Bonfanti A, Grassi F, Scarone S, et al. [¹⁸F]-FDG-PET study in obsessive-compulsive disorder: a clinical/metabolic correlation study after treatment. *British Journal of Psychiatry*. 1995; 166:244–50. [PubMed: 7728370]
- Petroff OA, Mattson RH, Rothman DL. Proton MRS: GABA and glutamate. *Advances in Neurology*. 2000; 83:261–71. [PubMed: 10999208]
- Pfund Z, Chugani DC, Juhasz C, Muzik O, Chugani HT, Wilds IB, et al. Evidence for coupling between glucose metabolism and glutamate cycling using FDG PET and ¹H magnetic resonance spectroscopy in patients with epilepsy. *Journal of Cerebral Blood Flow and Metabolism*. 2000; 20(5):871–8. [PubMed: 10826538]
- Picard N, Strick PL. Motor areas of the medial wall: a review of their location and functional activation. *Cerebral Cortex*. 1996; 6:342–53. [PubMed: 8670662]
- Provencher SW. Automatic quantitation of localized in vivo ¹H spectra with LCModel. *NMR in Biomedicine*. 2001; 14:260–4. [PubMed: 11410943]
- Ringman, J.; O'Neill, J.; Tseng, PB.; Apostolova, L.; Geschwind, D.; Schaffer, B., et al. Elevated hippocampal myo-inositol in subjects with or at-risk for familial AD. *ICAD 2006; 10th international conference on Alzheimer's disease and related disorders*; Madrid. July; 2006. p. 15-20.
- Robinson MB, Blakely RD, Couto R, Coyle JT. Hydrolysis of the brain dipeptide N-acetyl-L-aspartyl-L-glutamate Identification and characterization of a novel N-acetylated alpha-linked acidic dipeptidase activity from rat brain. *Journal of Biological Chemistry*. 1987; 262:14498–506. [PubMed: 3667587]
- Rojas C, Frazier ST, Flanary J, Slusher BS. Kinetics and inhibition of glutamate carboxypeptidase II using a microplate assay. *Analytical Biochemistry*. 2002; 310:50–4. [PubMed: 12413472]

- Rosenberg DR, Amponsah A, Sullivan A, Macmillan S, Moore GJ. Increased medial thalamic choline in pediatric obsessive-compulsive disorder as detected by quantitative in vivo spectroscopic imaging. *Journal of Child Neurology*. 2001; 16:636–41. [PubMed: 11575601]
- Rosenberg DR, Keshavan MS. Toward a neurodevelopmental model of obsessive-compulsive disorder. *Biological Psychiatry*. 1998; 43:623–40. [PubMed: 9582996]
- Rosenberg DR, MacMaster FP, Keshavan MS, Fitzgerald KD, Stewart CM, Moore GJ. Decrease in caudate glutamatergic concentrations in pediatric obsessive-compulsive disorder patients taking paroxetine. *Journal of the American Academy of Child and Adolescent Psychiatry*. 2000; 39(9): 1096–103. [PubMed: 10986805]
- Rosenberg DR, Mirza Y, Russell A, Tang J, Smith JM, Banerjee P, et al. Reduced anterior cingulate glutamatergic concentrations in childhood OCD and major depression versus healthy controls. *Journal of the American Academy of Child and Adolescent Psychiatry*. 2004; 43(9):1146–53. [PubMed: 15322418]
- Rothbaum BO. Critical parameters for d-cycloserine enhancement of cognitive-behavioral therapy for obsessive-compulsive disorder. *American Journal of Psychiatry*. 2008; 165(3):293–6. [PubMed: 18316423]
- Sawle GV, Hymas NF, Lees AJ, Frackowiak RS. Obsessional slowness: functional studies with positron emission tomography. *Brain*. 1991; 114(Pt 5):2191–202. [PubMed: 1933241]
- Saxena S, Bota RG, Brody AL. Brain-behavior relationships in obsessive-compulsive disorder. *Seminars in Clinical Neuropsychiatry*. 2001; 6:82–101. [PubMed: 11296309]
- Saxena S, Gorbis E, O'Neill J, Baker SK, Mandelkern MA, Maidment KM, et al. Rapid effects of brief intensive cognitive-behavioral therapy on brain glucose metabolism in obsessive-compulsive disorder. *Molecular Psychiatry*. 2009a; 14(2):197–205. [PubMed: 18180761]
- Saxena, S.; O'Neill, J.; Rauch, SL. The role of cingulate cortex dysfunction in obsessive-compulsive disorder. In: Vogt, BA., editor. *Cingulate neurobiology and disease*. New York: Oxford University Press; 2009b. p. 587–618.
- Schwartz, JM., editor. *Brain lock: free yourself from obsessive-compulsive disorder*. New York: Harper Collins; 1996.
- Schwartz JM, Stoessel PW, Baxter LR Jr, Martin KM, Phelps ME. Systematic changes in cerebral glucose metabolic rate after successful behavior modification treatment of obsessive-compulsive disorder. *Archives of General Psychiatry*. 1996; 53:109–13. [PubMed: 8629886]
- Seese RR, O'Neill J, Hudkins M, Siddarth P, Levitt J, Tseng B, et al. Proton magnetic resonance spectroscopy and thought disorder in childhood schizophrenia. *Schizophrenia Research*. 2011; 133:82–90. [PubMed: 21872444]
- Shattuck D, Sandor-Leahy S, Schaper K, Rottenberg DA, Leahy RM. Magnetic resonance image tissue classification using a partial volume model. *NeuroImage*. 2001; 13:856–76. [PubMed: 11304082]
- Sibson NR, Shen J, Mason GF, Rothman DL, Behar KL, Sulman RG. Functional energy metabolism: in vivo ¹³C-NMR spectroscopy evidence for coupling of cerebral glucose consumption and glutamatergic neuronal activity. *Developmental Neuroscience*. 1998; 20(4–5):321–30. [PubMed: 9778568]
- Simpson, HB.; Shungu, DC.; Bender, J., Jr; Mao, X.; Xu, X.; Slifstein, M., et al. Investigation of cortical glutamate-glutamine and -aminobutyric acid in obsessive-compulsive disorder by proton magnetic resonance spectroscopy. *Neuropsychopharmacology*. 2012. <http://dx.doi.org/10.1038/npp.2012.132> [Epub ahead of print]
- Smith EA, Russell A, Lorch E, Banerjee SP, Rose M, Ivey J, et al. Increased medial thalamic choline found in pediatric patients with obsessive-compulsive disorder versus major depression or healthy control subjects: a magnetic resonance spectroscopy study. *Biological Psychiatry*. 2003; 54:1399–405. [PubMed: 14675804]
- Starck G, Ljungberg M, Nilsson M, Jönsson L, Lundberg S, Ivarsson T, et al. A ¹H magnetic resonance spectroscopy study in adults with obsessive compulsive disorder: relationship between metabolite concentrations and symptom severity. *Journal of Neural Transmission*. 2008; 115:1051–62. [PubMed: 18528631]

