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Proton Magnetic Resonance Spectroscopy and Thought Disorder in Childhood Schizophrenia

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Abstract

Objective—Although magnetic resonance spectroscopy has identified metabolic abnormalities in adult and childhood schizophrenia, no prior studies have investigated the relationship between neurometabolites and thought disorder. This study examined this association in language-related brain regions using proton magnetic resonance spectroscopic imaging (¹H MRSI).

Method—MRSI was acquired bilaterally from 28 youth with childhood-onset schizophrenia and 34 healthy control subjects in inferior frontal, middle frontal, and superior temporal gyri at 1.5 T and short echo time (TR/TE=1500/30 ms). CSF-corrected “total NAA” (tNAA; *N*-acetyl-aspartate + *N*-acetyl-aspartyl-glutamate), glutamate+glutamine (Glx), creatine+phosphocreatine (Cr+PCr), choline compounds (Cho), and *myo*-inositol (mI) were assayed in manually drawn regions-of-interest partitioned into gray matter, white matter, and CSF and then coregistered with MRSI. Speech samples of all subjects were coded for thought disorder.

Results—In the schizophrenia group, the severity of formal thought disorder correlated significantly with tNAA in the left inferior frontal and superior temporal gyri and with Cr+PCr in left superior temporal gyrus.

Conclusions—Neurometabolite concentrations in language-related brain regions are associated with thought disorder in childhood-onset schizophrenia.

Keywords

Childhood-onset schizophrenia; Thought Disorder; Magnetic Resonance Spectroscopy; *N*-acetyl aspartate; Choline compounds

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Contributors

R.R.S., J.O.N., and R.C. designed the experiments, analyzed the data, and prepared the manuscript. J.O.N. and P.S. performed statistical analyses. R.R.S., M.H., J.L., B.T., K.N.W. were involved with data acquisition and processing. All authors have contributed to and have approved the final manuscript.

Conflict of Interest

None of the authors declare any conflicts of interest with this study.

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1. INTRODUCTION

Proton magnetic resonance spectroscopy (^1H MRS) is a non-invasive, *in vivo* neuroimaging technique that interrogates specific aspects of neurochemistry. As such, MRS offers possibilities to better understand underlying abnormalities and to develop therapeutics for brain-related illnesses like schizophrenia. Most MRS studies on schizophrenia (reviewed in (Steen et al., 2005)) have focused on the largest ^1H MRS peak thought to indicate neuronal integrity (Demougeot et al., 2001; Urenjak et al., 1993). About three-quarters of this resonance's intensity is generated by the amino acid *N*-acetyl-aspartate (NAA); the remaining quarter is due to its derivative *N*-acetyl-aspartyl-glutamate (NAAG) (Edden et al., 2007; Pouwels and Frahm, 1997). In this paper, we refer to the combined peak "total NAA" (tNAA). Other metabolites of interest in ^1H MRS include glutamate plus glutamine (Glx), creatine plus phosphocreatine (Cr+PCr), choline compounds (Cho), and *myo*-inositol (mI).

Several MRS studies have found that absolute tNAA levels and the ratios, tNAA/Cr+PCr and tNAA/Cho, are below-normal in the hippocampus (Bertolino et al., 1996; Deicken et al., 1998; Deicken et al., 1999; Maier et al., 1995), anterior cingulate cortex (Jessen et al., 2006; Yamasue et al., 2002; Yasukawa et al., 2005), and dorsolateral prefrontal cortex (Bertolino et al., 1996; Callicott et al., 2000; Molina et al., 2007; Zabala et al., 2007) of adult schizophrenia patients. Similar to these findings in adults, MRS studies in childhood schizophrenia demonstrated lower tNAA/Cr+PCr in the hippocampus and dorsolateral prefrontal cortex bilaterally compared to healthy control children (Bertolino et al., 1998) and significantly lower tNAA in left dorsolateral prefrontal cortex compared both to children experiencing first episodes of psychosis and to healthy controls (Zabala et al., 2007). Another study illustrated above-normal Cr+PCr and Cho in the anterior middle cingulate cortex, non-cingulate frontal cortex, and caudate head (O'Neill et al., 2004). Above-normal Glx was also found in the right medial frontal lobe in children at high genetic risk for schizophrenia (Tibbo et al., 2004).

Formal thought disorder, a symptom of childhood schizophrenia, includes illogical reasoning and loosening of associations (Caplan et al., 2000). Prior volumetric, functional magnetic resonance imaging (fMRI), and H_2^{15}O -PET studies demonstrated involvement of the superior temporal (Holinger et al., 1999; Kircher et al., 2003; McGuire et al., 1998; Shenton et al., 1992), middle frontal (Kircher et al., 2003), and inferior frontal gyri (Assaf et al., 2006; Cerullo et al., 2007; McGuire et al., 1998) in the thought disorder of adults with schizophrenia. A recent fMRI study showed that the severity of thought disorder in childhood schizophrenia was associated with reduced cortical activity in the left inferior frontal gyrus and superior temporal gyrus during semantic and syntactic tasks, in the dorsolateral prefrontal cortex and dorsal medial prefrontal cortex during the semantic task, and in the insula during the syntactic task (Borofsky et al., in press). Combined, these studies suggest that cortical regions that process language-related information may play a role in the thought disorder of schizophrenia.

Thus, the exploratory proton magnetic resonance spectroscopy investigation described here not only interrogated possible metabolite differences in patients compared to healthy controls, but also examined the relationship between metabolite concentrations and severity of thought disorder in childhood onset schizophrenia. We employed the magnetic resonance spectroscopic imaging (MRSI) variety of MRS to simultaneously examine multiple small (~1 cc) volume elements ("voxels") each containing a high percentage of the targeted region-of-interest (ROI) (Bertolino et al., 1999; Maudsley, 2002). Moreover, we acquired MRSI at short-TE (30 ms), which allows for superior detection of Glx and mI and also yields larger signal intensities for tNAA, Cr+PCr, and Cho. We anticipated significantly different regional metabolite levels in the superior temporal (Holinger et al., 1999; Kircher

et al., 2003; McGuire et al., 1998; Shenton et al., 1992), middle frontal (Kircher et al., 2003), and inferior frontal (Assaf et al., 2006; Cerullo et al., 2007; McGuire et al., 1998) gyri in patients with schizophrenia compared to the healthy control subjects and that, within the schizophrenia group, neurometabolite levels in these brain regions would vary in relation to the severity of thought disorder.

2. METHODS

2.1 Subjects

The study included 28 children with schizophrenia (15 boys, 13 girls), aged 8.4–17.8 years (mean age $\pm$ SD: 14.1 ± 3.0), recruited from community child psychiatry clinics. The 28 subjects were screened from a total population of 207. Two additional subjects met all inclusion and no exclusion criteria, but could not participate in the study because they were severely ill and unable to get to or lie in the scanner. A schizophrenia diagnosis was based on the Schedule for Affective Disorders and Schizophrenia for School-Age Children, Present and Lifetime Version (K-SADS-PL) (Kaufman et al., 1997), administered separately to each child and parent as described previously (Caplan et al., 2000). At the time of testing, the average duration of illness was (mean $\pm$ SD) 3.4 ± 3.1 years. To be included in the study, these children met DSM-IV criteria for schizophrenia with onset of symptoms by age 13. Exclusionary criteria included: (a) IQ < 70 , (b) bilingualism, (c) braces, (d) an underlying neurological disorder, (e) a metabolic disorder, (f) a hearing disorder, (g) left handedness, and (h) psychosis associated with an organic disorder or substance abuse. At the time of the study, 23 patients were being treated with medications: 11 with neuroleptics alone, 10 with neuroleptics plus non-neuroleptics, and 2 with non-neuroleptics alone. None of the healthy control subjects were medicated. Neuroleptic dose was expressed as chlorpromazine equivalents (CPZ) (mean $\pm$ SD for schizophrenia group: 162.3 ± 171.3).

