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Assessments of Function and Biochemistry of the Anterior Cingulate Cortex in Schizophrenia

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Abstract

Background—Neuroimaging and electrophysiological studies have consistently provided evidence of impairment in anterior cingulate cortex (ACC)/medial frontal cortex (MFC) function in people with schizophrenia. In this study, we sought to clarify the nature of this abnormality by combining proton magnetic resonance spectroscopy (^{1}H-MRS) with functional magnetic resonance imaging (fMRI) at 3T.

Methods—We used single-voxel MRS acquired in the dorsal ACC and fMRI during performance of a Stroop color-naming task to investigate the neurochemistry and functional response of the ACC/MFC in 26 stable, medicated, subjects with schizophrenia and 23 matched healthy controls.

Results—In schizophrenia subjects, we found decreased blood oxygen level-dependent (BOLD) signal in the medial frontal wall, with significant clusters restricted to more dorsal regions compared

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to healthy subjects. In addition, we observed a trend level decrease in *N*-acetylaspartate/creatine (NAA/Cr) levels, and a significant positive correlation between NAA/Cr level and the BOLD signal in schizophrenia subjects that did not exist in healthy subjects. Furthermore, in this group of medicated subjects, we did not find evidence of decreased glutamate + glutamine (Glx)/Cr levels, but there was a significant negative correlation between Glx/Cr levels and negative symptoms.

Conclusions—Our results suggest that abnormal NAA levels, which may reflect a neuronal dysfunction related to schizophrenia, affect neuronal physiology, as evidenced by reduced BOLD response.

Keywords

schizophrenia; ACC; fMRI; MRS; NAA; glutamate

INTRODUCTION

Neuroanatomical studies suggest that the anterior cingulate cortex (ACC) is composed of a number of different subregions with presumably different functional significance (1,2). Some human neuroimaging studies have identified the subcallosal ACC as being involved in emotional processing (3) or internal states (4) and the dorsal ACC (dACC) as being involved in mediating attention and executive functions, including error or conflict monitoring (5,6).

Functional magnetic resonance imaging (fMRI) (7-22) and electrophysiology (23,24) studies have provided consistent evidence of impairment in cingulate function in schizophrenia during cognitive processing.

Proton magnetic resonance spectroscopy (^{1}H-MRS) allows the *in vivo* measurement of several metabolites critically important for brain function, including *N*-acetylaspartate (NAA), an amino acid considered to be a marker of neuronal integrity (25-27), and glutamate (Glu), an amino acid involved in excitatory neurotransmission (28) and metabolism (29,30). While there have been consistent reports of reduced NAA, including in the ACC, in schizophrenia, both in first episode and chronic subjects (31-35), studies of Glu have been less common due to difficulty in quantifying this metabolite at low magnetic field strengths, and results have varied (35-40). However, there is preliminary evidence of increased ACC Glu levels in never (36) or minimally (40) treated subjects with schizophrenia and decreased ACC Glu levels in chronic, medicated subjects (37).

The purpose of this study was to investigate the relationship between function and neurochemistry of the ACC/medial frontal cortex (MFC) in subjects with schizophrenia and matched healthy controls using fMRI and MRS. This combined approach might offer clues regarding the biochemical interpretation of the BOLD signal abnormality. To activate the ACC during fMRI, we chose the Stroop color-naming task (41), which has been shown to produce robust activation during high conflict trials (3,42-44). We also acquired single-voxel ^{1}H-MRS measurements in the bilateral dorsal ACC in the area where we predicted significant activation during the Stroop task. Based on previous fMRI studies (9-14), we hypothesized that subjects with schizophrenia would show decreased BOLD response in the ACC/MFC compared to healthy controls. We further hypothesized that NAA levels would be reduced in schizophrenia, purportedly reflecting a disease-related neuronal dysfunction, and would correlate with neuronal physiology as evidence by reduced BOLD signal. Finally, we hypothesized that Glu levels would be decreased in this group of medicated subjects with schizophrenia.

METHODS AND MATERIALS

SUBJECTS

Twenty-eight subjects with schizophrenia and schizoaffective disorder (SZ) were recruited from the outpatient psychiatry clinic at The University of Alabama at Birmingham to participate in this study. Twenty-five healthy controls (HC), matched on age, gender, ethnicity, and parental occupation, were recruited by advertisement in flyers and the university's newspaper. Exclusion criteria were major medical conditions, substance abuse within six months of imaging, previous serious head injury, a neurological disorder, loss of consciousness for more than two minutes, and pregnancy. The study was approved by the Institutional Review Board of The University of Alabama at Birmingham, and all subjects gave written informed consent. Before signing consent, each SZ subject completed an Evaluation to Sign Consent Form.

Diagnoses were established using subjects' medical records and the Diagnostic Interview for Genetic Studies (DIGS) (45). General cognitive function of all subjects was characterized by the Repeatable Battery for the Assessment of Neuropsychological Status (RBANS) (46). The Brief Psychiatric Rating Scale (BPRS) (47) and its positive and negative subscales were used to assess mental status and symptom severity.

Several subjects were excluded from the analyses (incidental MR findings, image artifact, poor signal-to-noise ratio, excessive motion, and poor behavioral data). For the MRS data, two SZ and two HC were excluded, leaving twenty-six SZ and twenty-three HC in the analyses (Table 1). For the fMRI data, fourteen SZ and seven HC were excluded, leaving fourteen SZ and eighteen HC in the analyses (Table S1 in Supplement 1).