- Sumitani S, Harada M, Kubo H, Ohmori T. Proton magnetic resonance spectroscopy reveals an abnormality in the anterior cingulate of a subgroup of obsessive-compulsive disorder patients. *Psychiatry Research: Neuroimaging*. 2007; 154:85–92.
- Swedo SE, Schapiro MB, Grady CL, Cheslow DL, Leonard HL, Kumar A, et al. Cerebral glucose metabolism in childhood onset obsessive-compulsive disorder. *Archives of General Psychiatry*. 1989; 46:518–23. [PubMed: 2786402]
- Vogt, BA. Regions and subregions of the cingulate gyrus. In: Vogt, BA., editor. *Cingulate neurobiology and disease*. New York: Oxford University Press; 2009. p. 3-30.
- Whalen PJ, Bush G, McNally RJ, Wilhelm S, McInerney SC, Jenike MA, et al. The emotional counting Stroop paradigm: an fMRI probe of the anterior cingulate affective division. *Biological Psychiatry*. 1998; 44:1219–28. [PubMed: 9861465]
- Whiteside SP, Abramowitz JS, Port JD. The effect of behavior therapy on caudate N-acetyl-l-aspartic acid in adults with obsessive-compulsive disorder. *Psychiatry Research: Neuroimaging*. 2012; 201:10–6.
- Whiteside SP, Port JD, Deacon BJ, Abramowitz JS. A magnetic resonance spectroscopy investigation of obsessive-compulsive disorder and anxiety. *Psychiatry Research: Neuroimaging*. 2006; 146:137–47.
- Wilhelm S, Buhlmann U, Tolin DF, Meunier SA, Pearson G, Reese HE, et al. Augmentation of behavior therapy with d-cycloserine for obsessive-compulsive disorder. *American Journal of Psychiatry*. 2008; 165:335–41. [PubMed: 18245177]
- Yamanishi T, Nakaaki S, Omori IM, Hashimoto N, Shinagawa Y, Hongo J, et al. Changes after behavior therapy among responsive and nonresponsive patients with obsessive-compulsive disorder. *Psychiatry Research: Neuroimaging*. 2009; 172:242–50.
- Yücel M, Harrison BJ, Wood SJ, Fornito A, Wellard RM, Pujo J, et al. Functional and biochemical alterations of the medial frontal cortex in obsessive-compulsive disorder. *Archives of General Psychiatry*. 2007; 64(8):946–55. [PubMed: 17679639]
- Yücel M, Wood SJ, Wellard RM, Harrison BJ, Fornito A, Pujol J, et al. Anterior cingulate glutamate-glutamine levels predict symptom severity in women with obsessive-compulsive disorder. *Australia-new Zealand Journal of Psychiatry*. 2008; 42(6):467–77.
- Zurowski, B.; Kordon, A.; Weber-Fahr, W.; Voderholzer, U.; Kuelz, AK.; Freyer, T., et al. Relevance of orbitofrontal neurochemistry for the outcome of cognitive-behavioural therapy in patients with obsessive-compulsive disorder. *European Archives of Psychiatry and Clinical Neuroscience*. 2012. <http://dx.doi.org/10.1007/s00406-012-0304-0>
- Zurowski B, Weber-Fahr W, Freyer T, Wahli K, Büchert M, Kuel AK, et al. Neurochemical abnormalities in patients with obsessive-compulsive disorder diminish in the course of behavior therapy. *Society for Neuroscience Abstracts*. Nov 5.2007

