Compromised myelin integrity during psychosis with repair during remission in drug-responding schizophrenia

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Abstract

Functional connection among the information-processing (grey-matter) centres within the CNS are necessary for the coordinated processing of perception, affect, thought and behaviour. Myelinated neuronal bundles provide the links among such processing centres. Magnetic resonance diffusion tensor imaging (DTI) can assess the physical integrity of myelin. Using DTI, the authors assessed diffusivity (Dm) within whole brain in 14 controls and within 13 acutely psychotic, drug-free schizophrenics both before and after 28 d of antipsychotic drug treatment. Drug-responder schizophrenics (D-RS) (n = 8) were differentiated from poor responders (PR) (n = 5) according to previously defined criteria. Differences of Dm at both baseline and following treatment were assessed using Dm distributional analyses and Statistical Parametric Software (SPM2). Impaired physical integrity of myelin, demonstrated by an increase (overall p < 0.05) of Dm, was found in the D-RS patients, with multiple regions demonstrating p < 0.0005 patient-control differences. The pathological increase in Dm was reduced (p < 0.03) following treatment-associated reduction of psychotic symptoms by 84%. Dm of PR patients did not differ from controls at baseline or following subacute treatment. While the pathophysiology(ies) underlying psychosis in poorly responsive (PR) schizophrenics does not appear to be related to a disordered myelin, the findings are consistent with a partially reversible disorder of myelin integrity, and may underlie a dys-synchrony of information processing in a major subgroup of drug-responsive patients with schizophrenia. An antipsychotic drug-induced cascade may partially restore myelin integrity and functional connectivity concomitant with antipsychotic effects in such D-RS patients.

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Key words: Antipsychotic response, diffusion tensor imaging (DTI), diffusivity (Dm), myelin, schizophrenia.

Introduction

While there has been considerable focus on deficient grey-matter volumes (Gogtay et al., 2004; Mathalon et al., 2001) and altered cortical and subcortical metabolic activities (Carter et al., 2001; Davidson and Heinrichs, 2003; Hazlett et al., 2004) in patients with schizophrenia, recently an interest in a primary pathology of the structures which connect and integrate the activities of such information-processing regions has emerged. ‘Wiring’ within the brain, which is critical for coordination of subcortical to cortical, and intra-cortical information processing, is comprised of myelin-coated axonal projections which rapidly transmit information across reverberating circuitry. Intact circuitry facilitates the synchrony of focused attention, perceptual evaluation, recognition (with activation of memory banks), assessments of alternatives, decision making, and directing behaviour. Disruption of such synchrony may be responsible for inattention, cognitive slippage, perceptual...
misinterpretations, and distorted convictions characteristic of patients with schizophrenia.

**Connectivity in schizophrenia**

*Correlation* between metabolic activities at distant sites is a commonly used measure of functional connectivity among cortical regions and with subcortical centres (Josin and Liddle, 2001). Positron emission tomography (PET) studies have demonstrated such altered functional frontal-superior temporal (Frith et al., 1995) and frontal-cingulate (Spence et al., 2000) cortex connectivity, as well as altered prefrontal-striatal (Buchsbaum et al., 1998), prefrontal-hippocampal (Meyer-Lindenberg et al., 2005), and occipito-temporal connectivity to multiple regions (Kim et al., 2005). Disrupted correlations of metabolic activities between right and left frontal lobes, as well as between anterior and posterior cortical regions (Mallet et al., 1998) are also reported in patients with schizophrenia.

Functional magnetic resonance imaging (fMRI) studies have documented altered fronto-temporal (Lawrie et al., 2002) frontal-parietal (Schlosser et al., 2003a, 2005) frontal-cerebellar (Schlosser et al., 2003b; Whalley et al., 2005), cerebellar-thalamic and thalamo-cortical (Schlosser et al., 2003b) connectivity.

**Integrity of myelinated connections in schizophrenia from diffusion tensor imaging (DTI)**

Integrity of the myelinated fibres connecting such cortical and subcortical grey matter has been investigated further using magnetic resonance (MR) DTI. Compact cellular structures, such as myelinated axonal tracts, impede the motion of extracellular water (Dong et al., 2004). Within the central nervous system (CNS), myelination of axonal projections is associated with restriction of white-matter diffusivity ($D_m$) of water molecules and high anisotropy (directional movement of water molecules parallel to the direction of myelinated fibre bundles) (Huppi et al., 1998). Conversely, during demyelination processes, $D_m$ increases and anisotropy is reduced (Cassol et al., 2004; Filippi et al., 2001; Rose et al., 2000; Zelaya et al., 1999). The relative $D_m$ or anisotropy of myelinated structures (cerebral white matter) has been widely used as index of ‘integrity’ of the myelinated tracts interconnecting distant grey-matter information-processing centres.