FUNCTIONAL TASK

Subjects performed a version of the Stroop color-naming task as described by Becker et al. (48). Stimuli were three words – 'RED', 'GREEN', or 'BLUE' – printed in one of those three colors. Incongruent trials were those in which the word and color were different. Subjects were instructed to indicate the color and ignore the word. They were instructed to respond both quickly and accurately. Responses were recorded by button press. The event-related design consisted of three runs of 88 trials each. The 3 s trials comprised a word stimulus for 1.5 s and a fixation cross for 1.5 s.

IMAGE ACQUISITION

All imaging was performed on a 3T head-only scanner (Magnetom Allegra, Siemens Medical Solutions, Erlangen, Germany), equipped with a circularly polarized transmit/receive head coil. fMRI data were acquired using the gradient recalled echo-planar imaging (EPI) sequence (TR/TE = 2100/30 ms, flip angle = 70°, FOV = 24 × 24 cm^2, 64 × 64 matrix, 4 mm slice thickness, 1 mm gap, 26 axial slices). A high-resolution structural scan was acquired for anatomical reference using the 3D T1-weighted MP-RAGE sequence (TR/TE/TI = 2300/3.93/1100 ms, flip angle = 12°, 256 × 256 matrix, 1 mm isotropic voxels). An IFIS-SA system (Invivo Corp., Orlando, FL) running E-Prime software (version 1.2; Psychology Software Tools, Inc., Pittsburgh, PA) was used to control stimulus delivery and to record responses and reaction times.

A series of sagittal, coronal, and axial T1-weighted anatomical scans serving as MRS-localizers were acquired for spectroscopic voxel placement. Slices were aligned to anatomical midline to control for head tilt. The MRS voxel was placed in a region of the bilateral dorsal ACC (Figure 1) on the basis of the sagittal and coronal images. Manual shimming was performed to optimize field homogeneity across the voxel, and chemical shift selective (CHESS) pulses

were used to suppress the water signal. Spectra were acquired using the point-resolved spectroscopy sequence (49) (PRESS; TR/TE = 2000/80 ms to optimize the Glu signal, number of averages = 256 (scanning time = 8 min 32 s), voxel size 2.7 × 2 × 1 cm^3).

STATISTICAL ANALYSES

Behavioral Measures—Three behavioral measures of reaction time (RT) were analyzed: the Stroop effect, post-conflict adjustment, and post-error slowing. RTs were analyzed in a current trial type by previous trial type by group analysis of variance (ANOVA) (48) in SPSS (version 12). The Stroop effect was defined as the difference in RT between incongruent trials and congruent trials. Post-conflict adjustment was defined as [(iC – cC) + (cI – iI)], where iC is a congruent trial preceded by an incongruent trial, etc. (12,48). Post-error slowing was calculated as the difference in RT of trials following an error trial versus after a correct trial. Total number of errors was also calculated. Post-error slowing and number of errors were compared across groups using one-way ANOVA. An alpha level of 0.05 was used for these statistical tests.

fMRI—Data analysis was implemented in SPM5. In order to obtain correlations between the BOLD signal and the MRS measurements, the functional analyses were performed in native space. The high-resolution structural scan was co-registered to the T1-weighted sagittal localizer images acquired for MRS voxel placement. Pre-processing of the fMRI data included slice timing correction, motion correction, reslicing to 2 mm isotropic voxels, co-registration to the structural scan, and smoothing with a 6 mm full width at half maximum (FWHM) Gaussian kernel. Subjects were excluded from further analyses if the motion parameters showed greater than 2 mm translation or 2° rotation within a run. Statistical analysis consisted of a single-subject voxel-by-voxel general linear model. Five conditions were included in the model: incongruent trials, congruent trials, error trials, no response trials, and stimulus repetitions, defined as any trial that was an exact repetition of the previous trial (12,48,50). Motion parameters were used as regressors. The conditions were convolved with the canonical hemodynamic response function with a temporal derivative. The contrast of interest was correct incongruent trials versus correct congruent trials.

Functional data were also analyzed after normalization to standard MNI (Montreal Neurological Institute) space to facilitate group comparisons. The medial wall region of interest (ROI) was defined using the WFU Pickatlas (version 2.4) (51,52) and the Automated Anatomical Labeling (AAL) atlas (53). It comprised the middle and anterior cingulate, the supplementary motor area, and the medial frontal wall. For these analyses, the statistical significance threshold was $p < 0.001$ with an extent threshold of 8 contiguous voxels (54) after small volume correction for the ROI. Whole-brain between-group differences were also examined at $p < 0.001$ uncorrected with at least 8 contiguous voxels.

MRS—MRS data were analyzed in jMRUI (version 3.0) (55). The residual water peak was removed using the Hankel-Lanczos singular values decomposition (HLSVD) filter (56). Spectra were quantified in the time domain using the AMARES (advanced method for accurate, robust, and efficient spectral fitting) algorithm (57). Prior knowledge derived from *in vitro* and *in vivo* metabolite spectra was included in the model. A phantom solution of 20 mM glutamate in buffer was imaged using the MRS parameters from the *in vivo* study. The resulting spectrum was quantified in jMRUI, and this model was used to fit the *in vivo* data. The model consisted of peaks for NAA, choline (Cho), creatine (Cr), and 3 peaks for Glu + glutamine (Glx), which correspond to the H-4 resonance of Glu. Amplitude, line width, and chemical shift were optimized for each peak. Cramer-Rao lower bounds (CRLB) (58-60) were calculated for each peak. Exclusion criteria were (1) line width of water greater than 25 Hz at FWHM during manual shimming, (2) CRLB greater than 30%, and (3) failure of the fitting algorithm. NAA,

Glx, and Cho were quantified with respect to Cr and compared across groups using one-way ANOVA with an alpha level of 0.05. To assess the reproducibility of the MRS measurements, one HC was scanned on five consecutive days, and the coefficient of variation was calculated for each metabolite ratio.