Fig. 1. Position of pACC (upper) and aMCC (lower) MRSI slabs (yellow boxes) and sample spectra (left, from blue boxes). The PRESS (TR/TE = 1500/30 ms) volume (white box) is sized and positioned to sample target structures. Sample spectra are shown from left pACC and left aMCC. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

Fig. 2.

(Left) Yale–Brown Obsessive–Compulsive Scale (Y-BOCS) score before and after brief intensive cognitive-behavioral therapy (CBT) in 10 patients with obsessive–compulsive disorder (OCD) showing sharp drop in OCD symptoms post-treatment in 9/10 patients. (Patient 9 has no post-CBT datum.) Group-mean Y-BOCS (black horizontal bars) fell 60.2%. (Right) levels of glutamate(Glu) + glutamine(Gln), together abbreviated “Glx”, in left anterior middle cingulate cortex (aMCC) in the same patients, showing a post-CBT drop in 7/8 patients. (Mean drop 22.0%; patients 9–10 have no post-CBT data.) Glx levels are corrected for voxel cerebrospinal fluid content and expressed in institutional units (IU). $^{\dagger} p < 0.01$, $^{\dagger\dagger} p < 0.0005$ paired *T*-test of rank-transformed (non-parametric) data, following repeated-measures multivariate analysis of variance for Glx. Findings suggest OCD symptom improvement in response to CBT is associated with lateralized post-treatment reduction in glutamatergic metabolites in the aMCC.

Fig. 3.

(Left) CSF-corrected levels of *N*-acetyl-aspartate(NAA) + *N*-acetyl-aspartyl-glutamate (NAAG), together abbreviated “tNAA”, in right pregenual anterior cingulate cortex (pACC) in 10 patients with obsessive–compulsive disorder (OCD) before and after brief intensive cognitive-behavioral therapy (CBT) and in 8 age-matched healthy controls (one scan only). Group-mean tNAA was 13.8% lower in pre-CBT OCD patients than in controls, but then rose 10.2% after CBT (black horizontal bars; patients 9–10 have no post-CBT data). (Right) pre- to post-CBT change in OCD symptoms, expressed by Yale–Brown Obsessive–Compulsive Scale (Y-BOCS) score, as a function of pre-CBT right pACC tNAA for the 8 patients who completed pre- and post-CBT assessments. A strong negative correlation ($r = -0.86$) was observed. tNAA levels are corrected for voxel cerebrospinal fluid content and expressed in institutional units (IU). $^{**}p < 0.01$ Spearman; $^{***}p < 0.001$ independent *T*-test of rank-transformed data, following repeated-measures multivariate analysis of covariance; $^{\dagger}p < 0.05$ paired *T*-test of rank-transformed data, following repeated-measures multivariate analysis of variance. Findings suggest levels of *N*-acetyl compounds are depressed in the right-sided pACC in symptomatic OCD, that these levels rise after successful treatment, and that the pre-CBT levels predict subsequent CBT clinical response.

Fig. 4.

(Left) Hamilton Depression Scale (HamD) score before and after CBT in 10 patients with OCD showing decreases in depressive symptoms post-treatment in 8/10 patients. (Patient 2 scored “0” pre- and post-CBT, patient 9 has no post-CBT datum.) Group-mean HamD decreased by 60.4%. (Right) pre-CBT HamD score as a function of pre-CBT left pACC myo-inositol (mI) for the same patients. A positive correlation ($R = +0.70$) was observed. mI levels are corrected for voxel cerebrospinal fluid content and expressed in institutional units (IU). $^*p < 0.05$ Spearman; $^{\dagger\dagger}p < 0.01$ paired T-test of rank-transformed data. These results suggest comorbid depressive symptoms as possible contributor to metabolite findings of Figs. 2 and 3. Note, however, that depressive symptoms in all patients were considered secondary to OCD and that CBT targeted OCD explicitly, but did not treat specifically for depression. Also, while OCD symptoms appear to be sensitive to pACC levels of *N*-acetyl compounds, depressive symptoms appear to vary with pACC mI.

