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Proton Magnetic Resonance Spectroscopy and Illness Stage in Schizophrenia – A Systematic Review and Meta-Analysis

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**Background**

It is not known whether regional brain N-acetyl aspartate (NAA) changes in the progression from prodrome to chronic schizophrenia. We used effect size meta-analysis to determine which brain regions show the most robust reductions in NAA first episode and chronic schizophrenia as measured by proton magnetic resonance spectroscopy (1H-MRS), and to determine whether these changes are present in individuals at high risk of developing schizophrenia.

**Methods**

We identified 131 papers of which 97 met inclusion criteria. Data were separated by stage of illness (at risk, first episode schizophrenia, chronic schizophrenia) and by brain region. For each region, mean and standard deviation of the NAA measure was extracted.

**Results**

Significant reductions in NAA levels were found in frontal lobe, temporal lobe and thalamus in both patient groups (effect size > 0.3; p<0.01). In individuals at high risk of schizophrenia (of whom approximately 20% would be expected to undergo transition to psychosis), significant NAA reductions were present in thalamus (effect size = 0.78; p<0.05), with reductions at trend level only in temporal lobe (effect size = 0.32; p<0.1), and no reductions in frontal lobe (effect size = 0.05; p=0.5).

**Conclusions**

These data suggest that schizophrenia is associated with loss of neuronal integrity in frontal and temporal cortices and in the thalamus, and suggest that these changes in the frontal and temporal lobe may occur in the transition between the at-risk phase and the first episode.
**Introduction**

Proton magnetic resonance spectroscopy (1H-MRS) studies in patients with schizophrenia have frequently reported regional reductions in N-acetyl aspartate (NAA), a brain metabolite present at very high concentrations in neurons in the CNS. Reductions in NAA levels measured by 1H-MRS are associated with ischemic brain injury and dementia, leading to the suggestion that NAA represents a surrogate marker of neuronal integrity (1). NAA also plays a role in myelin lipid synthesis, the control of osmolality, energy metabolism and neurotransmission (through the synthesis of NAAG by combination with glutamate) (1). An earlier meta-analysis, by Steen and colleagues, of frontal and medial temporal lobe 1H-MRS findings in patients with schizophrenia and first episode schizophrenia (FES) revealed that NAA levels were reduced in both regions, but that there was not any difference between FES and chronic schizophrenia (2).

Recent years have seen an explosion of interest in the prodrome of schizophrenia, with many groups studying individuals with attenuated psychotic symptoms and people at risk of schizophrenia due to their family history. Of course, only a proportion of these individuals eventually go on to develop schizophrenia (3). Nonetheless, several MRI studies have shown that people at risk of developing schizophrenia have alterations in gray matter volume compared to healthy controls (4-6), and that they have further reductions in gray matter volume associated with transition to psychosis (5).

Reductions in NAA reported in 1H-MRS studies of individuals at risk of schizophrenia suggest that these grey matter changes may have a neuronal basis (7),
perhaps representing the loss of dendritic arborization that has been reported in schizophrenia (8). It should be noted that not all 1H-MRS measures in people at risk of schizophrenia have been in agreement (9-10). Given the wide variance with 1H-MRS, and the requirement for relatively large sample sizes to obtain data with reasonable power (11), this is not altogether surprising.

Here, using a meta-analysis of the current literature, we test the hypothesis that regional reductions in NAA are associated with illness progression. We hypothesize that NAA reductions are most marked in patients with chronic schizophrenia, and are less evident in individuals with FES and in individuals at risk of schizophrenia.

**Methods**

**Selection of studies**

We selected all English-language studies published (or made available electronically) before 31st August 2009, of proton spectroscopy in patients with schizophrenia and individuals at high risk of schizophrenia compared to normal controls. We employed a search strategy using PubMed with keywords “MRS” and one of “schizophrenia”, “psychosis”, “UHR”, “ARMS”, “ultra high risk”, “prodromal”, “prodrome” or “schizoaffective”, obtaining hard-copies of all papers reporting NAA levels in patients with schizophrenia. We also obtained all papers referenced in two previous systematic reviews of the field (12-13). We selected all papers reporting mean and standard deviation for NAA in patients with schizophrenia, FES, and high risk of schizophrenia with a matched control group. Where mean and/or standard deviations for NAA were not reported, the authors of the papers were contacted. Efforts were made not to double-count subject data – papers in which this was suspected to have
occurred were excluded from the analysis.

**Methodological considerations**

Analysis of proton magnetic resonance spectroscopy data is complicated by the fact that the brain chemicals detected by 1H-MRS (termed metabolites) occur in grey and white matter but not in CSF. If the area from which the spectrum is acquired (voxel) contains a large percentage of CSF, the estimated metabolite concentrations measured using 1H-MRS will underestimate their actual concentration in brain tissue (14).