Using DTI, reduced anisotropy has been reported in white matter of the prefrontal cortex of schizophrenic patients compared to controls (Buchsbaum et al., 1998). Reduction of fronto-temporal and fronto-parietal white-matter tract (uncinate and arcuate fasciculus) integrity has also been described (Burns et al., 2003). A similar decrease in anisotropy has been found within the cingulate fasciculus (Kubicki et al., 2003; Wang et al., 2004) and in the middle cerebral peduncle (Okugawa et al., 2005). Other reports suggest that such a reduction in anisotropy in schizophrenics is widespread (Ardekani et al., 2003; Lim et al., 1999).

In contrast to anisotropy, which assesses spatial orientation of fibre tracts, $D_m$ (trace) assesses mean free-diffusion of water in all directions. $D_m$ provides information concerning relative fibre and myelin density, and may afford higher sensitivity than anisotropy in the detection of oedematous states. Using MR scanners with 1.5 T magnets, the $D_m$ diffusion tensor increases are indicative of volumetric expansion primarily within extracellular compartments (Clark and LeBihan, 2000).

**White-matter volumes in schizophrenia**

Reports of white-matter volume deficits in schizophrenia are not fully consistent. While three recent studies have reported regional volumetric deficits in cerebral white matter (Kubicki et al., 2003; Paillere-Martino et al., 2001; Sigmudsson et al., 2001), many previous studies have not described white-matter volume deficits (Shenton et al., 2001).

Shifts in the apparent volume of cerebral white matter have been reported during the course of successful treatment of psychotic symptoms in patients with schizophrenia. Serial, within-subject assessments of white-matter volumes have demonstrated reduction of white-matter volume associated with psychosis remission; conversely, patients with further psychotic deterioration between scans showed expansion of white-matter volumes (Christensen et al., 2004). Such apparent plasticity of white-matter volume associated with state changes may reflect an increase of intracellular and/or extracellular volume associated with psychotic episodes.

**Myelin and myelin-producing oligodendrocytes in schizophrenia: electron microscopy**

Post-mortem electron-microscopy studies in schizophrenia have reported fragmented myelin, with inclusion bodies (vacuolization) between layers of myelin which encircle axonal projections. The myelin-producing oligodendrocytes themselves show swollen cytoplasm and organelles, ectopia of nuclei, clumping of heterochromatin, and accumulation of dense granular lipofuscin-like material in combination
with lipid droplets and vacuolization (Uranova et al., 2001). The appearance suggests not only disruption of myelin, but also of a mixed necrotic-apoptotic inflammatory process. Such oligodendrocyte and myelin pathology may be associated with a ‘functional-disconnect-syndrome’ (FDS) marked by altered velocity of transmission and consequent desynchronization of grey-matter information-processing centres. Such desynchronization may underlie information-processing deficits associated with psychosis.

Herein we report observations which are consistent with changes in myelination in white matter within a group of drug-responder schizophrenics (D-RS). We characterize between-group differences in diffusivity ($D_m$) at psychotic decompensation and at partial remission using two complementary approaches. First, we characterized the distribution of $D_m$ throughout the brain, based on histograms of $D_m$ over all voxels. While discarding spatial and anatomical information, between-group assessment of distributional differences in an anatomically unconstrained fashion avoids problems of multiple comparisons. Second, we employed conventional voxel-by-voxel statistical parametric mapping of $D_m$ measures at each point in the brain. This enabled us to identify regionally specific $D_m$ differences between normal controls and D-RS, and document regions of change of $D_m$ in D-RS associated with significant antipsychotic response.

Methods

Subjects

Thirteen patients with recent emergence/exacerbation of psychosis were admitted to in-patient services at the University of Louisville Hospital. Each had been free of antipsychotic medication for at least 2 months prior to admission. Previously treated patients had been non-compliant with antipsychotic medications. Based on a diagnostic assessment using the Comprehensive Assessment of Symptoms and History (CASH; Andreasen, 1985), each patient met DSM-IV criteria for schizophrenia. Exclusion criteria included a history of head injury, mental retardation, substance dependence, or medical/neurological impairment. All patients had negative toxicology screens at the time of admission to the study. Each of the 13 patients provided written informed consent in accordance with requirements of the University of Louisville IRB for serial DTI studies and treatment with an antipsychotic in hospital for a period of at least 4 wk.

Fourteen controls without history of mental or neurological disease were drawn from the university and surrounding communities. All denied previous history of substance dependence, and each had a negative toxicology screen at the time of the study. Each also provided written consent for serial DTI studies.

Schedule of assessments

At baseline for both controls and patients (during drug-free psychosis exacerbation), MR DTI was completed. Psychosis was quantitated, using the Schedule for Assessment of Positive Symptoms (SAPS; Andreasen, 1984) for both patients and controls. Following baseline studies, the first five patients began antipsychotic treatment with 4 mg/d risperidone. The subsequent eight study patients received double-blind either 7 mg/d haloperidol (plus one placebo for blind b.i.d. dosage) or 60 mg ziprasidone b.i.d. with meals, as determined by double-blind assignment to treatment group. All had weekly psychosis (SAPS) assessments in hospital for a period of 4 wk. Continued hospitalization ensured medication compliance and prevented use and potential effects of other psychoactive drugs between assessments. Four weeks into treatment, and following psychosis evaluation (SAPS), patients were again assessed with DTI. Controls underwent similar serial DTI assessments to verify reliability of methods.