Functional and MRS analyses were also obtained after controlling for (1) the effect of medications, by using antipsychotic drug (APD) dose (expressed in chlorpromazine equivalent) as a covariate, and (2) substance abuse disorder, by contrasting SZ with (n = 4) and without (n = 10) a prior history of substance dependence. These analyses did not affect the results.

fMRI/MRS Native Space Correlations—The volume of interest from the MRS experiment was used as a region of interest in the fMRI native space analysis. For each subject, an ROI was created in MarsBaR (61) and MATLAB from the size, orientation, and coordinate information read from the individual's MRS raw data header. Mean percentage signal changes in this ROI for the incongruent and congruent conditions were extracted using MarsBaR. The relationships between metabolite levels, percent signal change, Stroop effect (RT), RBANS total index, and BPRS positive and negative subscales were analyzed by Pearson correlation with an alpha level of 0.05.

RESULTS

BEHAVIORAL

A current trial type by previous trial type by group ANOVA showed that the main effect of group was not significant, indicating that SZ subjects were not significantly slower across all trial types than HC subjects (Tables S2 and S3 in Supplement 1). The main effect of current trial type (the Stroop effect) was significant, indicating the mean RT was significantly faster for congruent trials than incongruent trials. The current trial type by group interaction was significant, indicating that the Stroop effect was greater in the HC group than the SZ group. The current trial type by previous trial type interaction (the post-conflict adjustment (48)) was not significant, and there was no group interaction. There was no significant difference in post-error RT between the groups [$F(1, 30) = 0.325$, $p = 0.573$]. SZ subjects had a significantly greater number of errors than HC subjects (SZ: 12.79 ± 10.7; HC: 5.44 ± 5.62) [$F(1, 30) = 6.282$, $p = 0.018$].

fMRI

Within and between-group ROI results are presented in Figure 2. HC subjects activated a large region of the medial wall, including the supplementary motor area and the medial frontal gyrus extending into the cingulate gyrus (Table 2). SZ subjects exhibited activation restricted to dorsal regions including the supplementary motor area in addition to the medial frontal gyrus and superior frontal gyrus.

Whole-brain between-group differences are shown in Figure S1 and Table S4 (see Supplement 1). SZ subjects exhibited reduced activity in the medial wall in regions of the right supplementary motor area, superior frontal gyrus, and cingulate gyrus in addition to the left medial frontal gyrus. In addition, SZ subjects showed reduced activity relative to HC in the left parahippocampal gyrus and right dorsolateral prefrontal cortex (DLPFC). Furthermore, SZ subjects had reduced activity in the caudate, ventrolateral prefrontal cortex, fusiform, thalamus, occipital, and parietal regions. SZ did not show greater activation compared to HC in any region at the defined threshold.

MRS

The coefficients of variation for NAA/Cr, Glx/Cr, and Cho/Cr were 4.0%, 4.5%, and 2.7%, respectively, for the one HC scanned on five days to assess measurement reproducibility (Figure 1).

MRS results for the HC and SZ groups are presented in Table 3. NAA/Cr was decreased in SZ relative to HC. The difference was not significant but approaching trend level (Figure 3). There were no significant group differences in Glx/Cr and Cho/Cr ratios. There was a significant correlation between NAA/Cr and Glx/Cr in HC ($r = 0.436$, $p = 0.038$) that was not observed in the SZ group ($r = 0.192$, $p = 0.347$). Comparison of the correlation coefficients showed no group difference ($z = 0.89$, $p = 0.374$).

MRS results for the HC and SZ included in the fMRI analyses are summarized in Table 4. Levels of NAA/Cr were decreased in SZ compared to HC; however, the difference did not reach significance. There were also no significant group differences in levels of Glx/Cr and Cho/Cr. The SZ subjects showed a trend-level association between NAA/Cr and Glx/Cr ($r = 0.469$, $p = 0.090$) that was not observed in the HC group ($r = 0.195$, $p = 0.437$) (between-group comparison: $z = 0.78$, $p = 0.435$).

fMRI/MRS NATIVE SPACE CORRELATIONS

There was a trend-level difference in the percent BOLD signal change for the contrast incongruent versus congruent in each subject's MRS voxel ROI [$F(1, 30) = 2.428$, $p = 0.130$]. For SZ subjects, there was a significant positive correlation between the percent signal change and NAA/Cr ($r = 0.668$, $p = 0.009$) (Figure 4), which was not observed in the HC group ($r = -0.010$, $p = 0.969$) (between-group comparison: $z = 2.06$, $p = 0.039$). Similarly, the SZ group showed a positive correlation between percent signal change and Glx/Cr ($r = 0.540$, $p = 0.046$), that was not observed in the HC group ($r = 0.118$, $p = 0.641$) (between-group comparison: $z = 1.22$, $p = 0.223$). There was a significant negative correlation between the percent signal change and Cho/Cr in the SZ group ($r = -0.657$, $p = 0.011$) that was not observed in the HC group ($r = 0.099$, $p = 0.695$) (between-group comparison: $z = -2.23$, $p = 0.026$).

fMRI/MRS BEHAVIORAL CORRELATIONS

HC subjects showed a positive correlation between the Stroop effect (RT) and the percent signal change ($r = 0.463$, $p = 0.053$). Analysis of the Stroop effect in the SZ group revealed one outlier. When this data point was removed from the correlations, there was a positive trend-level association between the signal change and Stroop effect in SZ ($r = 0.483$, $p = 0.095$) (between-group comparison: $z = 0.06$, $p = 0.952$). Percent signal change did not correlate with the RBANS total index in SZ or HC (all $p > 0.05$).