There are two methods commonly used to correct for this problem. The first is to report a ratio of the metabolite of interest to one of the other metabolites that are thought to remain relatively constant (either creatine - Cr, or less commonly choline - Cho). This has the potential drawback that if Cr or Cho levels are altered by illness state, the resultant ratio will be affected. The second method is to correct metabolite measures (either water-scaled or “absolute”) for the amount of CSF and brain tissue in the voxel by dividing the metabolite measure by the percent tissue volume in the voxel, sometimes with a further correction made for the relative grey and white matter content of the voxel (voxel brain tissue corrected) (14).

The fact that there are different methods for correcting 1H-MRS data for voxel brain tissue content makes combination of papers in a meta-analysis problematic. However, NAA results using scaled to Cr measures and voxel brain tissue corrected values were not found to differ significantly when analysed separately by Steen and colleagues (2). They combined data from studies using different methods by calculating the ratio of patient to control NAA levels (regardless of voxel tissue correction method). Here,
rather than calculate a ratio for each study, we performed a meta-analysis of effect size derived from each study, calculated from the absolute mean and SD values for patients and controls regardless of outcome method.

Proton magnetic resonance spectroscopy studies require selection of the voxel for spectrum acquisition at the time of scanning. Many studies in patients with schizophrenia and in individuals at high risk of schizophrenia acquire spectra from slightly different brain regions, leading to difficulties in comparing findings. Steen and colleagues combined 1H-MRS data acquired from all frontal and all temporal lobe regions into two main brain regions. Although this approach could mask findings that are specific to a particular subregion of frontal or temporal cortices, they found that regardless of subregion chosen, the NAA findings were similar within each overall region. We took a similar approach, combining NAA findings into seven regions – frontal lobe, temporal lobe, thalamus, cerebellum, parietal lobe, occipital lobe and basal ganglia.

**Identification of individuals at high risk of schizophrenia**

There are two current methods of identifying individuals at high risk of schizophrenia. The first is to identify teenage relatives of patients with schizophrenia (high genetic risk of schizophrenia), and the second is to select subjects on the basis of their clinical presentation – either having attenuated (subthreshold) psychotic symptoms, or a brief (less than 7 day) psychotic episode, which resolved without intervention (“BLIP”), so-called ultra high risk (UHR) patients (15). In the present study, we included both high risk groups.
Identifying individuals with First Episode Schizophrenia

Criteria for FES were not explicitly reported in any of the studies identified. Where authors reported the research subjects as being FES they were included as such for the purposes of the present analysis. We also included subjects reported as having schizophreniform disorder. Studies that included patients with non-schizophrenic psychosis were excluded from the analysis unless data from the patients with FES could be extracted.

Analysis

Data were separated by phase of illness (at risk of schizophrenia, FES, chronic schizophrenia), and by brain region (thalamus, basal ganglia, cerebellum, parietal lobe, occipital lobe, temporal lobe, frontal lobe). Where a study reported measures from more than one subregion (e.g. anterior cingulate cortex and DLPFC), these were entered into the appropriate meta-analysis (i.e. frontal lobe) as two (or more) independent datasets. Studies reporting metabolite values from different brain hemispheres, different subdivisions of patients (within each broad group of at risk, FES and chronic schizophrenia), or different outcome measures were treated in the same manner. Sample sizes for both patients and controls were adjusted to ensure that participants in such studies were not double-counted. This is an accepted strategy for handling such data (16).

We conducted a meta-analysis of effect size, using the meta (version 1.5-0) and metafor (version 1.1-0) packages for the R statistical programming language (17). A random effects model was used for the analyses, as it was considered likely that variation in subregion, hemisphere, and choice of outcome measure could potentially
introduce significant heterogeneity (18). The standardized mean difference (SMD) effect size (Hedge’s g) was calculated for each study and combined as a weighted mean by the inverse variance method.

Meta-regression was used to test the hypothesis that the degree of reduction in NAA levels relative to healthy controls would increase with illness phase from risk of developing psychosis to chronic schizophrenia. Where evidence for such a relationship was found, the timing of these NAA losses was examined using further meta-regressions.

**Publication bias**

Publication bias was investigated qualitatively using funnel plots for each group and brain region. Funnel plot asymmetry was assessed quantitatively by linear regression analysis (19) and the Duval and Tweedie trim and fill procedure (20).