We also included 34 healthy control subjects (15 boys, 19 girls), aged 6.4–17.2 years (mean age $\pm$ SD: 11.5 ± 2.9), who were recruited from four public and two private schools in the Los Angeles community. Both healthy control and schizophrenia subjects completed face-to-face evaluations and were screened for psychiatric, neurological, language, and hearing disorders through a structured telephone interview with a parent. We excluded any children manifesting symptoms of these disorders in the past or after enrolling in the study.

2.2 Procedures

This study was conducted in accordance with the policies of the Human Subjects Protection Committees of the University of California, Los Angeles. Informed assent and consent were obtained from all subjects and parents, respectively.

2.2.1 Neuroimaging—Structural MRI and ^1H MRSI were acquired together at 1.5 T with a Siemens Sonata scanner using a standard quadrature head coil. Study subjects were not sedated at the time of scanning. In addition to localizer scout scans, structural MRI consisted of a pair of sagittal spoiled gradient recalled (SPGR) sequences yielding high-resolution T1-weighted whole-brain volumes. Two acquisitions were performed separately and subsequently co-registered to each other and averaged to diminish the contribution of any subject movement during scanning. MRSI was acquired with PRESS (TR/TE = 1500/30 ms, NEX = 4, slab thickness = 9 mm, in-plane resolution = $11 \times 11 \text{ mm}^2$) from three slabs. Figures 1–2 indicate slab positioning and anatomical parameters used to make sure that all slabs were localized approximately in the same brain regions across subjects. The first two slabs (Figure 1) were sagittal-oblique (roughly parallel to the ipsilateral temple) in orientation and sampled the left and right peri-Sylvian region including inferior frontal, superior temporal, and other nearby cortex. The third slab (Figure 2) was coronal-oblique

and sampled bilateral middle frontal (“dorsolateral prefrontal”) cortex, mesial prefrontal cortex (pregenual anterior, anterior middle paracingulate cortex, or superior frontal cortex depending on individual subject anatomy) and prefrontal white matter (mainly corona radiata) lying in between. We excluded any subjects with clinically relevant structural abnormalities.

Using software and protocols developed at the UCLA Laboratory of Neuroimaging (Blanton et al., 2004; Taylor et al., 2005), regions-of-interest (ROIs) consisting of cortex, superjacent sulcal CSF, and subjacent white matter were sketched manually for bilateral inferior frontal gyri (IFG), superior temporal gyri (STG), and middle frontal gyri (MFG) using the sagittal T1-weighted MRI volume of each subject in multiplanar view (Figure 3). Independently, the entire brain was segregated into gray matter, white matter, and CSF subvolumes (Shattuck et al., 2001). By overlapping the ROIs with the whole-brain gray matter, white matter, and CSF subvolumes, we divided each ROI into three separate “tissue-segmented ROIs.” All personnel involved in region measurement received extensive specialized training in neuroanatomy. Intra- and interrater reliability were maintained at an intraclass correlation coefficient of at least 0.90 for each ROI.

MRSI data were post-processed with the LCModel package (Provencher, 2001), yielding metabolite values for major resonances of tNAA (2.01 ppm), Glx (2.1–2.5 ppm), Cr+PCr (3.01 ppm), Cho (3.24ppm), and mI (3.54 ppm). Numerous minor metabolites, particularly lipids and macromolecules, were included in the fit. Our self-designed MRSI Voxel Picker (MVP) program (Figure 4) (O’Neill et al., 2006) was used for co-processing of MRI and MRSI data. For each MRSI slab, MVP reconstructed the subject’s whole-brain T1-weighted gray matter, white matter, and CSF volumes, as well as the tissue-segmented ROIs, which were manually delineated as described above, into the space of the chosen MRSI slab. MVP then returned the tissue content (both on whole-brain and ROI bases) and CSF-corrected metabolite levels for each voxel in the slab and given ROI. Using automated quality control features (aided by manual inspection), the operator selected and averaged together all voxels within the ROI meeting fixed tissue-content and spectral quality criteria. For each ROI, values were averaged together for voxels containing at least 75% by volume gray matter plus white matter, a signal-to-noise ratio of three or greater, and a full width at half maximum (FWHM) less than or equal to 0.1 ppm. Only metabolite peaks satisfying the LCModel criterion of less than or equal to 20% standard deviation were included in the average. MVP implemented these quality-control criteria automatically. Additionally, with the help of a guided user interface, all voxels that passed these criteria were manually inspected by raters blind to age, gender, and diagnosis. Voxels that showed significant artifact (e.g., contaminating signals from proximal extracranial tissue) were eliminated from analysis. Typically for both subject groups, 1–4 voxels per ROI passed all quality-control criteria and were included in the subject’s average for that ROI.

2.2.2 Thought Disorder—Formal thought disorder includes measures of impaired use of language to formulate and organize thoughts and reflect higher-level discourse deficits (Caplan et al., 2000). The listener has difficulty making sense of the speech of children with formal thought disorder because of unpredicted change in the topic of conversation (e.g., loose associations) and unsound reasoning (e.g., illogical thinking).

The Story Game was administered to both control subjects and patients to obtain adequate speech samples for rating formal thought disorder. During the Story Game, the children heard an audiotaped story about a ghost and an ostracized boy, and were presented with structured open-ended questions following each story. The children also chose to tell a story about a good child, a bad child, the Incredible Hulk, or a witch.

Two raters with no knowledge of the children's psychiatric diagnosis coded videotapes of the Story Game with the Kiddie Formal Thought Disorder Rating Scale (K-FTDS) which has operationalized loose associations and illogical thinking for use in children (Caplan et al., 1989). While speaking, the child with loose associations unpredictably changes the topic of conversation without preparing the listener for the upcoming change (i.e., Interviewer: What are reasons not to like a kid? Child: I hate going to the beach). Illogical thinking is coded when the child uses unsound reasoning during causal (i.e., Interviewer: Why was the child in the story afraid? Child: I am scared because my mother's name is Jane) and non-causal utterances (i.e., Interviewer: What kind of mean things do kids do to you? Child: They tease me. And then I know I am cool) or contradicts him/herself (e.g., Interviewer: How did the story end? Child: Peter woke up and was fast asleep).

Formal thought disorder scores are frequency counts of illogical thinking and loose association ratings divided by the number of sentences (clauses) made by the child. Higher loose associations and illogical thinking scores represent more formal thought disorder. To test for inter-rater reliability, the generalizability coefficients for illogical thinking and kappa for loose associations were determined to be 0.75 (SD = 0.15) and 0.66 (SD = 0.01), respectively (Caplan et al., 2000).

2.2.3 Cognition—The Wechsler Intelligence Scale for Children-III (Wechsler, 1974) administered to both healthy control and schizophrenia subjects generated Full Scale, Verbal and Performance IQ scores.