Separation of subgroups of patients on the basis of response parameters

Following 4 wk of antipsychotic drug treatment, drug-responder patients were separated from poor-responder patients on the basis of previously described criteria based on non-normal distributions of latency to antipsychotic response following treatment initiation (Garver et al., 1988, 1999, 2000, 2003). Patients responding with either >60% reduction of baseline psychosis (SAPS) scores or reduction of total SAPS psychosis score to <10 over a period of 28 d of treatment were designated as ‘drug-responder schizophrenics’ (D-RS). Patients who failed to meet either response criteria were designated as ‘poor responders’ (PR).

MR DTI: mean $D_m$

All MRI studies were performed using a 1.5 T clinical scanner with a quadrature head coil (Siemens Medical Systems, Iselin, NJ, USA). The effective diffusion tensor of water was measured using echo-planar imaging
transformation was then applied to the D standard anatomical atlas space; the same spatial during the DTI scan was spatially normalized to a the non-diffusion-weighted image that was acquired for each subject, tissue segmentation and statistical analysis of all 3D brain images. For each subject, (Ashburner and Friston, 2000; Frackowiak et al., 2003; Friston et al., 1995, Worsley et al., 1996) was used for the first acquisition had zero diffusion weighting. The intensity of this image was used to normalize that of the other six acquisitions, which had diffusion-encoding gradients of equal magnitude (24 mT/m) applied along six non-collinear directions \( G = \{(1, 0, 1), (-1, 0, 1), (0, 1, 1), (0, 1, -1), (1, 1, 0), (-1, 1, 0)\}; \) the diffusion-encoding \( b \) value magnitude was 1000 s/mm\(^2\) in each direction. The total diffusion imaging scan time was 5.5 min. The effective diffusion tensor was calculated by matrix inversion (Basser et al., 1994). The magnitudes and directions of the three principal axes of the diffusion tensor were determined by calculating the eigenvalues and eigenvectors, respectively, of the effective diffusion tensor. This method ensures that all terms (including non-diagonal terms) of the diffusion tensor are extracted from the data. The measured diffusion tensor therefore is rotationally invariant, so that changes in head orientation within the MRI scanner do not generate errors in the measured diffusion tensor. \( D_m \) was calculated as one-third the trace of the diffusion tensor.

Statistical parametric mapping software (SPM2) (Ashburner and Friston, 2000; Frackowiak et al., 2003; Friston et al., 1995, Worsley et al., 1996) was used for spatial normalization, tissue segmentation and statistical analysis of all 3D brain images. For each subject, the non-diffusion-weighted image that was acquired during the DTI scan was spatially normalized to a standard anatomical atlas space; the same spatial transformation was then applied to the \( D_m \) images. SPM2 also was used for automated tissue segmentation of the non-diffusion weighted EPI images. A brain volume mask was generated from the tissue probability images as the connected region with grey-matter plus white-matter probabilities exceeding 90\%. Each subject’s 3D \( D_m \) image was multiplied by this brain volume mask, and subsequent histogram analyses were performed on the masked image. A histogram of the proportional distribution of each resulting \( D_m \) image was computed using 20 bins over the \( D_m \) range of \( 0 \text{--} 20 \times 10^{-4} \text{mm}^2/\text{s} \). Each histogram was then normalized by the total brain volume, yielding a dimensionless histogram of the proportion of \( D_m \) per bin which was independent of the total brain size (Figure 1). Inspection of the histograms of each of the 13 patients and 14 controls revealed a similar pattern of distribution, with \( D_m \) peaks within bins 8 or 9 \((8 \text{--} 9 \times 10^{-4} \text{mm}^2/\text{s})\) and with a skew of \( D_m \) distribution to the right, towards high diffusivity. The high diffusivity region \((10 \text{--} 20 \times 10^{-4} \text{mm}^2/\text{s})\), in which movement of water molecules was less restricted by compact myelin, was examined among D-RS, PR and normal control groups at baseline using one-way ANOVA. Reduction of the pathological high diffusivity of the D-RS subgroup, associated with anti-psychotic response, was assessed with paired \( t \) test. Reliability of quantitative assessments of the proportion of \( D_m \) within the high diffusivity region was examined using test--test reliability from paired scans of five controls over a period of 9.5 ± 5.5 d, which demonstrated a within-subject coefficient of variation (CV) of 0.032 ± 0.026 (3.2\%) within the high diffusivity region.

**Data analysis**

**Histogram analysis**

Analyses of the \( D_m \) histogram at baseline and at 4 wk of treatment, and of patients subgroups (D-RS and PR) were performed using SigmaStat 3.0 (SPSS Inc., Chicago, IL, USA). Data were assessed for normality of distribution and equality of variance. Failure to meet
Myelin integrity in schizophrenia

First appearance of frank psychotic symptoms) ranged from 1 yr to 36 yr (11.8 ± 11.9 yr). Normal controls (seven males and seven females) were drawn from university students and employees and were aged 28.9 ± 7.2 yr (Table 1).