**Analysis of heterogeneity**

Cochran Q and I² statistics were calculated in order to evaluate within study heterogeneity. Meta-regression of experimental variables was performed to investigate possible sources of heterogeneity, including effects of laterality, echo time, 1H-MRS acquisition technique (PRESS, STEAM), outcome measure (scaled to creatine vs. voxel brain tissue correction) and subregion. The influences of important demographic and clinical variables (age, age of onset, genetic vs. clinical at risk group, duration of medication and PANSS subscales) were investigated using meta-regression. It was not possible to compare NAA levels in patients treated with typical vs. atypical antipsychotics using meta-regression as most studies did not report
metabolite values for these treatment groups separately. Results were corrected for multiple comparisons using the Holm-Bonferroni method, in order to guard against type I errors (21).

**Outliers and sensitivity analysis**

Where significant heterogeneity was found the papers contributing to this were identified by visual inspection and cumulative meta-analysis. Outliers were examined in order to determine possible explanations for their results. We then repeated the meta-analyses with the outliers excluded, in order to assess the extent to which their findings were dependent upon the influence of such outliers.

To investigate the effect of our use of a random effects meta-analysis, we reanalyzed the data using fixed effects meta-analysis.

Lastly, to investigate the effect of treating multiple results from a single study as separate dataset, we repeated the meta-analyses using a single ‘composite’ measure from each study: where studies reported results from different subregions or using different outcome measures in the same paper, these were combined into a single value and analyzed using a random effects meta-analysis.

**Results**

**Study selection**

The literature search described above yielded 269 papers, of which 131 were potentially suitable for this study. Of these, 12 papers were excluded because of overlapping or suspected overlapping data with other papers from the same research
group. Eleven were excluded for lack of a control group. Six were excluded because mean and standard deviation of NAA levels were not reported and 5 were excluded as it was impossible to separate patients with schizophrenia from those with other diagnoses. A total of 97 papers met the inclusion criteria and were thus included in further analysis (7, 9-10, 22-115).

**NAA measures by region and illness phase**

**Frontal lobe**

In keeping with previous reports, NAA in frontal lobe was significantly reduced compared to controls in patients with chronic (fig. 1) and FES (fig. 2). No significant reduction in NAA was found in individuals at high risk of schizophrenia, whether due to a genetic or clinical risk (figure 3).

**Temporal lobe**

NAA in temporal lobe was reduced significantly in patients with chronic (fig. 4) and FES (fig. 5). In individuals at risk of developing schizophrenia, a reduction at trend level was found, although the effect size was comparable to that reported FES and chronic groups (fig. 6).

**Thalamus**

Significant NAA reductions in thalamus were present in patients with chronic schizophrenia (fig. 7) and in patients with first-episode schizophrenia (fig. 8). Only two studies measuring NAA in thalamus in individuals at high risk of schizophrenia have been published to date, but both showed a significant reduction compared to healthy controls (table 1).
Other regions

One group reported significant reductions in NAA in patients with chronic schizophrenia, FES, and in individuals with an at risk mental state in corpus callosum (113-114). There are no replications of these data by other groups at present.

Studies reporting NAA data in basal ganglia, cerebellum, parietal and occipital lobe have been published in patients with chronic schizophrenia and FES.

A trend for lower NAA in cerebellum compared to healthy controls was found in studies of first episode and chronic schizophrenia, becoming significant when the two groups were combined (table 1). There was no evidence for a reduction in basal ganglia NAA levels in patients with chronic or first-episode schizophrenia (table 1). A single study examining the basal ganglia in individuals at risk of developing schizophrenia has been published, although its results are non-significant (24). Pooled results of studies examining patients with both first-episode and chronic schizophrenia, either taken separately or combined, found no evidence for a reduction in NAA levels in occipital or parietal lobe (table 1).

Difference in NAA measures with stage of illness

A statistically significant difference in NAA levels with illness phase was found in frontal lobe only, with NAA reductions being present in FES patients but not in the at risk group (table 2). No significant differences in NAA levels were found in the patients with first episode compared to chronic schizophrenia.
Publication Bias

Funnel plots revealed no obvious signs of publication bias for any phase of illness or region (see supplemental materials eFigures 1-7). The linear regression test for funnel plot asymmetry confirmed this finding for all regions except frontal lobe, where a trend towards asymmetry was found for patients with chronic schizophrenia (Table 2). However, the trim and fill procedure estimated the number of missing studies to be zero. It was therefore considered unlikely that significant publication bias existed.

Heterogeneity

Significant heterogeneity was found in frontal lobe for all phases of illness. In temporal lobe significant heterogeneity was found for patients with first-episode schizophrenia and chronic schizophrenia but not for individuals at increased risk of developing schizophrenia (table 3).