There were no significant correlations between MRS metabolite ratios and the Stroop effect (RT) or the RBANS total index (all $p > 0.05$).

fMRI/MRS BPRS CORRELATIONS

There were no significant correlations between the percent signal change and the BPRS subscales. In the SZ group, there was a significant negative correlation between Glx/Cr and the negative subscale of the BPRS ($r = -0.551$, $p = 0.005$). The BPRS negative subscale was positively correlated with Cho/Cr in SZ ($r = 0.517$, $p = 0.010$).

DISCUSSION

To our knowledge, the current study is the first to combine fMRI and ^{1}H-MRS to investigate the function and neurochemistry of the ACC/MFC in schizophrenia. We found decreased

BOLD signal in the medial frontal wall in subjects with schizophrenia, with significant clusters restricted to more dorsal regions compared to healthy controls. In addition, we observed a trend-level decrease in NAA/Cr levels in schizophrenia subjects and a significant positive correlation between NAA/Cr level and the BOLD signal in schizophrenia subjects that did not exist in controls. Furthermore, in this group of medicated schizophrenia subjects, we did not find evidence of decreased Glx/Cr levels, but there was a significant negative correlation between Glx/Cr and negative symptoms.

Consistent with previous fMRI studies (7-20,22), though not all (21), we observed decreased activation in the MFC in schizophrenia subjects. In general, studies that used the Multi-Source Interference Task, a task combining multiple sources of cognitive interference, have identified fewer differences between schizophrenia and healthy control groups (11,13). Like Heckers et al. (11), we found the locations of peak activation in the MFC within the schizophrenia group to be located dorsally to those identified in controls. In contrast, a topographic analysis of individual peak activations in MFC during interference failed to identify significant differences on measures of spatial distribution between subjects with schizophrenia and healthy controls (13).

In addition, consistent with recent meta-analyses (14,62), we found decreased activation in schizophrenia subjects in several regions consistently engaged across executive function tasks including in the DLPFC, the ventrolateral prefrontal cortex, the thalamus, and inferior/posterior cortical areas.

We observed a trend towards a decrease in NAA/Cr in the dACC in subjects with schizophrenia relative to healthy controls. These measurements were static and acquired independent of fMRI stimulation. While there have been several reports of decreased NAA in this region (31-33, 35), there are also reports of no difference (37,63-68), and even one report of increased ACC NAA (69). A meta-analysis of 64 ^{1}H-MRS studies in schizophrenia (34), found consistent evidence of reductions of NAA in frontal cortex, as well as a reduction in ACC NAA of about 4%, which is on the same order of our findings.

NAA is an amino acid synthesized in neuronal mitochondria, where it is thought to facilitate energy production from glutamate (30). In the cytosol, NAA acts as a precursor for the enzymatic synthesis of *N*-acetylaspartylglutamate (NAAG), which acts to regulate glutamate and dopamine release (70,71). NAA levels are reduced by inhibitors of the mitochondrial respiratory chain (25). Partial recovery of NAA levels have been reported in multiple sclerosis following treatment with various drugs (72-74). In non-human primates infected with the simian virus, reduced NAA levels were related to reductions in synaptophysin, a marker of synaptic integrity (26). Thus, NAA levels may reflect soma or synaptic integrity, the number of axon terminals and spines, and through its role in mitochondrial function, a measure of neuronal energetics.

There have been relatively fewer MRS studies of glutamate in schizophrenia. Based on two studies in chronic, medicated subjects, we had hypothesized that Glu levels would be decreased (37,39). However, we did not find significant differences in Glx/Cr between the groups, which is in agreement with the study of Wood et al. (35).

Glutamatergic abnormalities may point to disturbances in excitatory neurotransmission (28), altered flux through the tricarboxylic acid (TCA) cycle responsible for neuronal glutamate turnover (30), and/or altered flux through the cycling of glutamate from neurons to glia and back into the neurons after conversion to glutamine (29). One possibility, given that neuronal NAA and Glu metabolisms are linked, is reduced NAA levels without concomitant changes in Glx levels might point to abnormalities in soma size and/or density of dendrite/axon terminals rather than altered neuronal energetics.

The majority of postmortem studies of the ACC (75,76), but not all (77), have reported decreased cortical thickness in schizophrenia. While there is no evidence of cell loss (78,79), there is evidence of reduced neuronal density (77,80-82). Reports of decreased density of non-pyramidal neurons in layer II (77,81) suggest decreased interneuron GABA-ergic activity. There is also evidence of both decreased and increased tyrosine hydroxylate (TH) fibers on pyramidal neurons and interneurons, respectively (83). In addition, there are reports of reduced dendritic density (84,85), decreased expression of synaptic proteins (86-88) such as synaptophysin (89), and increased number of glutamatergic afferents entering layers II and III (90,91). Thus, there are a host of postmortem abnormalities that could affect neurometabolite levels in schizophrenia. Importantly, segmentation analysis of the *in vivo* data presented in this study revealed no differences in gray matter volume or tissue content fraction within the MRS voxel, excluding the possibility that reduced gray matter in schizophrenia subjects contributed to decreased NAA/Cr levels and activation.