2.2.4 Statistical Analyses—A summary measure of formal thought disorder ("formal thought disorder score") was used in all analyses of the relationship of thought disorder with MRSI neurometabolite levels, as previously described (Caplan et al., 2006). Thought disorder data from healthy control and schizophrenia subjects in all our studies ($n = 234$ healthy control, $n = 119$ schizophrenia) were used to derive the formal thought disorder score as follows. We computed a linear discriminant function for formal thought disorder, using illogical thinking and loose associations. Discriminant analysis develops a criterion (the discriminant function) to classify each subject into one of two groups. Employing a jackknife method to estimate the probability that each subject has schizophrenia, we obtained scores between 0 and 1. This process is analogous to the propensity score proposed by Rosenbaum and Rubin (Rosenbaum and Rubin, 1983). The discriminant functions were then applied to the subjects in the current study. Thus, every subject was assigned a formal thought disorder score. The propensity scores ranged from 0 to 1, with higher scores denoting greater thought disorder in subjects. These scores were then used as a continuous measure of the degree of thought disorder in the healthy controls and schizophrenia subjects.

For each ROI – namely IFG, MFG and STG – percent gray and white matter volumes, as compared to total volume of all tissues, including CSF, in the MRSI voxels were compared between the schizophrenia and control groups using univariate analysis of covariance (ANCOVA). Gender and age were used as covariates since the schizophrenia group was significantly older (14.1 ± 3.0 vs. 11.5 ± 2.9 years; $t(61) = 3.49$, $p < 0.001$) and had 6 fewer females (15 boys, 13 girls vs. 15 boys, 19 girls; $\chi^2 = 0.55$, $p = 0.46$) than the healthy control group. Even though the schizophrenia subjects had a significantly lower mean IQ score (mean $\pm$ SD, 86.1 ± 17.7) than the healthy control group (113.1 ± 15.9) ($t(55) = 5.60$, $p < 0.0005$), we did not covary IQ in the between-group comparisons, as reduced IQ reflects an illness effect (Gochman et al., 2005).

MRSI is a data-rich field-mapping neuroimaging modality. In this study MRSI yielded five metabolite endpoints in each of the three ROIs for both hemispheres, which raises an issue of multiple comparisons. We addressed this issue by carrying out repeated-measures

multivariate analyses of variance (R-MANOVAs) prior to comparing metabolite levels in individual ROIs between schizophrenia and healthy control groups. Separate R-MANOVAs were performed for each cerebral hemisphere, where ROI (IFG, MFG, STG) was used as the within-subjects factor and diagnostic group (schizophrenia, healthy control) was used as the between-subjects factor. R-MANOVAs were carried out across the five metabolite measures tNAA, Glx, Cr+PCr, Cho, and mI. Significant main effect of diagnosis or diagnosis-by-ROI interactions in the R-MANOVAs were deemed sufficient cause to compare the corresponding metabolite levels in *post-hoc* ANCOVAs covarying gender and age within the individual ROIs.

Associations between formal thought disorder scores and MRSI regional metabolite levels in the patient group were examined using R-MANOVA omnibus tests across the five metabolites tNAA, Glx, Cr+PCr, Cho, and mI computed for each hemisphere as described above. ROI and the summary measure of thought disorder were the within-subjects and between-subjects factors, respectively. Post-hoc thought disorder-metabolite correlations (age-partialled Spearman) were calculated within individual ROIs. Age and the current neuroleptic dose, expressed as chlorpromazine equivalents (CPZ), were partialled out due to possible influences on thought disorder (Caplan et al., 2000). An alpha level of 0.05 was considered to be statistically significant for all tests.

3. RESULTS

Univariate ANCOVAs of the percent of gray matter or white matter volume in the MRSI voxel between schizophrenia and healthy control groups revealed no significant differences for any ROI. The R-MANOVA also demonstrated no significant between-group differences for either set of metabolites in both hemispheres. Table 1 summarizes average metabolite values in all cortical regions studied for both schizophrenia and healthy control groups.

For the schizophrenia group, the R-MANOVA omnibus testing demonstrated a significant multivariate main effect of formal thought disorder ($F_{5,13} = 6.2, p < 0.005$) in the left hemisphere. There were no significant R-MANOVA findings in the right hemisphere of the schizophrenia group.

Figure 5 and Table 2 summarize age-partialled Spearman correlations between metabolite levels and formal thought disorder score for the schizophrenia group. In the left hemisphere controlling for age and CPZ equivalents, the severity of thought disorder was significantly and positively correlated with tNAA in the superior temporal and inferior frontal gyri and with Cr+PCr in the superior temporal gyrus. These correlations enjoyed omnibus protection for multiple comparisons.

4. DISCUSSION

Corroborating the findings of prior PET (McGuire et al., 1998), fMRI (Assaf et al., 2006; Kircher et al., 2003), and volumetric studies (Holinger et al., 1999; Shenton et al., 1992), this study demonstrated that increased metabolite levels (tNAA and Cr+PCr) in language-related brain regions (left superior temporal gyrus and/or inferior frontal gyrus) were associated with formal thought disorder. We found no significant metabolite differences between the schizophrenia and control groups.

Unlike many studies of adult subjects with schizophrenia, schizophreniform disorder, or at risk for schizophrenia (Bertolino et al., 1996; Bertolino et al., 1998; Bertolino and Weinberger, 1999; Bertolino et al., 2001; Bertolino et al., 2003; Block et al., 2000; Bustillo et al., 2001; Callicott et al., 2000; Cecil et al., 1999; Choe et al., 1994; Deicken et al., 1997; Heimberg et al., 1998; Jessen et al., 2006; Lim et al., 1998; Molina et al., 2005; Molina et

al., 2006), but similar to several other studies (Bartha et al., 1999; Rusch et al., 2008; Sigmundsson et al., 2003; Szulc et al., 2005; Wood et al., 2003), we did not observe below-normal tNAA in the middle frontal gyri. Findings are also inconsistent in the few pediatric studies of subjects with or at risk for schizophrenia or schizophreniform disorders (reviewed in (Mehler and Warnke, 2002)) with five finding below-normal tNAA (Bertolino et al., 1998; Brooks et al., 1998; Hagino et al., 2002; Pae et al., 2004; Stanley et al., 2007) and three reporting no abnormality (Bartha et al., 1999; Wood et al., 2006; Wood et al., 2003). Four methodological considerations may explain why our results vary with the majority of other studies: subject age, chronicity, MRS voxel location and tissue composition, and use of metabolite ratios.

In terms of age, Bertolino et al. (Bertolino et al., 1996) and our two prior studies (O'Neill et al., 2004; Thomas et al., 1998) investigated patients with onset by age 13, whereas other studies (Stanley et al., 2007; Wood et al., 2006; Wood et al., 2003; Zabala et al., 2007) included adolescent-onset subjects. Findings of tNAA/Cr+PCr abnormalities in children with schizophrenia (Bertolino et al., 1998), similar to those shown in adults, might reflect both the older mean age (16.4 ± 1.7 years) and smaller sample size ($N=14$) of the subjects with schizophrenia compared to a mean age of 14.1 ± 3.0 years and larger sample size of 28 in our study.

Regarding the chronicity of the illness, our subjects might not have been ill long enough for the development of the metabolite abnormalities found in adult schizophrenia. In support of this explanation, two studies (Molina et al., 2005; Molina et al., 2006) found tNAA/Cr+PCr and tNAA/Cho deficits in chronically ill but not in first-episode schizophrenia. As for medication effects, there is evidence that neuroleptics, especially atypical antipsychotics, increase cortical NAA (Bertolino et al., 2001; Braus et al., 2001; Braus et al., 2002; Ende et al., 2000; Heimberg et al., 1998; Molina et al., 2007). Therefore, atypical antipsychotics, prescribed to most of our patients, may have masked tNAA deficits.