Meta-regression

This meta-analysis included studies of two different categories of individuals considered to be at heightened risk of developing schizophrenia – those related to patients diagnosed with schizophrenia and those experiencing prodromal symptoms of schizophrenia. Meta-regression revealed no significant effect of these categories on NAA levels. Furthermore, no significant effect of hemisphere or variation in frontal lobe subregion was found for any patient group (Table 3), in line with previous findings (116). We therefore felt justified in our decision to include results from both hemispheres and from different frontal subregions in a single analysis. In temporal lobe an effect of outcome measure (ratio to Cr vs. voxel brain tissue correction) was found (uncorrected p<0.5) for both patients with FES and individuals at risk of
schizophrenia, with studies employing ratio to Cr reporting a greater effect size, although this was not statistically significant after correction for multiple comparisons (table 4).

One significant effect was found for chronic patients: that of negative symptoms (as measured by PANSS) on NAA levels in the temporal lobe (with lower NAA being associated with higher levels of negative symptoms). For first episode patients, in frontal lobe, significant effects of age of onset of illness (lower NAA with earlier onset) and duration of antipsychotic treatment (lower NAA with longer duration of treatment) on NAA levels were found (table 4). No effect was found for duration of illness on NAA levels, however. The complete results of the meta-regression are presented as supplemental material (eTable 2).

**Outliers and sensitivity analysis**

Although there was a high overall degree of consistency between findings, there were a number of outliers which we considered individually.

**Frontal Lobe**

The study by Wood and colleagues (10), emerged as a significant outlier in the analysis of the frontal lobe in individuals at risk of schizophrenia. The authors note that this increase in NAA/Cr ratio might be due to a reduction in Cr levels, representing hypometabolism, although other groups have not replicated this finding.

The study of FES patients by Yasukawa and colleagues was a significant outlier in frontal lobe NAA measures (89). This particular study reported results two patient
groups: one containing patients diagnosed with Gilbert’s syndrome (the outlying result); the other patients who did not have this condition.

In patients with chronic schizophrenia, Sanches and colleagues (71), reported a markedly greater effect size for NAA reduction in frontal lobe in patients, whilst Tebartz van Elst and colleagues, reported a marked increase in NAA in patients in this brain region (80). The reason for these discrepancies is not clear, but Sanches and colleagues employed a ratio to creatine plus choline for their measurement of NAA, a measure rarely used by other groups. The paper by Tebartz van Elst has been criticized for failing to employ adequate quality control of acquired magnetic resonance spectra for the reliable measurement of glutamate (117), and it is possible that this may have contributed to the difference in their reported data.

**Temporal lobe**

The same group that studied NAA in FES patients with Gilbert’s syndrome in frontal lobe reported data from temporal lobe in a separate paper (33). This result was similarly the only outlier for this region and patient group.

For chronic patients, the only outlier in temporal lobe was the left hemisphere subgroup from Maier and colleagues (99). One possible explanation is that the patients and controls in this study were not well matched in terms of age; the patients being significantly older. Age-related reductions in hippocampal NAA levels have been reported in healthy subjects (118-119), and it seems plausible that these should also occur to at least the same degree in patients with schizophrenia. However, it is not clear why this would affect only the left hippocampus.
**Other regions**

In thalamus, one subgroup of Heimburg and colleagues (32), was identified as an outlier in the chronic group. It is likely that its outlier status is due to its diminutive size – only two patients.

**Removal of outliers**

Meta-analyses were repeated with outliers excluded. In most cases the result was a slight reduction in the magnitude of the effect size but not to a degree that alters the findings of this study (table 5). The exception to this is for first episode patients in temporal lobe. Here, the result of removing the outlying study was to reduce the effect size to trend level.

Repeating the analyses using a single composite measure for each study yielded no significant differences in outcomes for any region. Likewise, there were no significant differences in effect sizes when the analyses were carried out using a fixed effects model (table 5).

**Discussion**

This is the first meta-analysis of 1H-MRS data comparing regional findings in patients with chronic, first episode and genetic and clinical risk of schizophrenia. We found that there was little difference in NAA findings in FES and chronic schizophrenia. In both groups, reduced NAA levels compared to controls were found in frontal lobe, temporal lobe and thalamus. The degree of reduction did not differ significantly between patients with FES and chronic schizophrenia, suggesting a lack
of progression. In individuals at risk of schizophrenia, whether due to genetic risk or subthreshold clinical features of schizophrenia, reductions in NAA were not present in frontal lobe. As discussed above, many of these individuals would not go on to develop schizophrenia (3), and so results in this group are likely to be significantly diluted.