We found a significant positive correlation between NAA/Cr level and the BOLD signal in schizophrenia subjects that did not exist in healthy controls, suggesting that this association is driven by schizophrenia-related neuronal pathology in the ACC. Importantly, this correlation was observed from data extracted from the exact same location. However, the placement of the MRS voxel was not functionally defined to optimally capture the interference-related BOLD signal. Others have sought similar fMRI/MRS relationships. Callicott et al. (92) reported a negative correlation between working memory task activation and DLPFC NAA in schizophrenia subjects only. Yucel et al. found reduced dACC NAA levels both in opiate addicts and obsessive-compulsive disorder (OCD) subjects. However, opiate addicts were comparable with controls in their dACC activation (93), and BOLD signal and NAA were not correlated, suggesting that NAA reductions were unrelated to dACC physiological activity. In OCD subjects, dACC activation was increased compared to controls (94), and NAA levels and BOLD signal were inversely correlated, suggesting that excessive activation could reflect a compensatory response to a neuronal dysfunction (94). Interestingly, all the above studies are in agreement with respect to a lack of correlation between NAA and the BOLD signal in healthy controls. Taken together, these data may indicate that abnormal NAA levels in different psychiatric conditions reflect specific neuronal abnormalities that in turn differentially affect the physiological response of the dACC during interference tasks.

Several studies have found evidence of correlations between negative symptoms and NAA levels in the DLPFC (95-97), the thalamus (98), and the ACC (33,35,68). We did not find such a correlation. However, we found a positive and a negative correlation between negative symptoms and Cho/Cr and Glx/Cr levels, respectively. Two studies (96,99) failed to identify correlations between frontal glutamate and choline and negative symptoms. These results clearly need replication.

Our results should be considered in view of the limitations of the study. In the current study, the NAA effect size was small, so in future studies a larger sample size will be needed. Moreover, we did not include a control region in MRS to test the specificity of the ACC findings. Furthermore, all subjects with schizophrenia were being treated with APD. Some studies have reported increases in NAA (100-102), in conjunction with APD treatment, while others have failed to observe such a change (103,104). In contrast to Bustillo (40), Theberge et al. (105) detected ACC glutamine reductions after prolonged APD treatment. While there is sufficient evidence to support decreased NAA levels in the ACC, it is possible that our negative findings for Glx levels represent a Type II statistical error. These results should be replicated with a larger and well-characterized group of subjects.

In summary, we show that in this group of chronic, medicated subjects with schizophrenia, decreased BOLD signal in the ACC is observed in the face of reduced NAA levels. Our results

suggest that abnormal NAA levels, which presumably reflect schizophrenia-related neuronal dysfunction, affect neuronal physiology as evidenced by reduced BOLD response. As the MR field moves towards higher field strengths, the relation between neuronal function and biochemistry might be a useful biomarker of illness as it could characterize, *in vivo*, key elements underlying the neuropathology of psychiatric conditions. Such biomarkers could provide a valuable tool to unravel the heterogeneity of schizophrenia or to assess novel treatment strategies, such as those targeting neuronal or spine plasticity.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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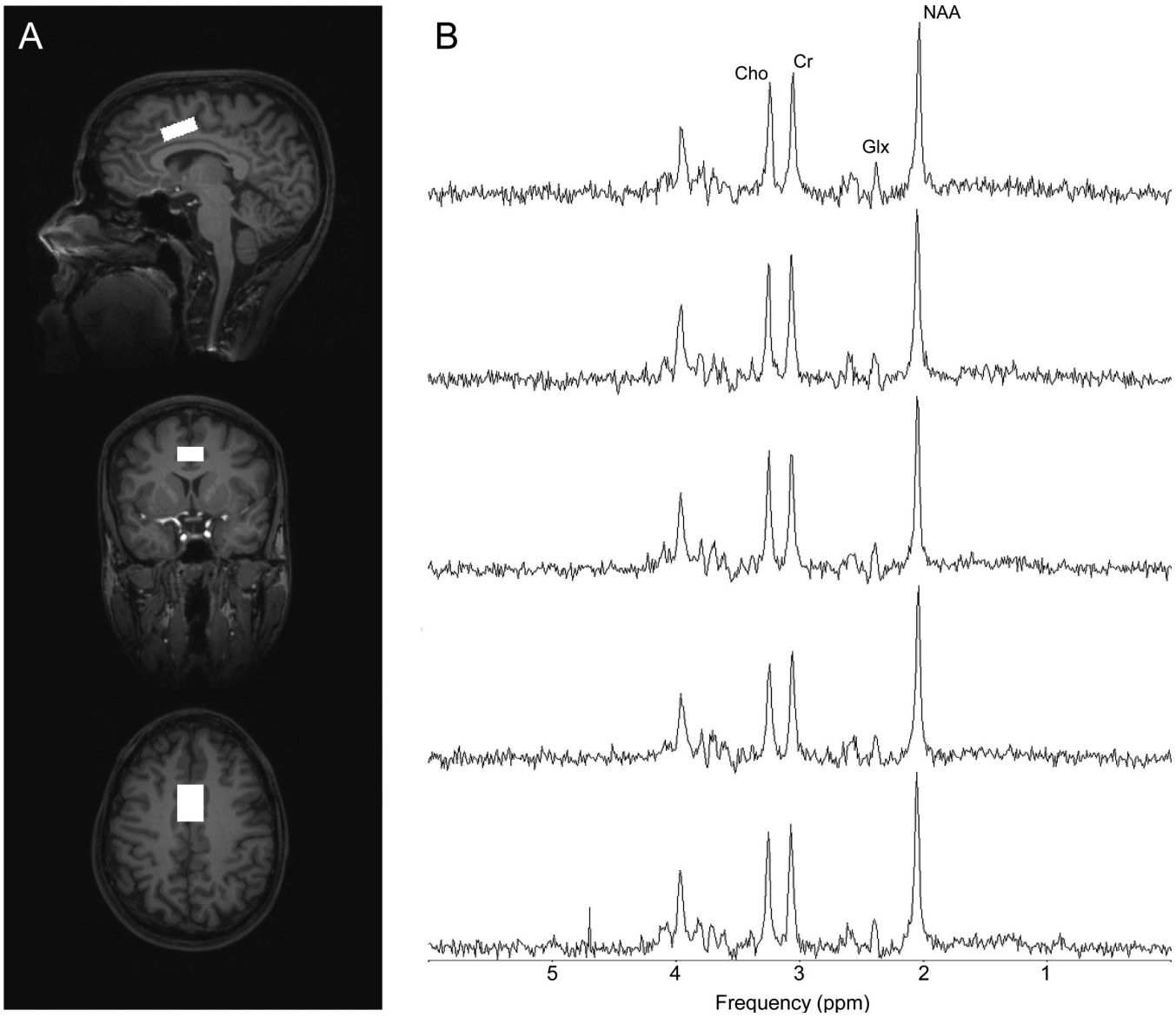