D-RS and PR schizophrenics

Patients were dichotomized into two groups or ‘endophenotypes’ on the basis of previously described distributions of response (Garver et al., 1999, 2000, 2003). D-RS (n = 8) demonstrated a reduction of baseline psychosis scores by ≥ 60%, or a residual SAPS psychosis score < 10 during the course of a 28-d period of antipsychotic drug treatment. PR patients (n = 5) failed to meet either improvement criteria by the end of 28 d of similar treatment. Thus, D-RS patients demonstrated a SAPS reduction of 39.9 ± 19.4 (84.4 ± 15.3%), while PR patients had a more modest reduction of 22.4 ± 16.0 (34.6 ± 21.0%) during the 28 d of in-patient treatment on the research ward. SAPS scores at the end of the 4-wk treatment period were 7.1 ± 10.0 for D-RS and 35.6 ± 7.1 for PR patients.

Histogram of Dm in controls (n = 14)

Dm was quantitatively assessed within all brain voxels, fractionating the relative magnitude of Dm across a 20 bin histogram. Inspection of each resulting histogram of individual controls revealed that all peaked between 7–9 × 10^-4 mm^2/s. The mean proportion (or %) of Dm within each of the 1 × 10^-4 mm^2/s graduated bins is shown in Figure 1. The range from 3–10 × 10^-4 mm^2/s reflects relatively compact myelin-neuronal bundles. Freer movement of water molecules (1/Dm), reflecting less compact myelin, was represented in the skewed distribution of higher Dm bins from 10–20 × 10^-4 mm^2/s. In total, 25.2 ± 2.28 (s.d.) % of brain voxels of controls were contained within the high Dm region in the group of 14 normal controls.

Dm in schizophrenics (n = 13); D-RS and PR

Inspection of the individual histograms of schizophrenics also documented peak Dm distributions within the 7–9 × 10^-4 mm^2/s range. Yet, compared to controls, there was a significant shift to the right (towards higher Dm) in the entire cohort of 13 schizophrenics at drug-free baseline. For the schizophrenics, 29.0 ± 3.94 % of brain voxels (vs. 25.2 ± 2.3 % for controls) were found within the 10–20 × 10^-4 mm^2/s, high Dm region (Mann–Whitney rank sum test: t = 236.5, p = 0.009).

criteria for normality or equality of variance resulted in use of non-parametric analyses. ANOVA or Kruskal–Wallis ANOVA on ranks with post-hoc comparisons were used to compare the two response groups to one another and to controls. Unpaired t tests and Mann–Whitney U tests were used for treatment group comparisons. Paired t tests compared changes within patient groups associated with treatment.

Image analysis

Statistical parametric maps were constructed using SPM2, which tested for group differences at baseline and for within-group differences associated with treatment using unpaired or paired t tests, respectively. These are shown at an uncorrected threshold of p = 0.01. Inferences about these regionally specific differences are restricted to a maxima whose corrected p value was < 0.05. These corrected p values are adjusted for the search volume and implicit multiple comparison problem using random field theory.

Results

Subjects

Patients with schizophrenia (nine males and four females) were in psychotic exacerbation and had been antipsychotic drug-free for at least 2 months (drug naive or compliance failure). They were aged 33.7 ± 11.3 yr (mean ± s.d.). Illness duration (since the first appearance of frank psychotic symptoms) ranged from 1 yr to 36 yr (11.8 ± 11.9 yr). Normal controls (seven males and seven females) were drawn from university students and employees and were aged 28.9 ± 7.2 yr (Table 1).

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However, increased baseline $D_m$ in the schizophrenics could be accounted for primarily by an ‘endophenotype’ which, following 4 wk of antipsychotic treatment demonstrated clear antipsychotic response. At baseline, such D-RS ($n=8$) had 30.2 ± 3.2% of voxels within the high $D_m$ region ($10^{-4}$–20 $\times$ $10^{-4}$ mm$^2$/s), while PR patients ($n=5$) showed 27.0 ± 4.5% and controls ($n=14$), 25.2 ± 2.3% (Kruskal–Wallis one-way ANOVA on ranks: $H=9.609$, d.f. = 2, $p=0.008$ with D-RS vs. controls in Dunn’s pairwise multiple comparison: $Q=3.096$, $p<0.05$). (Insufficient power was present to assess...
Table 1. Demographics, psychosis and high diffusivity (Dm) in drug-responder schizophrenics (DRS), poor responder (PR) schizophrenics and controls