The most robust differences in NAA levels in individuals at risk of schizophrenia compared to controls were found in thalamus. Relative NAA reductions in temporal lobe were present only at trend level (although this may have been due to the lower power in the at risk group), and NAA levels in frontal lobe did not differ significantly from controls. Taken together, these findings suggest that NAA reductions in thalamus, and possibly in corpus callosum, may be a trait marker of schizophrenia risk, or, alternatively, may be the first regions to be affected by the illness.

As longitudinal data about which of the at risk individuals went on to develop a first episode of schizophrenia were not reported, it is not possible to determine the reason that reduced NAA levels were not present in frontal lobe, and were present at trend level only in temporal lobe. It is possible to speculate that NAA reductions in these regions might occur only with transition to psychosis, mirroring findings in gray matter change (5). Alternatively, reductions in NAA levels may already have occurred in these regions but only those individuals destined to develop psychosis.

Another possibility for differences in temporal lobe measures is suggested by the meta-regression. The magnitude of the NAA reduction in the temporal lobe for patients with FES was greater in studies using ratio to Cr as an outcome measure than
in those employing voxel brain tissue correction. The same pattern is seen for at-risk individuals. The reason for this finding is not clear, but it is possible temporal lobe creatine levels were increased in these groups. There is some evidence to support increased hippocampal activity as a possible mechanism in the early stages of schizophrenia (121-122), which could conceivably be associated with increased hippocampal creatine levels (a marker of cellular metabolism). Thus, significant reductions in temporal lobe NAA may arise only in the later stages of illness, with increased metabolism, as indicated by increased creatine levels, occurring earlier.

There is a need for well-designed longitudinal studies to answer this question.

This study provides the first evidence of a significant correlation between negative symptoms and temporal lobe NAA levels in patients with chronic schizophrenia. Correlations between severity of negative symptoms and NAA levels have been observed before, notably in frontal lobe (78, 88). Given the suggested close relationship between NAA and glutamatergic neurotransmission (123), the current finding fits with previous reports of negative symptoms correlating with measures of abnormal glutamatergic transmission in hippocampus and temporal lobe (124-125).

This study indicates that an earlier age of onset is associated with lower frontal NAA levels in individuals with FES, supporting the hypothesis that an earlier age of onset may be associated with a more severe illness and more marked anatomical changes (126-127). The study also indicates that antipsychotic exposure is associated with reduced frontal NAA levels in FES patients. This finding is in keeping with earlier studies which have reported correlations between duration of treatment with typical antipsychotic medication and low NAA levels in this region (particularly anterior
cingulate cortex), while atypical neuroleptics appear to be associated with less of a reduction and perhaps even an increase in NAA levels (56, 128-130). No significant effect was found for duration of illness, supporting the hypothesis that NAA reductions are not progressive in schizophrenia.

**Weaknesses**

There are a number of shortcomings and potential weaknesses of this study that we acknowledge. The combination of results from different sub-regions, into a single analysis may have obscured some findings. This was a relatively unavoidable problem, given that different research groups use different a-priori regions of interest. A previous study did not find any significant differences in NAA changes in sub-regions (12), and we attempted to address this issue by analyzing these data using meta-regression. We did not find any evidence of significant differences in findings between different frontal subregions, nor between left and right hemispheres, and so feel that the inclusion of these data in a single analysis was justified. We cannot, however, exclude the possibility that some laterality or subregional effects in NAA levels may exist in the three groups.

Similarly, by including studies reporting different outcome measures in a single analysis, we were potentially obscuring other subtle effects. While meta-regression did not uncover any systematic influence of outcome measure, a near-significant effect was found in temporal lobe for first episode patients. In these groups, significantly lower NAA compared to controls was found only with scaled to metabolite measures. It is possible that this reflects increased temporal lobe creatine levels. It is not known whether or to what extent alterations in creatine levels in other
studies have affected the results.

Conclusions
These data suggest that schizophrenia is associated with reductions in neuronal integrity in frontal and temporal lobe and in the thalamus. Preliminary findings suggest that the first regions to be affected by loss of NAA in psychotic illness may be the thalamus, and possibly the corpus callosum. Further studies examining longitudinal changes in NAA in individuals at risk of schizophrenia are required to investigate this hypothesis.

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93:23-32.
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117. Theberge J, Williamson KE, Aoyama N, Drost DJ, Manchanda R, Malla AK,


Figure 1: Forest plot (random effects meta-analysis) of frontal cortex NAA levels in patients with chronic schizophrenia compared to controls.
Figure 2: Forest plot (random effects meta-analysis) of frontal cortex NAA levels in patients with first episode psychosis compared to controls.
Figure 3: Forest plot (random effects meta-analysis) of frontal cortex NAA levels in individuals at high risk of psychosis compared to controls.