Voxel location and tissue composition may also help explain the differences across studies. For example, the frontal cortical tNAA deficits in several studies (Block et al., 2000; Deicken et al., 1997; Heimberg et al., 1998; Jessen et al., 2006; Pae et al., 2004) included regions outside of the middle frontal gyrus. Regarding tissue composition, in many cases (Brooks et al., 1998; Choe et al., 1994; Lim et al., 1998; Molina et al., 2005; Molina et al., 2006) the voxel included white matter. This is relevant because two groups (Lim et al., 1998); (Bartha et al., 1999) suggest that low tNAA/Cr+PCr in schizophrenia may be restricted to white matter or due to higher voxel white matter content. We delineated the middle frontal gyri of all the subjects in our study according to a standardized procedure using voxels that contained $\geq 75\%$ middle frontal gyrus. Moreover, volume percent gray matter and white matter did not differ significantly between the patient and healthy control groups in these voxels (data not shown). Thus, metabolite values in our study may more accurately represent the middle frontal gyrus and may be less influenced by between-group differences in tissue content than those of other studies.

Additionally, many studies detecting tNAA deficits (Block et al., 2000; Callicott et al., 2000; Cecil et al., 1999; Choe et al., 1994; Jessen et al., 2006; Molina et al., 2005; Molina et al., 2006; Molina et al., 2007) expressed their findings as the metabolite ratios tNAA/Cr +PCr and/or tNAA/Cho. However, similar to our study, studies that found no metabolite deficits (Bartha et al., 1999; Rusch et al., 2008; Sigmundsson et al., 2003; Szulc et al., 2005) employed absolute metabolite analyses. Effects observed with ratios can be attributable to either the numerator or the denominator (e.g., to tNAA or to Cr+PCr). One group (Wood et al., 2003), in fact, observed *above-normal* tNAA/Cr+PCr in adolescent and adult subjects at ultra-high risk for schizophrenia, an effect they attributed to Cr hypometabolism.

Overall, the inconsistent findings and methodological differences across studies imply that frontal cortical tNAA may not necessarily be diminished in childhood-onset schizophrenia. Our study of subjects with childhood-onset schizophrenia acquired MRSI at high spatial resolution and short-TE from voxels with carefully defined anatomic localization and tissue composition. Its findings provide additional evidence against below-normal tNAA levels in middle and inferior frontal gyri and superior temporal gyri in pediatric schizophrenia. Some studies have found cortical atrophy of the superior temporal gyrus in adult schizophrenia (Holinger et al., 1999; Shenton et al., 1992). Such atrophy, however, may occur through mechanisms that do not necessarily imply a drop in tNAA, e.g., uniform loss of all neurons and glia, selective loss of glia, and/or dehydration. Studies with larger sample size are needed to replicate these results.

To our knowledge, this is the first study to investigate associations between neurometabolites and thought disorder in schizophrenia. A positive correlation was found between tNAA and thought disorder. This is somewhat counterintuitive, as tNAA has been posited as a neuronal marker (Demougeot et al., 2001; Urenjak et al., 1993) and higher levels might be indicative of healthier neurons. As discussed above, however, the presence of a tNAA deficit in schizophrenia is by no means assured, particularly in children or early in the disease. The next paragraph suggests a mechanism whereby the NAAG molecule contributing to the tNAA signal may be elevated in microglial pathology and help bring about thought disorder. The significant correlations found in this study were illustrated in the left superior temporal and inferior frontal gyri. This corroborates previous structural and functional imaging work demonstrating that anatomical and functional abnormalities in these language-related regions are associated with thought disorder (Assaf et al., 2006; Cerullo et al., 2007; Holinger et al., 1999; Kircher et al., 2003; McGuire et al., 1998; Shenton et al., 1992). Functional MRI findings that normal topic maintenance (the opposite of loose associations) and use of logical reasoning (in contrast to illogical thinking) in conversation are related to cortical activation of the same regions lend further support to the finding that metabolites in these regions are correlated with thought disorder (Caplan and Dapretto, 2001). All of the above observations suggest that neurochemical imbalances in language-related cortical regions are associated with illogical thinking and loosening of associations in childhood-onset schizophrenia.

The specific role of tNAA and Cr+PCr, however, in thought disorder requires extensive future biochemical investigation. NMDA receptors are a necessary element for associative memory formation (Tsien, 2000) and thus might be involved in the normal associative chains engaged in thought and speech (Hall et al., 2009; Kircher, 2008). Additionally, they have been proposed to be hypoactive in schizophrenia (Coyle et al., 2002; Coyle et al., 2003). NAAG, a component of the tNAA peak (Edden et al., 2007; Pouwels and Frahm, 1997), antagonizes NMDA receptors (Bergeron et al., 2005; Westbrook et al., 1986), suggesting that future studies should delineate the biochemistry of NAAG in schizophrenia. Specifically, microgliosis, an already demonstrated phenomenon of schizophrenia (Bayer et al., 1999; van Berckel et al., 2008), may contribute to the above NMDA receptor antagonism since microglia contain high levels of NAAG (Passani et al., 1998). PCr levels are also high in microglia (de Gannes et al., 1998), which may help explain the observed Cr+PCr association with thought disorder if the microglia are activated in childhood onset schizophrenia. Thus, this study's correlations emphasize that the neurochemistry of language-related cortical regions are associated with thought disorder, and they also suggest specific biochemical pathways for future analysis.

Limitations of the study include residual partial voluming (even at 1.1 cc voxel size) as well as the age and gender differences between the schizophrenia and control subjects. Because the concentration of NAA changes as the brain matures (Kadota et al., 2001), age and

gender differences (although statistically covaried) may contribute to our findings. Psychotropic medication effects (as described above) other than partialled-out CPZ equivalents, is a further limitation. Our calculations of CPZ equivalents addressed atypical antipsychotic doses, but it should be noted that the precise conversions for atypicals to CPZ equivalents is disputed in the literature (Woods, 2003). While IQ was not statistically covaried because of its assumed illness effect, it might have nevertheless influenced the study's findings because lower IQ is related to below-normal tNAA (Jung et al., 1999), above-normal Cho (Jung et al., 1999), and more thought disorder in adult patients with schizophrenia (Willis-Shore et al., 2000). A further limitation was MRSI acquisition at 1.5 T since acquisition at 3 T might have enabled us to separately quantitate the NAA and NAAG peaks with the aid of specialized pulse sequences (Edden et al., 2007).

Despite these limitations, the present findings highlight that neurometabolite levels in language regions of the brain are associated with formal thought disorder in childhood onset schizophrenia. Although preliminary, they also imply a need to further investigate the possible biochemical roles of tNAA and Cr+PCr in language and how they may be abnormal in the brain of schizophrenia patients with thought disorder.