<table>
<thead>
<tr>
<th>Group (drug)</th>
<th>Age (yr)</th>
<th>Sex</th>
<th>Years since onset</th>
<th>SAPS</th>
<th>% high Dm (10–20 × 10−4 mm²/s)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Base Rx % change</td>
<td>Base Rx Δ with Rx</td>
</tr>
<tr>
<td>Controls (n = 14)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean ± s.d.</td>
<td>29 ± 7</td>
<td>7F, 7M</td>
<td>–</td>
<td>0 –</td>
<td>25.2 ± 2.3³</td>
</tr>
<tr>
<td>D-RS (n = 8)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Risperidone</td>
<td>24</td>
<td>M</td>
<td>4</td>
<td>23  2</td>
<td>25.3</td>
</tr>
<tr>
<td></td>
<td>30</td>
<td>M</td>
<td>4</td>
<td>68  5</td>
<td>31.4</td>
</tr>
<tr>
<td>Risperidone</td>
<td>24</td>
<td>M</td>
<td>1</td>
<td>14  6</td>
<td>29.3</td>
</tr>
<tr>
<td>Risperidone</td>
<td>33</td>
<td>M</td>
<td>5</td>
<td>37  8</td>
<td>33.2</td>
</tr>
<tr>
<td>Ziprasidone</td>
<td>47</td>
<td>M</td>
<td>28</td>
<td>94  31</td>
<td>78.3</td>
</tr>
<tr>
<td>Ziprasidone</td>
<td>36</td>
<td>F</td>
<td>12</td>
<td>46  2</td>
<td>77.3</td>
</tr>
<tr>
<td>Haloperidol</td>
<td>56</td>
<td>F</td>
<td>36</td>
<td>43  3</td>
<td>78.3</td>
</tr>
<tr>
<td>Ziprasidone</td>
<td>27</td>
<td>F</td>
<td>2</td>
<td>47  0</td>
<td>78.3</td>
</tr>
<tr>
<td>Mean ± s.d.</td>
<td>35 ± 11</td>
<td>3F, 5M</td>
<td>12 ± 13</td>
<td>47 ± 25</td>
<td>7 ± 10</td>
</tr>
<tr>
<td>PR (n = 5)</td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Ziprasidone</td>
<td>31</td>
<td>M</td>
<td>8</td>
<td>45  45</td>
<td>78.3</td>
</tr>
<tr>
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<td>37</td>
<td>M</td>
<td>18</td>
<td>39  35</td>
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<tr>
<td>Haloperidol</td>
<td>50</td>
<td>F</td>
<td>28</td>
<td>42  36</td>
<td>78.3</td>
</tr>
<tr>
<td>Haloperidol</td>
<td>19</td>
<td>M</td>
<td>6</td>
<td>25  58</td>
<td>78.3</td>
</tr>
<tr>
<td>Haloperidol</td>
<td>24</td>
<td>M</td>
<td>1</td>
<td>37  46</td>
<td>78.3</td>
</tr>
<tr>
<td>Mean ± s.d.</td>
<td>32 ± 12</td>
<td>1F, 4M</td>
<td>12 ± 11</td>
<td>58 ± 18</td>
<td>36 ± 7</td>
</tr>
</tbody>
</table>

* D-RS significantly greater diffusivity at baseline than controls [Kruskal–Wallis one-way ANOVA on ranks (H = 9.61, d.f. = 2, p = 0.008) with pairwise multiple comparison (Dunn’s method; Q = 3.096, p < 0.05)].

* Significant reduction of excess diffusivity in good responders (paired t = 2.75, d.f. = 7, p = 0.028); poor responders showed a trend toward increased diffusivity during treatment (paired t = 2.06, d.f. = 4, p = 0.108); Dm change differences between patient groups between baseline and 28 d of treatment, t = 3.439, d.f. = 11, p = 0.006.

Potentially significant difference in high Dm between PR patients and either controls or D-RS patients at baseline.) Figure 2 demonstrates the shift to higher Dm of D-RS patients during the actively psychotic, initial drug-free baseline vs. controls.

Statistical parametric mapping found voxels with higher Dm in the eight D-RS patients, which were widely distributed throughout the brain, but appeared localized within the white matter and its boundary with grey matter. The t test maxima are reported in Table 2 in terms of their location in standard stereotactic space (Collins et al., 1994; Lancaster et al., 2000), the voxel’s t value and the adjusted p value. Regions with the most significantly increased Dm included the right pyramidal tract, left optical radiation and left superior temporal gyrus, although nearly every white-matter brain region showed an excess of Dm compared to controls (Figure 3a). There were no brain regions with lower Dm in the schizophrenia group, even when extremely loose thresholds were used for significance testing (uncorrected voxel-wise p < 0.05 and volume threshold >0.1 cc).