Figure 1.
(A) Example of MRS voxel placement in the bilateral dorsal ACC. (B) Spectra obtained from one healthy control scanned on five consecutive days to assess reproducibility of MRS data.

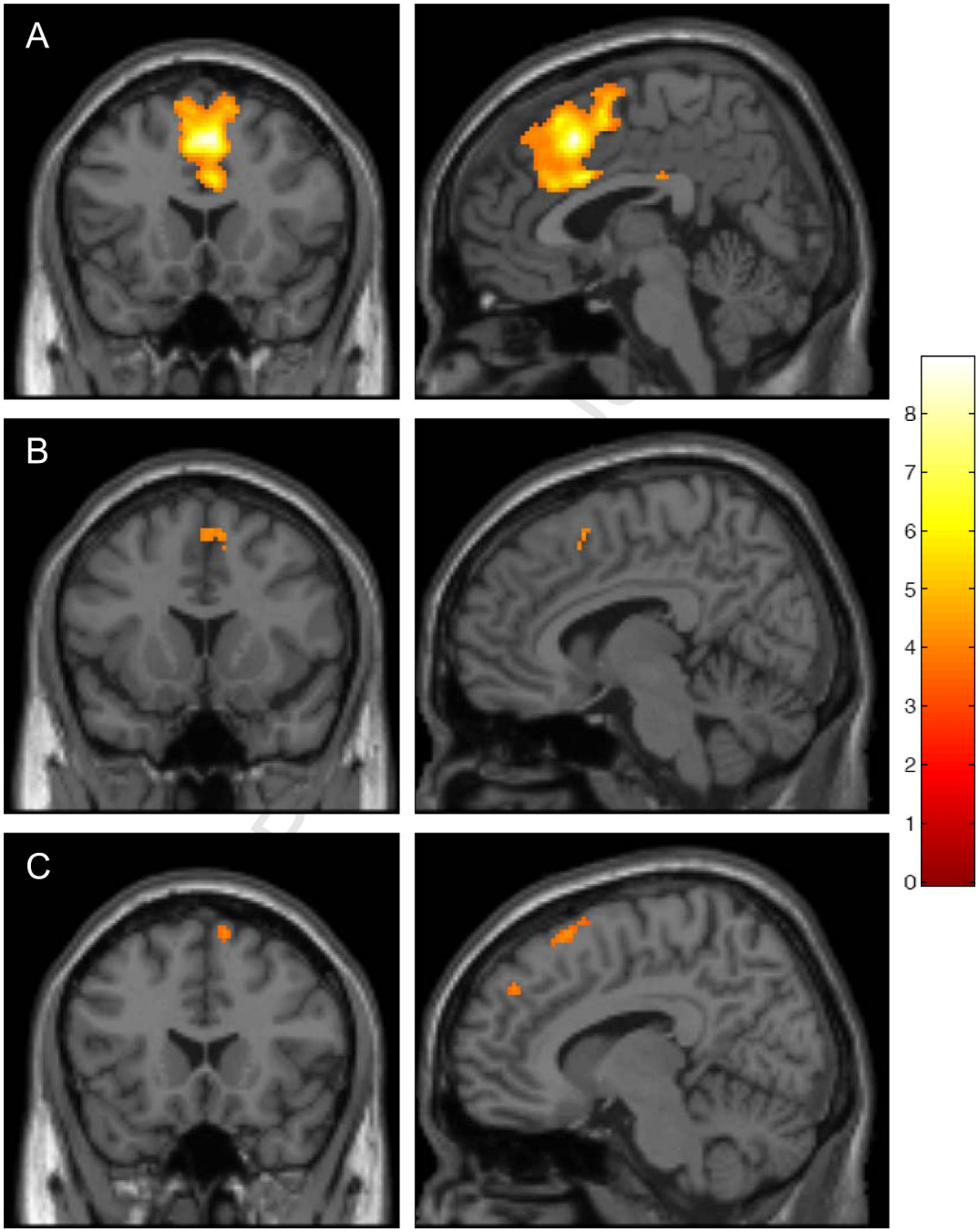

Figure 2.
Brain activation during the Stroop task (incongruent correct trials > congruent correct trials) in the medial frontal wall region of interest. (A) Healthy controls (n = 18). (B) Subjects with schizophrenia (n = 14). (C) Between-group differences (healthy controls > subjects with schizophrenia). $p < 0.001$ and at least 8 contiguous voxels with small volume correction for the medial frontal wall. Activation overlaid on the SPM5 T1 single-subject template. Left, coronal slices; right, sagittal slices. Color bar indicates t-values.