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Random effects model 0.05 [-0.33; 0.43] 100%
Figure 4: Forest plot (random effects meta-analysis) of temporal cortex NAA levels in patients with chronic schizophrenia compared to controls.
Figure 5: Forest plot (random effects meta-analysis) of temporal cortex NAA levels in patients with first episode psychosis compared to controls.
Figure 6: Forest plot (random effects meta-analysis) of temporal cortex NAA levels in individuals at high risk of psychosis compared to controls.
Figure 7: Forest plot (random effects meta-analysis) of thalamic NAA levels in patients with chronic schizophrenia compared to controls.
Figure 8: Forest plot (random effects meta-analysis) of thalamic NAA levels in patients with first episode psychosis compared to controls.
Table 1: Effect size by region for individuals at risk of psychosis (At risk), patients with first-episode schizophrenia (FES), chronic schizophrenia (Chronic), and for both patient groups combined (All pts). Studies reporting multiple regions or outcome measures were entered as separate datasets, with sample sizes adjusted where necessary to ensure that participants were not double-counted.

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<td>19</td>
<td>32</td>
<td>376</td>
<td>428</td>
<td>-0.45</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
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<td>Chronic</td>
<td>41</td>
<td>71</td>
<td>880</td>
<td>799</td>
<td>-0.45</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td></td>
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<td>61*</td>
<td>104*</td>
<td>1279</td>
<td>1256</td>
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<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Temporal</td>
<td>At risk</td>
<td>4</td>
<td>5</td>
<td>82</td>
<td>100</td>
<td>-0.38</td>
<td>0.0703</td>
</tr>
<tr>
<td></td>
<td>FES</td>
<td>11</td>
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<td>232</td>
<td>189</td>
<td>-0.53</td>
<td>0.0025</td>
</tr>
<tr>
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<td>Chronic</td>
<td>22</td>
<td>35</td>
<td>524</td>
<td>530</td>
<td>-0.60</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
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<td>33</td>
<td>54</td>
<td>756</td>
<td>719</td>
<td>-0.58</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
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<td>2</td>
<td>3</td>
<td>49</td>
<td>49</td>
<td>-0.72</td>
<td>0.0006</td>
</tr>
<tr>
<td></td>
<td>FES</td>
<td>5</td>
<td>8</td>
<td>102</td>
<td>88</td>
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<td>0.0203</td>
</tr>
<tr>
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<td>Chronic</td>
<td>12</td>
<td>20</td>
<td>291</td>
<td>255</td>
<td>-0.32</td>
<td>0.0041</td>
</tr>
<tr>
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<td>17</td>
<td>28</td>
<td>393</td>
<td>313</td>
<td>-0.34</td>
<td>0.0002</td>
</tr>
<tr>
<td>Basal Ganglia</td>
<td>FES</td>
<td>6</td>
<td>9</td>
<td>125</td>
<td>91</td>
<td>-0.09</td>
<td>0.599</td>
</tr>
<tr>
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<td>11</td>
<td>17</td>
<td>200</td>
<td>181</td>
<td>-0.07</td>
<td>0.4981</td>
</tr>
<tr>
<td></td>
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<td>17</td>
<td>26</td>
<td>325</td>
<td>272</td>
<td>-0.09</td>
<td>0.3195</td>
</tr>
<tr>
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<td>All pts</td>
<td>5</td>
<td>6</td>
<td>108</td>
<td>75</td>
<td>-0.50</td>
<td>0.0114</td>
</tr>
<tr>
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<td>All pts</td>
<td>7</td>
<td>11</td>
<td>127</td>
<td>132</td>
<td>0.06</td>
<td>0.6397</td>
</tr>
<tr>
<td>Parietal</td>
<td>All pts</td>
<td>5</td>
<td>10</td>
<td>92</td>
<td>83</td>
<td>-0.08</td>
<td>0.6233</td>
</tr>
</tbody>
</table>
Table 2: Differences in regional NAA between illness phase as determined using meta-regression (* p<0.05) for individuals at risk of psychosis (At risk), patients with first-episode schizophrenia (FES), chronic schizophrenia (Chronic).

<table>
<thead>
<tr>
<th>Region</th>
<th>All phases</th>
<th>At-risk v. FES</th>
<th>At-risk v. Chronic</th>
<th>FES v. Chronic</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>QM(df=1)</td>
<td>P-value</td>
<td>QM(df=1)</td>
<td>P-value</td>
</tr>
<tr>
<td>Frontal</td>
<td>4.0544</td>
<td>0.0441*</td>
<td>6.1078</td>
<td>0.0135*</td>
</tr>
<tr>
<td>Temporal</td>
<td>1.0072</td>
<td>0.3156</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Thalamus</td>
<td>1.7934</td>
<td>0.1805</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Basal Ganglia</td>
<td>1.1202</td>
<td>0.2899</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Table 3: Measures of heterogeneity (Cochran Q and $I^2$), and estimates of publication bias (* p<0.05) for individuals at risk of psychosis (At risk), patients with first-episode schizophrenia (FES), chronic schizophrenia (Chronic), and for both patient groups combined (All pts).