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References

- Assaf M, Rivkin P, Kuzu C, Calhoun V, Kraut M, Groth K, Yassa M, Hart JJ, Pearlson G. Abnormal object recall and anterior cingulate overactivation correlate with formal thought disorder in schizophrenia. *Biol Psychiatry*. 2006; 59(5):452–459. [PubMed: 16199012]
- Bartha R, al-Semaan YM, Williamson PC, Drost DJ, Malla AK, Carr TJ, Densmore M, Canaran G, Neufeld RW. A short echo proton magnetic resonance spectroscopy study of the left mesial-temporal lobe in first-onset schizophrenic patients. *Biol Psychiatry*. 1999; 45(11):1403–1411. [PubMed: 10356621]
- Bayer TA, Buslei R, Havas L, Falkai P. Evidence for activation of microglia in patients with psychiatric illnesses. *Neurosci Lett*. 1999; 271(2):126–128. [PubMed: 10477118]
- Bergeron R, Coyle J, Tsai G, Greene R. NAAG reduces NMDA receptor current in CA1 hippocampal pyramidal neurons of acute slices and dissociated neurons. *Neuropsychopharmacology*. 2005; 30(1):7–16. [PubMed: 15354184]
- Bertolino A, Nawroz S, Mattay V, Barnett A, Duyn J, Moonen C, Frank J, Tedeschi G, Weinberger D. Regionally specific pattern of neurochemical pathology in schizophrenia as assessed by multislice proton magnetic resonance spectroscopic imaging. *Am J Psychiatry*. 1996; 153(12):1554–1563. [PubMed: 8942451]
- Bertolino A, Kumra S, Callicott J, Mattay V, Lestz R, Jacobsen L, Barnett I, Duyn J, Frank J, Rapoport J, Weinberger D. Common pattern of cortical pathology in childhood-onset and adult-onset schizophrenia as identified by proton magnetic resonance spectroscopic imaging. *Am J Psychiatry*. 1998; 155(10):1376–1383. [PubMed: 9766769]
- Bertolino A, Knable MB, Saunders RC, Callicott JH, Kolachana B, Mattay VS, Bachevalier J, Frank JA, Egan M, Weinberger DR. The relationship between dorsolateral prefrontal N-acetylaspartate measures and striatal dopamine activity in schizophrenia. *Biol Psychiatry*. 1999; 45(6):660–667. [PubMed: 10187995]

- Bertolino A, Weinberger D. Proton magnetic resonance spectroscopy in schizophrenia. *Eur J Radiol.* 1999; 30(2):132–141. [PubMed: 10401594]
- Bertolino A, Callicott JH, Mattay VS, Weidenhammer KM, Rakow R, Egan MF, Weinberger DR. The effect of treatment with antipsychotic drugs on brain N-acetylaspartate measures in patients with schizophrenia. *Biol Psychiatry.* 2001; 49(1):39–46. [PubMed: 11163778]
- Bertolino A, Sciota D, Brudaglio F, Altamura M, Blasi G, Bellomo A, Antonucci N, Callicott JH, Goldberg TE, Scarabino T, Weinberger DR, Nardini M. Working memory deficits and levels of N-acetylaspartate in patients with schizophreniform disorder. *Am J Psychiatry.* 2003; 160(3):483–489. [PubMed: 12611829]
- Blanton R, Levitt J, Peterson J, Fadale D, Sporty M, Lee M, To D, Mormino E, Thompson P, McCracken J, Toga A. Gender differences in the left inferior frontal gyrus in normal children. *Neuroimage.* 2004; 22(2):626–636. [PubMed: 15193591]
- Block W, Bayer TA, Tepest R, Traber F, Rietschel M, Muller DJ, Schulze TG, Honer WG, Maier W, Schild HH, Falkai P. Decreased frontal lobe ratio of N-acetyl aspartate to choline in familial schizophrenia: a proton magnetic resonance spectroscopy study. *Neurosci Lett.* 2000; 289(2):147–151. [PubMed: 10904141]
- Borofsky L, McNealy K, Siddarth P, Wu K, Dapretto M, Caplan R. Semantic processing and thought disorder in childhood-onset schizophrenia: Insights from fMRI. *J Neuroling Res.* in press.
- Braus DF, Ende G, Weber-Fahr W, Demirakca T, Henn FA. Favorable effect on neuronal viability in the anterior cingulate gyrus due to long-term treatment with atypical antipsychotics: an MRSI study. *Pharmacopsychiatry.* 2001; 34(6):251–253. [PubMed: 11778146]
- Braus DF, Ende G, Weber-Fahr W, Demirakca T, Tost H, Henn FA. Functioning and neuronal viability of the anterior cingulate neurons following antipsychotic treatment: MR-spectroscopic imaging in chronic schizophrenia. *Eur Neuropsychopharmacol.* 2002; 12(2):145–152. [PubMed: 11872332]
- Brooks WM, Hodde-Vargas J, Vargas LA, Yeo RA, Ford CC, Hendren RL. Frontal lobe of children with schizophrenia spectrum disorders: a proton magnetic resonance spectroscopic study. *Biol Psychiatry.* 1998; 43(4):263–269. [PubMed: 9513735]
- Bustillo JR, Lauriello J, Rowland LM, Jung RE, Petropoulos H, Hart BL, Blanchard J, Keith SJ, Brooks WM. Effects of chronic haloperidol and clozapine treatments on frontal and caudate neurochemistry in schizophrenia. *Psychiatry Res.* 2001; 107(3):135–149. [PubMed: 11566430]
- Callicott JH, Bertolino A, Mattay V, Langheim F, Duyn J, Coppola R, Goldberg T, Weinberger D. Physiological dysfunction of the dorsolateral prefrontal cortex in schizophrenia revisited. *Cereb Cortex.* 2000; 10(11):1078–1092. [PubMed: 11053229]
- Caplan R, Guthrie D, Fish B, Tanguay P, David-Lando G. The Kiddie Formal Thought Disorder Rating Scale: clinical assessment, reliability, and validity. *J Am Acad Child Adolesc Psychiatry.* 1989; 28(3):408–416. [PubMed: 2738008]
- Caplan R, Guthrie D, Tang B, Komo S, Asarnow R. Thought disorder in childhood schizophrenia: replication and update of concept. *J Am Acad Child Adolesc Psychiatry.* 2000; 39(6):771–778. [PubMed: 10846312]
- Caplan R, Dapretto M. Making sense during conversation: an fMRI study. *Neuroreport.* 2001; 12(16):3625–3632. [PubMed: 11733724]
- Caplan R, Siddarth P, Bailey C, Lanphier E, Gurbani S, Donald Shields W, Sankar R. Thought disorder: A developmental disability in pediatric epilepsy. *Epilepsy Behav.* 2006; 8(4):726–735. [PubMed: 16678493]
- Cecil KM, Lenkinski RE, Gur RE, Gur RC. Proton magnetic resonance spectroscopy in the frontal and temporal lobes of neuroleptic naive patients with schizophrenia. *Neuropsychopharmacology.* 1999; 20(2):131–140. [PubMed: 9885793]
- Cerullo M, Adler C, Strakowski S, Eliassen J, Nasrallah H, Nasrallah A. Memantine normalizes brain activity in the inferior frontal gyrus: a controlled pilot fMRI study. *Schizophr Res.* 2007; 97(1–3):294–296. [PubMed: 17855055]
- Choe BY, Kim KT, Suh TS, Lee C, Paik IH, Bahk YW, Shinn KS, Lenkinski RE. 1H magnetic resonance spectroscopy characterization of neuronal dysfunction in drug-naive, chronic schizophrenia. *Acad Radiol.* 1994; 1(3):211–216. [PubMed: 9419488]