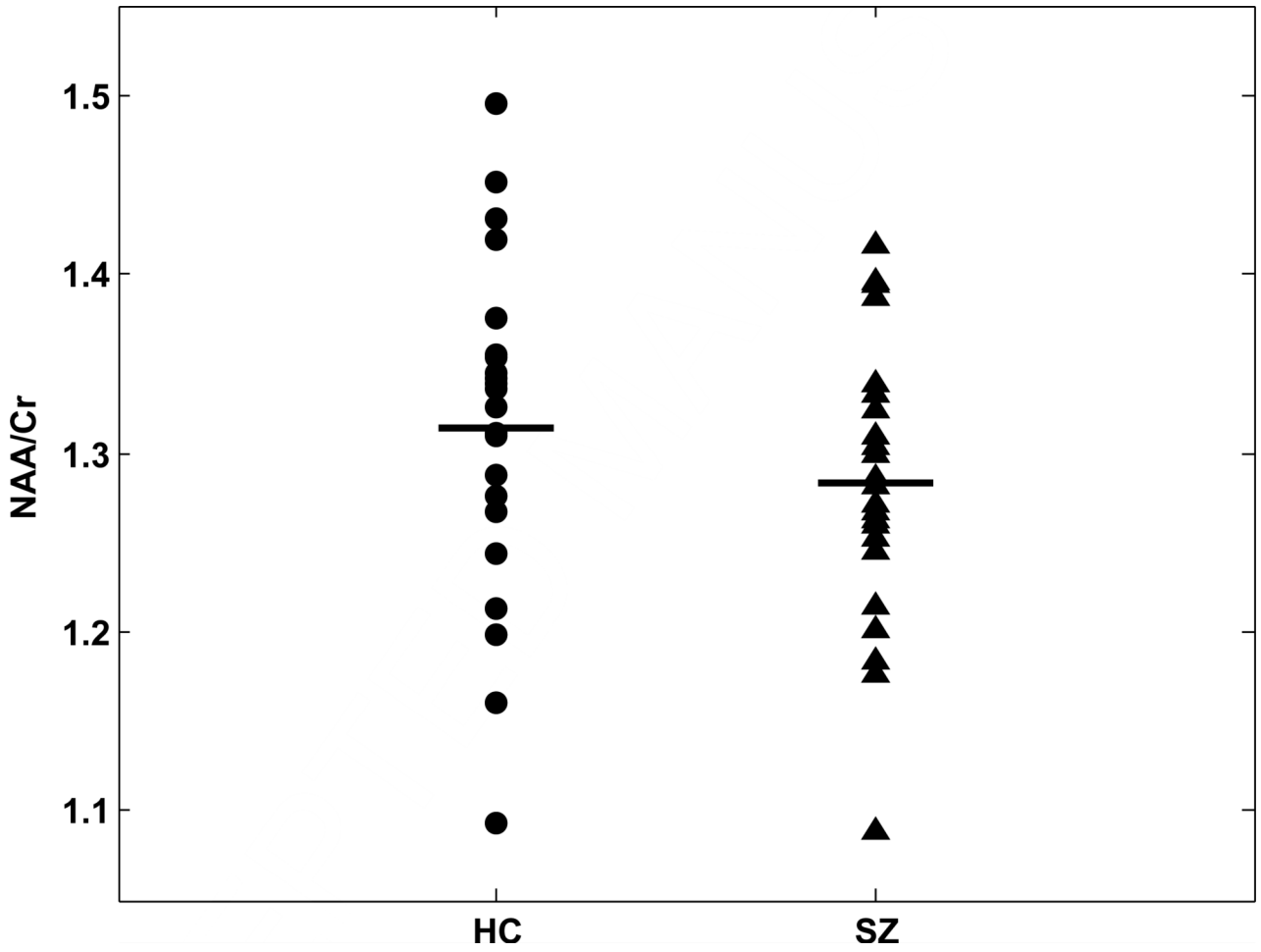

Figure 3.
Comparison of NAA/Cr levels in healthy controls (HC, n = 23) and subjects with schizophrenia (SZ, n = 26). Horizontal lines indicate means.

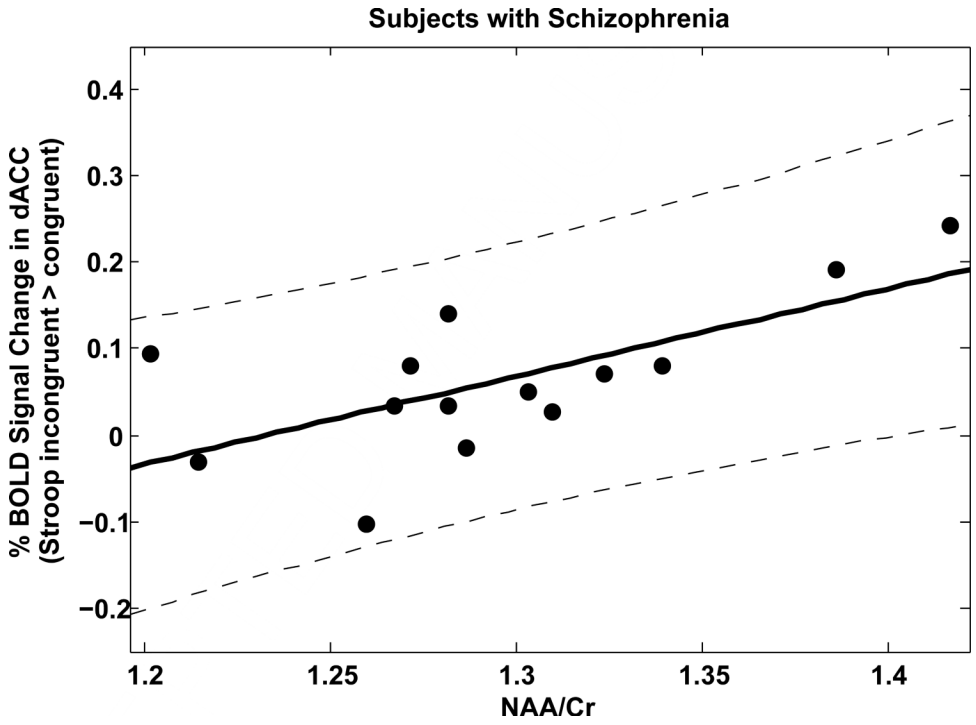

Figure 4.