Partial restoration myelin integrity associated with drug treatment and psychosis reduction in D-RS patients (n = 8)

At 28 d of antipsychotic drug treatment, a significant but partial reversal of the pathological excess of Dm observed at baseline in D-RS patients was documented. The excess Dm found in D-RS patients within the 10–20 × 10−4 mm²/s region during baseline psychotic decompensation was reduced by 1.01 ± 0.04% [from 30.2 ± 3.2% to 29.2 ± 3.5% (paired t = 2.754, d.f. = 7, p = 0.028)] (Figure 4) as SAPS psychosis scores fell from 47 ± 25 to 7 ± 10 at the fourth week of treatment. Regional reduction of Dm, shown in Figure 3b, indicated a highly significant decrease of Dm in D-RS patients within the right pyramidal tract, left temporal lobe, and cingulate gyrus. Each of these regions had Dm excess at baseline (Figure 3a).
Table 2. Locations of $D_m$ differences/changes found by Statistical Parametric Mapping

<table>
<thead>
<tr>
<th>Brain region (Brodmann area or white matter)</th>
<th>Side</th>
<th>MNI coordinates $(x, y, z)$</th>
<th>Maxima $t$ value</th>
<th>Cluster $p$ value (corrected)</th>
<th>Cluster size $(cm^3)$</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Temporal lobe</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sup. temp. gyrus (BA 21)</td>
<td>L</td>
<td>$-52, -24, -2$</td>
<td>7.8</td>
<td>$&lt;0.0005$</td>
<td>3.8</td>
</tr>
<tr>
<td>Mid. temp. gyrus (WM)</td>
<td>L</td>
<td>$-50, -56, -10$</td>
<td>4.5</td>
<td>0.001</td>
<td>2.4</td>
</tr>
<tr>
<td>Angular gyrus (WM)</td>
<td>R</td>
<td>$-36, -74, 30$</td>
<td>5.9</td>
<td>0.002</td>
<td>2.2</td>
</tr>
<tr>
<td>Fusiform gyrus (WM)</td>
<td>R</td>
<td>$-38, -54, -16$</td>
<td>5.0</td>
<td>$&lt;0.0005$</td>
<td>7.5</td>
</tr>
<tr>
<td>Subgyral (WM) – optic radiation</td>
<td>L</td>
<td>$-40, -36, -2$</td>
<td>4.7</td>
<td>$&lt;0.0005$</td>
<td>3.9</td>
</tr>
<tr>
<td><strong>Parietal lobe</strong></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Precuneus (BA 7)</td>
<td>L</td>
<td>$-4, -72, 38$</td>
<td>5.4</td>
<td>$&lt;0.0005$</td>
<td>10.8</td>
</tr>
<tr>
<td>Precuneus (BA 7)</td>
<td>R</td>
<td>$18, -76, 36$</td>
<td>5.3</td>
<td>0.001</td>
<td>2.4</td>
</tr>
<tr>
<td>Inf. par. lobe (BA 40)</td>
<td>L</td>
<td>$-32, -34, 36$</td>
<td>4.9</td>
<td>0.001</td>
<td>2.5</td>
</tr>
<tr>
<td><strong>Occipital lobe</strong></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Lingual gyrus (WM)</td>
<td>L</td>
<td>$-20, -62, -6$</td>
<td>5.1</td>
<td>$&lt;0.0005$</td>
<td>3.2</td>
</tr>
<tr>
<td><strong>Subcortical</strong></td>
<td></td>
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<td></td>
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<td></td>
</tr>
<tr>
<td>Internal capsule (WM)</td>
<td>R</td>
<td>$28, -22, 16$</td>
<td>5.2</td>
<td>0.005</td>
<td>2.0</td>
</tr>
<tr>
<td>Cingulate gyrus (WM)</td>
<td>L</td>
<td>$-14, -20, 34$</td>
<td>4.5</td>
<td>$&lt;0.0005$</td>
<td>2.8</td>
</tr>
<tr>
<td><strong>Cerebellum</strong></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Posterior lobe</td>
<td>R</td>
<td>$12, -46, -36$</td>
<td>5.4</td>
<td>$&lt;0.0005$</td>
<td>18.2</td>
</tr>
<tr>
<td><strong>Brainstem</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Midbrain</td>
<td>L</td>
<td>$-12, -24, -4$</td>
<td>6.9</td>
<td>$&lt;0.0005$</td>
<td>1.6</td>
</tr>
</tbody>
</table>

$^a$SPM2 was used to perform $t$ tests on $D_m$ images with voxel-level $p$ threshold = 0.01, corrected cluster-level $p$ threshold = 0.01, volume threshold = 125 voxels (1 cm$^3$).

$^b$Brain region labels and approximate Brodmann areas were obtained using the Talairach Daemon (Collins et al., 1994).

$^c$The Montreal Neurological Institute stereotaxic brain atlas space (Lancaster et al., 2000).
Myelin integrity in schizophrenia

Figure 4. High diffusivity ($D_m$ of $10^{-20} \times 10^{-4} \text{mm}^2/\text{s}$) in controls and in drug-responder schizophrenia (D-RS) before and during successful antipsychotic treatment; contrasts with poor responders (PR). Distributions of $D_m$ in controls ($n = 14$), in D-RS patients ($n = 8$), and in PR patients ($n = 5$) at drug-free baseline and following 28 d of antipsychotic drug treatment (distributions and mean $\pm$ s.d.). At psychotic baseline, D-RS had a significant increase of voxels within the high $D_m$ region ($10^{-20} \times 10^{-4} \text{mm}^2/\text{s}$) compared to controls (Kruskal–Wallis one-way ANOVA on ranks: $H = 9.609$, d.f. = 2, $p = 0.008$ with D-RS vs. controls in Dunn’s pairwise multiple comparison: $Q = 3.096, p < 0.05$). The excess $D_m$ found in D-RS patients during baseline psychotic decompensation was reduced (paired $t = 2.754$, d.f. = 7, $p = 0.028$) as SAPS psychosis scores fell from 47.25 to 7.10 at 28 d treatment. PR patients showed no such excess $D_m$ at baseline, nor were there significant changes with antipsychotic drug treatment.