- Coyle J, Tsai G, Goff D. Ionotropic glutamate receptors as therapeutic targets in schizophrenia. *Curr Drug Targets CNS Neurol Disord.* 2002; 1(2):183–189. [PubMed: 12769626]
- Coyle J, Tsai G, Goff D. Converging evidence of NMDA receptor hypofunction in the pathophysiology of schizophrenia. *Ann N Y Acad Sci.* 2003; 1003:318–327. [PubMed: 14684455]
- de Gannes F, Merle M, Canioni P, Voisin P. Metabolic and cellular characterization of immortalized human microglial cells under heat stress. *Neurochem Int.* 1998; 33(1):61–73. [PubMed: 9694044]
- Deicken R, Zhou L, Corwin F, Vinogradov S, Weiner MW. Decreased left frontal lobe N-acetylaspartate in schizophrenia. *Am J Psychiatry.* 1997; 154(5):688–690. [PubMed: 9137129]
- Deicken R, Zhou L, Schuff N, Fein G, Weiner M. Hippocampal neuronal dysfunction in schizophrenia as measured by proton magnetic resonance spectroscopy. *Biol Psychiatry.* 1998; 43(7):483–488. [PubMed: 9547926]
- Deicken R, Pegues M, Amend D. Reduced hippocampal N-acetylaspartate without volume loss in schizophrenia. *Schizophr Res.* 1999; 37(3):217–223. [PubMed: 10403193]
- Demougeot C, Garnier P, Mossiat C, Bertrand N, Giroud M, Beley A, Marie C. N-Acetylaspartate, a marker of both cellular dysfunction and neuronal loss: its relevance to studies of acute brain injury. *J Neurochem.* 2001; 77(2):408–415. [PubMed: 11299303]
- Edden R, Pomper M, Barker P. In vivo differentiation of N-acetyl aspartyl glutamate from N-acetyl aspartate at 3 Tesla. *Magn Reson Med.* 2007; 57(6):977–982. [PubMed: 17534922]
- Ende G, Braus D, Walter S, Weber-Fahr W, Soher B, Maudsley A, Henn F. Effects of age, medication, and illness duration on the N-acetyl aspartate signal of the anterior cingulate region in schizophrenia. *Schizophr Res.* 2000; 41(3):389–395. [PubMed: 10728716]
- Gochman PA, Greenstein D, Sporn A, Gogtay N, Keller B, Shaw P, Rapoport JL. IQ stabilization in childhood-onset schizophrenia. *Schizophr Res.* 2005; 77(2–3):271–277. [PubMed: 15913958]
- Hagino H, Suzuki M, Mori K, Nohara S, Yamashita I, Takahashi T, Kurokawa K, Matsui M, Watanabe N, Seto H, Kurachi M. Proton magnetic resonance spectroscopy of the inferior frontal gyrus and thalamus and its relationship to verbal learning task performance in patients with schizophrenia: a preliminary report. *Psychiatry Clin Neurosci.* 2002; 56(5):499–507. [PubMed: 12193238]
- Hall J, Romaniuk L, McIntosh A, Steele J, Johnstone E, Lawrie S. Associative learning and the genetics of schizophrenia. *Trends Neurosci.* 2009
- Heimberg C, Komoroski RA, Lawson WB, Cardwell D, Karson CN. Regional proton magnetic resonance spectroscopy in schizophrenia and exploration of drug effect. *Psychiatry Res.* 1998; 83(2):105–115. [PubMed: 9818736]
- Holinger D, Shenton M, Wible C, Donnino R, Kikinis R, Jolesz F, McCarley R. Superior temporal gyrus volume abnormalities and thought disorder in left-handed schizophrenic men. *Am J Psychiatry.* 1999; 156(11):1730–1735. [PubMed: 10553736]
- Jessen F, Scherk H, Traber F, Theyson S, Berning J, Tepest R, Falkai P, Schild H, Maier W, Wagner M, Block W. Proton magnetic resonance spectroscopy in subjects at risk for schizophrenia. *Schizophr Res.* 2006; 87(1–3):81–88. [PubMed: 16842971]
- Jung R, Brooks W, Yeo R, Chiulli S, Weers D, Sibbitt WJ. Biochemical markers of intelligence: a proton MR spectroscopy study of normal human brain. *Proc Biol Sci.* 1999; 266(1426):1375–1379. [PubMed: 10445292]
- Kadota T, Horinouchi T, Kuroda C. Development and aging of the cerebrum: assessment with proton MR spectroscopy. *AJNR Am J Neuroradiol.* 2001; 22(1):128–135. [PubMed: 11158898]
- Kaufman J, Birmaher B, Brent D, Rao U, Flynn C, Moreci P, Williamson D, Ryan N. Schedule for Affective Disorders and Schizophrenia for School-Age Children-Present and Lifetime Version (K-SADS-PL): initial reliability and validity data. *J Am Acad Child Adolesc Psychiatry.* 1997; 36(7):980–988. [PubMed: 9204677]
- Kircher T, Liddle P, Brammer M, Murray R, McGuire P. [Neural correlates of “negative” formal thought disorder]. *Nervenarzt.* 2003; 74(9):748–754. [PubMed: 14504772]
- Kircher T. [Neurobiological foundations of thought and language disorder in schizophrenia]. *Fortschr Neurol Psychiatr.* 2008; 76(Suppl 1):S24–32. [PubMed: 18461541]

- Lim K, Adalsteinsson E, Spielman D, Sullivan E, Rosenbloom M, Pfefferbaum A. Proton magnetic resonance spectroscopic imaging of cortical gray and white matter in schizophrenia. *Arch Gen Psychiatry*. 1998; 55(4):346–352. [PubMed: 9554430]
- Maier M, Ron M, Barker G, Tofts P. Proton magnetic resonance spectroscopy: an in vivo method of estimating hippocampal neuronal depletion in schizophrenia. *Psychol Med*. 1995; 25(6):1201–1209. [PubMed: 8637950]
- Maudsley, A. MR spectroscopic imaging. In: Toga, AW.; Mazziotta, JC., editors. *Brain Mapping: The Methods*. Academic Press; Amsterdam: 2002. p. 351-378.
- McGuire P, Quested D, Spence S, Murray R, Frith C, Liddle P. Pathophysiology of ‘positive’ thought disorder in schizophrenia. *Br J Psychiatry*. 1998; 173:231–235. [PubMed: 9926099]
- Mehler C, Warnke A. Structural brain abnormalities specific to childhood-onset schizophrenia identified by neuroimaging techniques. *J Neural Transm*. 2002; 109(2):219–234. [PubMed: 12075863]
- Molina V, Sanchez J, Reig S, Sanz J, Benito C, Santamarta C, Pascau J, Sarramea F, Gisbert JD, Misiego JM, Palomo T, Desco M. N-acetyl-aspartate levels in the dorsolateral prefrontal cortex in the early years of schizophrenia are inversely related to disease duration. *Schizophr Res*. 2005; 73(2–3):209–219. [PubMed: 15653263]
- Molina V, Sanz J, Sarramea F, Luque R, Benito C, Palomo T. No association between dorsolateral prefrontal gray matter deficit and N-acetyl aspartate ratios in schizophrenia. *Neuropsychobiology*. 2006; 54(3):171–178. [PubMed: 17230035]
- Molina V, Sanchez J, Sanz J, Reig S, Benito C, Leal I, Sarramea F, Rebolledo R, Palomo T, Desco M. Dorsolateral prefrontal N-acetyl-aspartate concentration in male patients with chronic schizophrenia and with chronic bipolar disorder. *Eur Psychiatry*. 2007; 22(8):505–512. [PubMed: 17904824]
- O’Neill J, Levitt J, Caplan R, Asarnow R, McCracken J, Toga A, Alger J. 1H MRSI evidence of metabolic abnormalities in childhood-onset schizophrenia. *Neuroimage*. 2004; 21(4):1781–1789. [PubMed: 15050598]
- O’Neill, J.; Tseng, PB.; Pagare, R.; Frew, AJ.; Tafazoli, S.; Alger, JR. MRSI Voxel Picker (MVP): Software for co-evaluation of structural and spectroscopic information. *ISMRM Workshop on Quantitative Spectroscopy*; Warrenton, VA. 2006.
- Pae CU, Choe BY, Joo RH, Lim HK, Kim TS, Yoo SS, Choi BG, Kim JJ, Lee SJ, Lee C, Paik IH, Lee CU. Neuronal dysfunction of the frontal lobe in schizophrenia. *Neuropsychobiology*. 2004; 50(3): 211–215. [PubMed: 15365217]
- Passani L, Elkabes S, Coyle J. Evidence for the presence of N-acetylaspartylglutamate in cultured oligodendrocytes and LPS activated microglia. *Brain Res*. 1998; 794(1):143–145. [PubMed: 9630582]
- Pouwels P, Frahm J. Differential distribution of NAA and NAAG in human brain as determined by quantitative localized proton MRS. *NMR Biomed*. 1997; 10(2):73–78. [PubMed: 9267864]
- Provencher S. Automatic quantitation of localized in vivo 1H spectra with LCModel. *NMR Biomed*. 2001; 14(4):260–264. [PubMed: 11410943]
- Rosenbaum P, Rubin D. The central role of the propensity score in observational studies. *Biometrika*. 1983; 70:41–45.
- Rusch N, Tebartz van Elst L, Valerius G, Buchert M, Thiel T, Ebert D, Hennig J, Olbrich H. Neurochemical and structural correlates of executive dysfunction in schizophrenia. *Schizophr Res*. 2008; 99(1–3):155–163. [PubMed: 17616347]
- Shattuck D, Sandor-Leahy S, Schaper K, Rottenberg D, Leahy R. Magnetic resonance image tissue classification using a partial volume model. *Neuroimage*. 2001; 13(5):856–876. [PubMed: 11304082]
- Shenton M, Kikinis R, Jolesz F, Pollak S, LeMay M, Wible C, Hokama H, Martin J, Metcalf D, Coleman M. Abnormalities of the left temporal lobe and thought disorder in schizophrenia. A quantitative magnetic resonance imaging study. *N Engl J Med*. 1992; 327(9):604–612. [PubMed: 1640954]