Fig. 5.

CSF-corrected levels of tNAA (left) and Glx (right) in midline (left + right) dorsal posterior cingulate cortex (dPCC) in an auxiliary cohort of 5 healthy controls (2 male, 3 female; mean age 36.0 ± 8.7 years, range 28–46 years) for two identical scans separated by 23–37 days. Scan-to-scan variability of metabolite levels was modest (mean 1.7% tNAA –1.8% Glx) and random. Abbreviations and other details as in Figs. 2 and 3.

Table 1

Clinical characteristics of obsessive–compulsive disorder patients.

Patient	Sex	Age, yr	Medication	Current and prior comorbidities
1	Female	34.3	Fluvoxamine, clonazepam	Dysthymic disorder, current and prior major depression
2	Female	29.6	Paroxetine, risperidone, clonazepam	Prior major depression
3	Female	27.9	None	Specific phobia – spiders
4	Male	51.7	None	Prior alcohol, marijuana abuse
5	Male	46.0	Escitalopram, bupropion	Current and prior major depression
7	Male	34.9	None	None
8	Male	30.8	Sertraline, methylphenidate	Prior ADHD, current major depression
9	Female	25.7	None	Prior major depression
10	Male	34.0	None	Current major depression

All but patients 9–10 received both pre- and post-CBT MRSI scans. For current depression scores of patients see Fig. 4.

MRSI voxel tissue composition and neurometabolites in cingulate subregions in obsessive-compulsive disorder patients before and after brief intensive cognitive-behavioral therapy (CBT) and in age-matched healthy controls. Statistically significant comparisons in boldface.

Table 2

	Left hemisphere		Right hemisphere	
	Control	Pre-CBT	Post-CBT	Post-CBT
<i>Pregenual anterior cingulate cortex (pACC)</i>				
vol% GM	72.7 (4.0)	74.1 (3.2)	72.7 (4.4)	72.9 (5.8)
vol% WM	11.4 (5.3)	7.0 (4.1)	7.6 (4.0)	12.3 (5.8)
tNAA	8.4 (1.5)	8.1 (0.7)	8.6 (1.0)	7.0 (0.4) *** 7.8 (0.8) [†]
Glx	14.1 (2.3)	14.7 (2.5)	16.5 (2.1)	13.3 (1.5)
Cr	5.2 (1.2)	5.5 (0.9)	6.0 (0.8)	4.9 (0.5)
Cho	1.8 (0.4)	1.9 (0.4)	2.0 (0.2)	1.6 (0.3)
mI	4.7 (1.0)	4.8 (1.0)	5.5 (1.4)	4.3 (1.1)
<i>Anterior middle cingulate cortex (aMCC)^a</i>				
vol% GM	72.6 (5.0)	74.2 (5.5)	74.9 (6.9)	71.8 (6.1)
vol% WM	7.1 (4.4)	12.2 (9.2)	15.4 (6.3)	18.2 (10.5)
tNAA	8.5 (1.0)	7.5 (1.4)	7.9 (0.8)	8.1 (1.6)
Glx	14.6 (2.2)	11.9 (2.5) ^{††}	13.3 (2.0)	12.6 (3.1)
Cr	6.1 (0.9)	5.3 (1.3)	5.6 (1.0)	5.6 (1.2)
Cho	2.0 (0.3)	1.8 (0.5)	1.8 (0.3)	1.9 (0.4)
mI	5.3 (1.2)	5.1 (1.2)	4.9 (1.1)	5.1 (0.8)

MRSI = magnetic resonance spectroscopic imaging, vol% = percent of MRSI voxel volume, GM = gray matter, WM = white matter, tNAA = *N*-acetyl-aspartate + *N*-acetyl-aspartyl-glutamate, Glx = glutamate + glutamine, Cr = creatine + phosphocreatine, Cho = choline-containing compounds, mI = *myo*-inositol. Values are group means (std.) neurometabolite levels corrected for voxel CSF content and expressed in institutional units (IU).

^aMRSI not acquired for controls in aMCC.

[†] $p < 0.05$.

^{††} $p < 0.01$ paired *T*-test of rank-transformed (non-parametric) data, following repeated-measures multivariate analysis of variance.

*** $p < 0.001$ independent *T*-test of rank-transformed data, following omnibus multivariate analysis of covariance. Note that vol% GM and WM refer to proportions of tissue inside the MRSI voxels sampled and not to the total anatomic gray and white matter volumes of the pACC and aMCC.