Association between BOLD signal change (%) during the Stroop task (incongruent correct trials > congruent correct trials) and levels of NAA/Cr in the dorsal ACC (dACC) in subjects with schizophrenia (n = 14). Solid line is fitted linear regression, and dashed lines are 95% confidence intervals.

Table 1

Demographics and clinical measures[a]

Characteristic	SZ (n = 26)[f]	HC (n = 23)	t/χ^2	p-value
Age, years	40.4 ± 13.1	37.2 ± 12.4	0.88	0.39
Gender, M/F	18/8	15/8	0.09	0.77
Ethnicity, A/AA/C/H[b]	0/17/9/0	1/8/13/1	5.81	0.12
Parental occupation[c]	6.7 ± 5.1	6.8 ± 5.0	0.07	0.94
RBANS[d]				
Total index	72.9 ± 14.4	96.4 ± 11.1	6.16	< 0.001
Immediate memory	73.2 ± 13.9	98.0 ± 12.0	6.43	< 0.001
Visuospatial	85.8 ± 17.7	96.8 ± 14.1	2.31	0.03
Language	83.5 ± 14.7	95.6 ± 12.8	2.95	0.01
Attention	76.8 ± 20.9	101.1 ± 14.2	4.58	< 0.001
Delayed memory	72.6 ± 19.7	97.0 ± 8.9	5.38	< 0.001
BPRS[e]				
Total	35.1 ± 9.7	---	---	---
Positive	7.9 ± 4.2	---	---	---
Negative	4.2 ± 1.4	---	---	---

[a] Mean ± SD unless indicated otherwise; SZ, schizophrenia; HC, healthy control

[b] A, Asian; AA, African American; C, Caucasian; H, Hispanic

[c] Ranks determined from Diagnostic Interview for Genetic Studies (1 – 18 scale); higher rank (lower numerical value) corresponds to higher socioeconomic status; information not available for 2 SZ and 1HC

[d] Repeatable Battery for the Assessment of Neuropsychological Status; data not available for 4 SZ

[e] Brief Psychiatric Rating Scale (1 – 7 scale); positive (conceptual disorganization, hallucinatory behavior, and unusual thought content); negative (emotional withdrawal, motor retardation, and blunted affect); data not available for 2 SZ

[f] 21 SZ were treated with a second generation and 4 SZ with a first generation antipsychotic; information not available for 1 SZ

Table 2

Within-group activation in medial frontal wall during Stroop task performance[a]

Region	Hem	BA	x, y, z	Cluster size	t-value
SZ (n = 14)					
Supplementary motor area/medial frontal gyrus	L	6	-12, -6, 66	8	5.05
Supplementary motor area/superior frontal gyrus	R	6	8, 14, 56	37	4.36
HC (n = 18)					
Supplementary motor area/medial frontal gyrus	R	8	4, 18, 48	3098	8.91
Cingulate gyrus	L	23	-6, -22, 32	33	5.88
Medial frontal gyrus	L	9	-10, 48, 26	16	4.51

[a] $p < 0.001$ and at least 8 contiguous voxels after small volume correction for the medial frontal wall; SZ, schizophrenia; HC, healthy control; Hem, hemisphere; BA, Brodmann's Area; x, y, z refer to MNI coordinates

Table 3

Metabolite ratios in dorsal anterior cingulate cortex[a]

Metabolite	SZ (n = 26) Mean ± SD *CRLB*	HC (n = 23) Mean ± SD *CRLB*	F(1, 47)	p-value
NAA/Cr[b]	1.282 ± 0.074 *0.033*	1.315 ± 0.093 *0.031*	1.80	0.19
Glx/Cr	0.676 ± 0.068 *0.069*	0.674 ± 0.061 *0.060*	0.01	0.91
Cho/Cr	0.870 ± 0.102 *0.026*	0.840 ± 0.066 *0.024*	1.48	0.23

[a] SZ, schizophrenia; HC, healthy control; CRLB, Cramer-Rao Lower Bounds; NAA, *N*-acetylaspartate; Glx, glutamate and glutamine; Cho, choline; Cr, creatine

[b] There was no significant difference in the raw amplitudes of Cr between the groups [F(1, 47) = 2.788, p = 0.102].

Table 4

Metabolite ratios in dorsal anterior cingulate cortex (fMRI subgroup)[a]

Metabolite	SZ (n = 14) Mean ± SD *CRLB*	HC (n = 18) Mean ± SD *CRLB*	F(1, 30)	p-value
NAA/Cr[b]	1.296 ± 0.058 *0.033*	1.323 ± 0.087 *0.031*	0.99	0.33
Glx/Cr	0.672 ± 0.079 *0.067*	0.681 ± 0.057 *0.060*	0.13	0.72
Cho/Cr	0.842 ± 0.083 *0.026*	0.843 ± 0.073 *0.024*	0.001	0.97

[a] SZ, schizophrenia; HC, healthy control; CRLB, Cramer-Rao Lower Bounds; NAA, *N*-acetylaspartate; Glx, glutamate and glutamine; Cho, choline; Cr, creatine

[b] There was no significant difference in the raw amplitudes of Cr between the groups [F(1, 30) = 2.640, p = 0.115].