- Sigmundsson T, Maier M, Toone BK, Williams SC, Simmons A, Greenwood K, Ron MA. Frontal lobe N-acetylaspartate correlates with psychopathology in schizophrenia: a proton magnetic resonance spectroscopy study. *Schizophr Res*. 2003; 64(1):63–71. [PubMed: 14511802]
- Stanley J, Vemulapalli M, Nutche J, Montrose DM, Sweeney JA, Pettegrew JW, MacMaster FP, Keshavan MS. Reduced N-acetyl-aspartate levels in schizophrenia patients with a younger onset age: a single-voxel 1H spectroscopy study. *Schizophr Res*. 2007; 93(1–3):23–32. [PubMed: 17498928]
- Steen R, Hamer R, Lieberman J. Measurement of brain metabolites by 1H magnetic resonance spectroscopy in patients with schizophrenia: a systematic review and meta-analysis. *Neuropsychopharmacology*. 2005; 30(11):1949–1962. [PubMed: 16123764]
- Szulc A, Galinska B, Tarasow E, Dzienis W, Kubas B, Konarzewska B, Walecki J, Alathiaki AS, Czernikiewicz A. The effect of risperidone on metabolite measures in the frontal lobe, temporal lobe, and thalamus in schizophrenic patients. A proton magnetic resonance spectroscopy (1H MRS). *Pharmacopsychiatry*. 2005; 38(5):214–219. [PubMed: 16189748]
- Taylor J, Blanton R, Levitt J, Caplan R, Nobel D, Toga A. Superior temporal gyrus differences in childhood-onset schizophrenia. *Schizophr Res*. 2005; 73(2–3):235–241. [PubMed: 15653266]
- Thomas MA, Ke Y, Levitt J, Caplan R, Curran J, Asarnow R, McCracken J. Preliminary study of frontal lobe 1H MR spectroscopy in childhood-onset schizophrenia. *J Magn Reson Imaging*. 1998; 8(4):841–846. [PubMed: 9702885]
- Tibbo P, Hanstock C, Valiakalayil A, Allen P. 3-T proton MRS investigation of glutamate and glutamine in adolescents at high genetic risk for schizophrenia. *Am J Psychiatry*. 2004; 161(6):1116–1118. [PubMed: 15169703]
- Tsien J. Linking Hebb's coincidence-detection to memory formation. *Curr Opin Neurobiol*. 2000; 10(2):266–273. [PubMed: 10753792]
- Urenjak J, Williams S, Gadian D, Noble M. Proton nuclear magnetic resonance spectroscopy unambiguously identifies different neural cell types. *J Neurosci*. 1993; 13(3):981–989. [PubMed: 8441018]
- van Berckel B, Bossong M, Boellaard R, Kloet R, Schuitemaker A, Caspers E, Luurtsema G, Windhorst A, Cahn W, Lammertsma A, Kahn R. Microglia Activation in Recent-Onset Schizophrenia: A Quantitative (R)-[(11)C]PK11195 Positron Emission Tomography Study. *Biol Psychiatry*. 2008
- Wechsler, D. Manual for the Wechsler Intelligence Scale for Children. Revised. Psychological Corp; New York: 1974.
- Westbrook G, Mayer M, Namboodiri M, Neale J. High concentrations of N-acetylaspartylglutamate (NAAG) selectively activate NMDA receptors on mouse spinal cord neurons in cell culture. *J Neurosci*. 1986; 6(11):3385–3392. [PubMed: 3021930]
- Willis-Shore J, Poole J, Skinner H, Benioff L, Vinogradov S. Two commonly used indices of thought disorder. *Schizophr Res*. 2000; 42(3):261–262. [PubMed: 10896411]
- Wood S, Berger G, Lambert M, Conus P, Velakoulis D, Stuart G, Desmond P, McGorry P, Pantelis C. Prediction of functional outcome 18 months after a first psychotic episode: a proton magnetic resonance spectroscopy study. *Arch Gen Psychiatry*. 2006; 63(9):969–976. [PubMed: 16952999]
- Wood SJ, Berger G, Velakoulis D, Phillips LJ, McGorry PD, Yung AR, Desmond P, Pantelis C. Proton magnetic resonance spectroscopy in first episode psychosis and ultra high-risk individuals. *Schizophr Bull*. 2003; 29(4):831–843. [PubMed: 14989417]
- Woods SW. Chlorpromazine equivalent doses for the newer atypical antipsychotics. *J Clin Psychiatry*. 2003; 64(6):663–667. [PubMed: 12823080]
- Yamasue H, Fukui T, Fukuda R, Yamada H, Yamasaki S, Kuroki N, Abe O, Kasai K, Tsujii K, Iwanami A, Aoki S, Ohtomo K, Kato N, Kato T. 1H-MR spectroscopy and gray matter volume of the anterior cingulate cortex in schizophrenia. *Neuroreport*. 2002; 13(16):2133–2137. [PubMed: 12438941]
- Yasukawa R, Miyaoka T, Mizuno S, Inagaki T, Horiguchi J, Oda K, Kitagaki H. Proton magnetic resonance spectroscopy of the anterior cingulate gyrus, insular cortex and thalamus in schizophrenia associated with idiopathic unconjugated hyperbilirubinemia (Gilbert's syndrome). *J Psychiatry Neurosci*. 2005; 30(6):416–422. [PubMed: 16327875]

Zabala A, Sanchez-Gonzalez J, Parellada M, Moreno D, Reig S, Burdalo M, Robles O, Desco M, Arango C. Findings of proton magnetic resonance spectrometry in the dorsolateral prefrontal cortex in adolescents with first episodes of psychosis. *Psychiatry Res.* 2007; 156(1):33–42. [PubMed: 17764911]

Figure 1. Sagittal-oblique (*center*), coronal (*upper right*), and axial-oblique (*lower right*) MRI depicting positioning of 9-mm thick MRSI slabs (one left hemisphere, one right) in peri-Sylvian region. The PRESS volume was aligned parallel to the temple, set ~2 cm deep into the brain, sized to the subject's anatomy and rotated counterclockwise parallel to the Sylvian fissure or beyond to sample inferior frontal (*blue square*) and superior temporal cortex. (*Left*) ^1H MR spectrum (TR/TE = 1500/30 milliseconds) from voxel sampled. *Red* trace is post-acquisition fit of signal by scanner software; more precise fits were obtained offline using LCModel.