Mean $D_m$ in PR ($n = 5$): (a) comparisons with controls ($n = 14$) and (b) following antipsychotic drug treatment

In contrast to the pathological increase of $D_m$ in D-RS patients, PR patients during psychosis at drug-free baseline failed to demonstrate significant $D_m$ differences from controls within the $10^{-20} \times 10^{-4} \text{mm}^2/\text{s}$, high $D_m$ region, as noted above. Following 4 wk of hospital treatment (milieu and antipsychotic drug) and as SAPS psychosis scores fell more modestly from $58.0 \pm 18.0$ to $35.6 \pm 7.1$ (34.6 ± 21.0%), there was no reduction in $D_m$ within the $10^{-20} \times 10^{-4} \text{mm}^2/\text{s}$, high $D_m$ region [27.0 $\pm$ 4.56% to 27.5 $\pm$ 4.0% (paired $t = 2.062$, d.f. = 4, $p = 0.108$)]. PR patients remained statistically indistinguishable from controls following treatment (Mann–Whitney $t = 50.5$, $p = 0.459$).

Age, gender, duration of illness and pathological $D_m$

There was no age-effect of baseline $D_m$ in the 13 patients within the $10^{-20} \times 10^{-4} \text{mm}^2/\text{s}$, high $D_m$ region ($r_p = -0.233, p = 0.443$). Duration of illness did not differ between the D-RS and PR patients (11.5 $\pm$ 13.3 vs. 12.2 $\pm$ 10.8 yr; $t = 0.099$, d.f. = 11, $p = 0.923$). $D_m$ within the high $D_m$ region was not correlated with illness duration ($r_p = 0.289, p = 0.338$). Neither was illness duration associated with drug and response-associated changes in high $D_m$ ($r_p = -0.135, p = 0.661$). Nor was there indication that gender was associated with differences in high $D_m$ at baseline [nine males and four females: 29.2 $\pm$ 3.1 vs. 28.5 $\pm$ 6.0% ($t = 0.279$, d.f. = 11, $p = 0.785$)] or associated with change during treatment [0.6 $\pm$ 1.3 vs. 0.1 $\pm$ 0.9% ($t = 1.067$, d.f. = 11, $p = 0.305$)]. In the D-RS group itself, high $D_m$ was not related to gender at baseline [five males 30.0 $\pm$ 2.2% vs. three females 30.7 $\pm$ 5.1% ($t = 0.272$, d.f. = 6, $p = 0.705$)] or change with treatment [−1.1 $\pm$ 1.3 vs. −0.03 $\pm$ 0.4% ($t = 1.017$, d.f. = 6, $p = 0.348$)].

Discussion

The finding of pathological changes of myelin, with increased $D_m$ in a subgroup of schizophrenic patients, is consistent with recent reports of widespread reduced directionality (anisotropy) (Ardekani et al., 2003) and altered myelin water fraction (Flynn et al., 2003) associated with white matter in schizophrenics. Such changes in myelin are also consistent with functional disconnection of subcortical to cortical, and intra-cortical linkage of metabolic activities from both PET (Kim et al., 2005; Mallet et al., 1998; Meyer-Lindenberg et al., 2005) and fMRI blood-flow studies (Lawrie et al., 2002; Schlosser et al., 2003a,b, 2005; Whalley et al., 2005) in patients with schizophrenia.

Recent studies have focused upon underlying pathology of oligodendroglia, which repetitively wrap axons with their phospholipid sheaths to insulate and hasten the speed of signal conduction (Aboitiz et al., 1992). Such sheathed axons carry partially processed information in reverberating circuits from subcortical to cortical, across intra-cortical, and back to subcortical sites. Proper synchrony of the arrival of such information at post-synaptic sites is critical for information processing associated with perception, thought and action (Miller, 2000). Demyelinating diseases, which cause focal disruption of such myelinated networks and associated with dys-synchrony of reverberating networks, have been associated with emergence of psychotic symptoms (Hyde et al., 1992). Oligodendrocytes have themselves been shown to be exquisitely sensitive to neurotoxicity, especially that induced by hyper-glutamatergic states (McDonald et al., 1998) which has been...
suggested to be a component of stress (Moghaddam, 2002) and of psychotic decompensation (Keshavan, 1999).