<table>
<thead>
<tr>
<th>Region</th>
<th>Phase</th>
<th>Q</th>
<th>df</th>
<th>Q P Value</th>
<th>$I^2$ (%)</th>
<th>Publication bias t(df)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frontal</td>
<td>At risk</td>
<td>50.71</td>
<td>16</td>
<td>&lt;0.0001</td>
<td>68.4</td>
<td>-0.868(15)</td>
</tr>
<tr>
<td></td>
<td>FES</td>
<td>60.76</td>
<td>31</td>
<td>0.0011</td>
<td>49</td>
<td>-0.531(30)</td>
</tr>
<tr>
<td></td>
<td>Chronic</td>
<td>209.76</td>
<td>70</td>
<td>&lt;0.0001</td>
<td>66.6</td>
<td>-1.836(69)*</td>
</tr>
<tr>
<td></td>
<td>All pts</td>
<td>271.26</td>
<td>103</td>
<td>&lt;0.0001</td>
<td>62</td>
<td>-1.709(102)*</td>
</tr>
<tr>
<td>Temporal</td>
<td>At risk</td>
<td>7.08</td>
<td>4</td>
<td>0.132</td>
<td>43.5</td>
<td>-0.558(3)</td>
</tr>
<tr>
<td></td>
<td>FES</td>
<td>48.11</td>
<td>18</td>
<td>0.0001</td>
<td>62.6</td>
<td>-0.381(17)</td>
</tr>
<tr>
<td></td>
<td>Chronic</td>
<td>110.73</td>
<td>34</td>
<td>&lt;0.0001</td>
<td>69.3</td>
<td>0.075(33)</td>
</tr>
<tr>
<td></td>
<td>All pts</td>
<td>159.39</td>
<td>53</td>
<td>&lt;0.0001</td>
<td>66.7</td>
<td>0.051(52)</td>
</tr>
<tr>
<td>Thalamus</td>
<td>At risk</td>
<td>1.83</td>
<td>2</td>
<td>0.3998</td>
<td>0</td>
<td>0.446(1)</td>
</tr>
<tr>
<td></td>
<td>FES</td>
<td>9.05</td>
<td>7</td>
<td>0.2491</td>
<td>22.7</td>
<td>-0.789(6)</td>
</tr>
<tr>
<td></td>
<td>Chronic</td>
<td>25.67</td>
<td>19</td>
<td>0.1398</td>
<td>26</td>
<td>0.801(18)</td>
</tr>
<tr>
<td></td>
<td>All pts</td>
<td>34.89</td>
<td>27</td>
<td>0.1418</td>
<td>22.6</td>
<td>0.405(26)</td>
</tr>
<tr>
<td>Basal Ganglia</td>
<td>FES</td>
<td>10.56</td>
<td>8</td>
<td>0.2281</td>
<td>24.2</td>
<td>1.089(7)</td>
</tr>
<tr>
<td></td>
<td>Chronic</td>
<td>13.58</td>
<td>16</td>
<td>0.6301</td>
<td>0</td>
<td>0.858(15)</td>
</tr>
<tr>
<td></td>
<td>All pts</td>
<td>24.18</td>
<td>25</td>
<td>0.509</td>
<td>0</td>
<td>1.305(24)</td>
</tr>
<tr>
<td>Cerebellum</td>
<td>All pts</td>
<td>7.72</td>
<td>5</td>
<td>0.1725</td>
<td>35.2</td>
<td>1.011(1)</td>
</tr>
<tr>
<td>Occipital</td>
<td>All pts</td>
<td>10.21</td>
<td>10</td>
<td>0.4223</td>
<td>2</td>
<td>1.939(4)</td>
</tr>
<tr>
<td>Parietal</td>
<td>All pts</td>
<td>2.83</td>
<td>9</td>
<td>0.9707</td>
<td>0</td>
<td>-0.560(4)</td>
</tr>
</tbody>
</table>
Table 4: Significant modifiers of NAA levels by region and illness phase, as determined using meta-regression. Significance (p) values are presented both as raw values and as values corrected for multiple comparisons (* corrected p<0.1; ** corrected p<0.05).