Figure 2.

Coronal-oblique (*left*), sagittal (*right upper*), and axial (*right lower*) MRI depicting positioning of 9-mm thick “dorsolateral prefrontal cortex” MRSI slab. The coronal-oblique plane was acquired 43° counterclockwise to the genu-splenium line as seen in the sagittal plane. The PRESS volume was oriented parallel to the aforementioned coronal-oblique plane then, in the sagittal plane, aligned parallel to the first cortical gyrus (paracingulate or superior frontal gyrus depending on subject) exterior to the rostral cingulate gyrus. Within the coronal-oblique plane, the PRESS volume was then sized and positioned to encompass as much lateral cortex as possible without contacting extracranial lipids. In addition to bilateral middle frontal (“dorsolateral prefrontal”) cortex, the PRESS volume samples medial prefrontal cortex and prefrontal white matter.

Figure 3. Delineation of inferior frontal (*top*), superior temporal (*bottom right*), and middle frontal (*bottom left*) gyri. Figures adapted from Blanton *et al.* (Blanton et al., 2004) and Taylor *et al.* (Taylor et al., 2005).

Figure 4.

Guided-user interface for the MRSI Voxel Picker (MVP) software package developed at UCLA. A high-resolution T1 weighted whole-brain MRI volume from a subject with childhood schizophrenia (upper right) is co-registered with a PRESS MRSI voxel grid (green; PRESS volume outlined lightly in white) and an anatomic region of interest (blue) from the same subject. The user selects an arbitrary MRSI voxel (maroon) and the matching LCModel-fit MR spectrum appears (lower right). Voxel volume% gray matter, white matter, and CSF (for whole-brain and region-of-interest); LCModel-derived QC-parameters and metabolite levels; and levels corrected for voxel whole-brain CSF content also appear (lower left). Example is for MRSI (1.5 T, TR/TE = 1500/30 ms) acquired in a bilateral “dorsolateral prefrontal cortex” (DLPFC) slab. Left middle frontal cortex (L-MFG-gm) is the highlighted region of interest.

Figure 5. Sample Spearman correlations between MRSI tNAA (NAA+NAAG; upper) and Cr+PCr (lower) in left superior temporal gyrus (Institutional Units, IU) and formal thought disorder controlling for age and chlorpromazine (CPZ) equivalents in schizophrenia (blue diamonds) and control subjects (red squares). $p < *0.05$, $**0.01$ (after repeated-measures MANOVA). FTD = Formal Thought Disorder propensity score between 0 and 1 represents the estimated probability that a subject will exhibit thought disorder.

Table 1Mean $\pm$ SD Regional Metabolite Values in Childhood Onset Schizophrenia and Healthy Control Groups

	Left Hemisphere		Right Hemisphere	
Metabolite	Patients	Controls	Patients	Controls
	Inferior Frontal Gyrus			
tNAA	8.0 $\pm$ 1.0	7.4 $\pm$ 1.0	7.4 $\pm$ 1.2	8.0 $\pm$ 1.1
Glx	10.4 $\pm$ 2.5	10.6 $\pm$ 1.7	11.9 $\pm$ 2.6	11.8 $\pm$ 2.0
Cr+PCr	4.1 $\pm$ 0.5	4.3 $\pm$ 0.7	4.3 $\pm$ 0.9	4.3 $\pm$ 0.6
Cho	1.1 $\pm$ 0.2	1.1 $\pm$ 0.2	1.2 $\pm$ 0.2	1.3 $\pm$ 0.2
mI	3.5 $\pm$ 0.5	3.6 $\pm$ 0.6	3.7 $\pm$ 0.6	3.8 $\pm$ 0.6
	Middle Frontal Gyrus			
tNAA	6.8 $\pm$ 1.9	6.4 $\pm$ 2.1	7.1 $\pm$ 1.7	6.8 $\pm$ 1.7
Glx	11.1 $\pm$ 4.2	11.1 $\pm$ 3.5	11.1 $\pm$ 3.3	11.3 $\pm$ 3.8
Cr+PCr	4.2 $\pm$ 1.0	4.0 $\pm$ 1.0	4.3 $\pm$ 0.9	4.7 $\pm$ 1.0
Cho	1.0 $\pm$ 0.4	1.0 $\pm$ 0.2	1.2 $\pm$ 0.2	1.2 $\pm$ 0.4
mI	4.2 $\pm$ 1.4	4.0 $\pm$ 0.7	4.3 $\pm$ 0.9	4.2 $\pm$ 0.9
	Superior Temporal Gyrus			
tNAA	7.1 $\pm$ 1.7	7.1 $\pm$ 1.6	7.3 $\pm$ 1.0	7.2 $\pm$ 1.2
Glx	10.3 $\pm$ 2.1	10.8 $\pm$ 2.7	11.8 $\pm$ 2.2	11.7 $\pm$ 1.7
Cr+PCr	4.1 $\pm$ 1.0	4.2 $\pm$ 1.3	4.4 $\pm$ 1.0	4.5 $\pm$ 0.9
Cho	1.2 $\pm$ 0.2	1.1 $\pm$ 0.2	1.2 $\pm$ 0.3	1.3 $\pm$ 0.4
mI	3.5 $\pm$ 1.4	4.0 $\pm$ 1.0	4.0 $\pm$ 0.8	4.0 $\pm$ 1.0

Metabolite levels in Institutional Units (IU). No significant between-group differences were found.

Table 2

Spearman Correlations Between MRSI Metabolites and Formal Thought Disorder Score in the Childhood Onset Schizophrenia Group

Hemisphere	mI	Cho	Cr+PCr	Glx	tNAA
	Inferior Frontal Gyrus				
Left	0.72**	0.14	0.46	0.53	0.58*
Right	0.45	0.60*	0.51*	0.36	0.41
	Middle Frontal Gyrus				
Left	0.29	0.38	0.43	0.03	0.18
Right	0.08	0.57*	0.08	0.05	0.08
	Superior Temporal Gyrus				
Left	0.53	0.54	0.76**	0.60	0.53*
Right	0.66*	0.42	0.23	0.56	0.10

tNAA: *N*-acetyl-aspartate+ *N*-acetyl-aspartyl-glutamate, Glx: glutamate+glutamine, Cr: creatine+phosphocreatine, Cho: choline compounds, mI: *myo*-inositol. $p < *0.05$, $**0.01$. Significant correlations in **bold** are protected from multiple comparisons by repeated-measures MANOVA, other significant correlations not protected. All correlations partialled for age, correlations within the schizophrenia group also partialled for neuroleptic dose in chlorpromazine (CPZ) equivalents.