Ultrastructural studies of oligodendrocytes from patients with schizophrenia have reported a paucity of oligodendrocytes (Orlovskaya et al., 2002). Residual oligodendrocytes frequently show degenerative changes, with swollen cytoplasm and organelles, ectopia of nuclei, and accumulation of lipofuscin-like material and lipid droplets (vacuolization). Oligodendrocyte projections, wrapping individual axonal projections, are themselves swollen, with vacuolization between layers of the encircling myelin (Uranova et al., 2001).

Vacuolization of cellular components in patients with schizophrenia is not limited to oligodendroglia and its myelinated processes. Mononuclear cells (lymphocytes and monocytes) within CSF of schizophrenics also show vacuolization. Such vaculated cytoplasm with convoluted nuclei and perinuclear halos of monocytic cells is characteristic of immune activation within the CNS (Nikkila et al., 2001).

We have previously provided evidence of immune activation within the CNS in many of these same D-RS patients. The pro-inflammatory cytokine IL-6 was significantly elevated (by a mean of 53%) in 23 neuroleptic-free D-RS contrasted to eight PR schizophrenia (Karlsson et al., 2003). Seven of the D-RS patients within the present cohort, who were dually assessed with DTI and CSF IL-6, also demonstrated a significant elevation of CSF IL-6 at baseline ($p < 0.05$) in comparison to controls. These findings suggest an association of the pathological increase in $D_m$ found in D-RS patients with an active inflammatory process in the CNS during psychosis exacerbation.

We have also reported that schizophrenic patients display an apparent volumetric reduction of total white-matter volume associated with psychosis reduction (Christensen et al., 2004). Repair of myelin during antipsychotic drug treatment, with reduction of $D_m$ and of extracellular oedema (Clark and LeBihan, 2000) may be associated with such reduction of white-matter volume.

It is unclear whether an inflammatory process within the CNS during psychosis exacerbation is primary, itself initiating pathology, or is secondary, a response to an ongoing degenerative process. Retroviral fragments have been reported in the brain and CSF of patients with schizophrenia (Karlsson et al., 2001). Such retroviral activation or, alternatively, a primary auto-immune process may underlie immune activation of the central immune systems, releasing pro-inflammatory cytokines. Such pro-inflammatory cytokines may themselves be activators of apoptosis/necrosis (Hu et al., 1997) seen in ultrastructural studies. Alternatively, excitotoxicity of oligodendroglia by excess glutamate might be responsible for the deteriorating picture reported in oligodendrocytes and myelin, with a consequent (secondary) immune response associated with a necrotic process.

The role of antipsychotic drugs in the partial restoration of physiological $D_m$ in D-RS is also unclear. The link between $D_2$ receptor blockade by antipsychotics and oligodendrocyte and myelin integrity has not been established. Neither has the link between $D_2$ receptor blockade and IL-6 or other pro-inflammatory cytokines been established within the CNS. It is clearly desirable to study the effects of antipsychotic drug treatment upon both $D_m$ and CSF IL-6 in a larger D-RS patient group. It is also important to determine whether second-generation antipsychotic drugs are more effective in reducing excess $D_m$ (and psychosis) than first-generation antipsychotics. Despite the apparent greater reduction of pathological $D_m$ during treatment by second-generation drugs, it should be noted that the random assignment of medications resulted in second-generation antipsychotics being given to patients with pathological excess of $D_m$; baseline $D_m$ in patients assigned to haloperidol was virtually identical to controls. Moreover, the present study was clearly not powered to detect differences among the three antipsychotic drugs.

The documented increase in $D_m$ in a subgroup of individuals with schizophrenia is interpreted herein as deriving from disruption of myelin integrity. However, if CSF infiltration into white matter is increased in D-RS patients, especially during the baseline pre-treatment state, some of the observed difference could be accounted for by partial volume averaging.

The limited group of PR patients, who did not show altered (increased) $D_m$, still presented with a similar range of psychotic symptoms. The underlying pathophysiology in such patients may be different, perhaps reflecting a neurodevelopmental subtype of schizophrenia, characterized by anomalies of neuronal migration, cellular location, and relatively stable but dysfunctional connections among information-processing centres (Garver et al., 1999).

The present study is supportive of a disconnect syndrome associated with physically compromised myelinated links among grey-matter centres during periods of psychosis. Such ‘disconnect’ may be present during psychotic exacerbation in a D-RS patient.
subgroup of patients with DSM-IV schizophrenia. The pathological increase of $D_m$ within myelinated structures is consistent with the presence of excess extracellular water (oedema). Such disturbances of myelin may be associated with immune activation within the central compartment during such periods of psychotic exacerbation. Antipsychotic response associated with risperidone or ziprasidone treatment appears to be associated with partial restoration of myelin integrity; similar potential effects of haloperidol could not be assessed in the present study owing to skewed (random) assignment of haloperidol to schizophrenics with a paucity of pathological $D_m$ at baseline. The mechanism(s) underlying such benefits of antipsychotic drugs on myelin and functional connectivity are still obscure.

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Statement of Interest

None.

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