<table>
<thead>
<tr>
<th>region</th>
<th>phase</th>
<th>modifier</th>
<th>intercept</th>
<th>co-efficient</th>
<th>P-value</th>
<th>Adjusted P-value (BH)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frontal</td>
<td>FES</td>
<td>Treatment duration</td>
<td>-0.3261424</td>
<td>-0.656463833</td>
<td>0.000181</td>
<td>0.01269461 **</td>
</tr>
<tr>
<td>Frontal</td>
<td>FES</td>
<td>Age of onset</td>
<td>-4.3600038</td>
<td>0.175791899</td>
<td>0.001226</td>
<td>0.02942189 **</td>
</tr>
<tr>
<td>Frontal</td>
<td>FES</td>
<td>Overall symptoms (PANSS)</td>
<td>-2.7258645</td>
<td>0.028745427</td>
<td>0.004484</td>
<td>0.08070678 *</td>
</tr>
<tr>
<td>Frontal</td>
<td>Chr</td>
<td>Age of onset</td>
<td>-5.4310586</td>
<td>0.22006038</td>
<td>0.021902</td>
<td>0.19711422</td>
</tr>
<tr>
<td>Temporal</td>
<td>At risk</td>
<td>Outcome measure</td>
<td>0.6180435</td>
<td>-0.65117903</td>
<td>0.03494</td>
<td>0.23579182</td>
</tr>
<tr>
<td>Temporal</td>
<td>FES</td>
<td>Outcome measure</td>
<td>0.7288</td>
<td>-0.7386</td>
<td>0.0064</td>
<td>0.09216 *</td>
</tr>
<tr>
<td>Temporal</td>
<td>FES</td>
<td>Age</td>
<td>1.3257</td>
<td>-0.0701</td>
<td>0.012186</td>
<td>0.14622605</td>
</tr>
<tr>
<td>Temporal</td>
<td>Chr</td>
<td>Negative symptoms (PANSS)</td>
<td>3.1049593</td>
<td>-0.150365854</td>
<td>0.000353</td>
<td>0.01269461 **</td>
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<tr>
<td>Temporal</td>
<td>Chr</td>
<td>Overall symptoms (PANSS)</td>
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<td>0.020307</td>
<td>0.19711422</td>
</tr>
<tr>
<td>Temporal</td>
<td>Chr</td>
<td>Positive Symptoms (PANSS)</td>
<td>4.9573865</td>
<td>-0.359261152</td>
<td>0.030048</td>
<td>0.23579182</td>
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</tbody>
</table>
Table 5: Comparison of effect size by region and illness phase when combining all data from each paper into one composite measure (“combined”), when using a fixed effects meta-analysis (“fixed”) and when removing outliers (“outliers removed”)

<table>
<thead>
<tr>
<th>Region</th>
<th>Phase</th>
<th>Combined effect size</th>
<th>Combined effect size p-value</th>
<th>Fixed effect size</th>
<th>Fixed effect size p-value</th>
<th>Outliers removed effect size</th>
<th>Outliers removed effect size p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frontal</td>
<td>At risk</td>
<td>0.12</td>
<td>0.25</td>
<td>0.0495</td>
<td>0.7995</td>
<td>-0.0628</td>
<td>0.6587</td>
</tr>
<tr>
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<td>FES</td>
<td>-0.42</td>
<td>&lt; 0.0001</td>
<td>-0.4474</td>
<td>&lt; 0.0001</td>
<td>-0.3883</td>
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<tr>
<td></td>
<td>Chronic</td>
<td>-0.39</td>
<td>&lt; 0.0001</td>
<td>-0.4489</td>
<td>&lt; 0.0001</td>
<td>-0.4046</td>
<td>&lt; 0.0001</td>
</tr>
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<td>At risk</td>
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<td>0.02</td>
<td>-0.3816</td>
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<td>FES</td>
<td>-0.46</td>
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<td>-0.5325</td>
<td>0.0025</td>
<td>-0.253</td>
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<td>&lt; 0.0001</td>
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<td>-0.4948</td>
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<td>At risk</td>
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<td>0.00</td>
<td>-0.7198</td>
<td>0.0006</td>
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<tr>
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<td>0.01</td>
<td>-0.4048</td>
<td>0.0203</td>
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</tr>
<tr>
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<td>-0.3174</td>
<td>0.0041</td>
<td>-0.3468</td>
<td>0.0002</td>
</tr>
<tr>
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<td>FES</td>
<td>-0.11</td>
<td>0.45</td>
<td>-0.0884</td>
<td>0.599</td>
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</tr>
<tr>
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<td>0.50</td>
<td>-0.0718</td>
<td>0.4981</td>
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<td></td>
</tr>
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<td>-0.49</td>
<td>0.00</td>
<td>-0.4989</td>
<td>0.0114</td>
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</tr>
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<td>0.6397</td>
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