Progressive striatal and hippocampal volume loss in initially antipsychotic-naive, first-episode schizophrenia patients treated with quetiapine: relationship to dose and symptoms

Bjørn H. Ebdrup1,2,3, Arnold Skimminge4, Hans Rasmussen1,2, Bodil Aggernaes1, Bob Oranje1,2, Henrik Lublin1,2, William Baare3,4 and Birte Glenthøj1,2
1 Centre for Neuropsychiatric Schizophrenia Research, CNSR, Psychiatric Centre Glostrup, Copenhagen University Hospital, Glostrup, Denmark
2 Centre for Clinical Intervention and Neuropsychiatric Schizophrenia Research, CINS, Psychiatric Center Glostrup, Copenhagen University Hospital, Glostrup, Denmark
3 Danish Research Centre for Magnetic Resonance, DRCMR, MR Department, Copenhagen University Hospital Hvidovre, Hvidovre, Denmark
4 Center for Integrated Molecular Brain Imaging, CIMBI, Copenhagen, Denmark

Abstract
First-generation antipsychotics have been associated with striatal volume increases. The effects of second-generation antipsychotics (SGAs) on the striatum are unclear. Moreover, SGAs may have neuroprotective effects on the hippocampus. Dose-dependent volumetric effects of individual SGAs have scarcely been investigated. Here we investigated structural brain changes in antipsychotic-naive, first-episode schizophrenia patients after 6 months treatment with the SGA, quetiapine. We have recently reported on baseline volume reductions in the caudate nucleus and hippocampus. Baseline and follow-up T1-weighted images (3 T) from 22 patients and 28 matched healthy controls were analysed using tensor-based morphometry. Non-parametric voxel-wise group comparisons were performed. Dose-dependent medication effects and associations with psychopathology were assessed. Patients had significant bilateral striatal and hippocampal loss over the 6-month treatment period. When compared to controls the striatal volume loss was most pronounced with low quetiapine doses and less apparent with high doses. Post-hoc analyses revealed that the striatal volume loss was most pronounced in the caudate and putamen, but not in accumbens. Conversely, hippocampal volume loss appeared more pronounced with high quetiapine doses than with low doses. Clinically, higher baseline positive symptoms were associated with more striatal and hippocampal loss over time. Although patients' ventricles did not change significantly, ventricular increases correlated with less improvement of negative symptoms. Progressive regional volume loss in quetiapine-treated, first-episode schizophrenia patients may be dose-dependent and clinically relevant. The mechanisms underlying progressive brain changes, specific antipsychotic compounds and clinical symptoms warrant further research.

Received 22 March 2010; Reviewed 15 May 2010; Revised 15 June 2010; Accepted 18 June 2010; First published online 12 August 2010

Key words: Brain imaging (MRI), hippocampus, psychopathology, schizophrenia, second-generation antipsychotic (SGA), striatum.

Introduction
Magnetic resonance imaging (MRI) studies have shown widespread progressive brain alterations during the course of schizophrenia. These brain changes seem to occur most dramatically in the early stages of the disease; however, it is still unclear to what extent specific regional changes are related to antipsychotic medication and psychopathology (Arango et al. 2008; Borgwardt et al. 2009; Hulshoff Pol & Kahn, 2008).

A growing body of evidence indicates that first-generation antipsychotic (FGA) compounds may induce striatal hypertrophy possibly linked to the blockade of the dopamine D2 receptors and the

Although second-generation antipsychotic (SGA) compounds also target the $D_2$ receptors, SGAs may not cause volume increases (Chakos et al. 1994; Dazzan et al. 2005; Smieskova et al. 2009) and they may even decrease striatal volumes (Crespo-Facorro et al. 2008). Notably, SGAs do not constitute a homogeneous category (Leucht et al. 2009) and both preclinical (Andersson et al. 2002; Lidow & Goldman-Rakic, 1997; Tarazi et al. 2001) and clinical (Crespo-Facorro et al. 2008; Markianos et al. 2001) studies indicate differential effects depending on the specific SGA administered, emphasizing the importance of investigating individual antipsychotic compounds preferably in antipsychotic-naive patients.

Among SGAs, both clozapine and quetiapine have been shown to reverse volumetric increases caused by FGAs in striatum (Scheepers et al. 2001; Stip et al. 2008). Interestingly, both compounds are characterized by a low affinity and a loose binding (a fast $k_{cat}$), for dopamine $D_2$ receptors and a broad receptor profile (Kapur & Seeman, 2001; Kessler et al. 2006). However, clozapine is not recommended as a first choice in first-episode schizophrenia patients (Kerwin, 2007). The biochemical mechanisms underlying the antipsychotic effect of quetiapine are not fully understood. Some have argued that the effect may be mediated solely via a transiently high $D_2$ occupancy (Kapur et al. 2000; Tauscher-Wisniewski et al. 2002), while others have proposed that a weak $D_2$ antagonism in combination with a potent $5HT_2A$ antagonism underlie the effect (Meltzer et al. 2003; Meltzer & Huang, 2008). Interestingly, in a positron emission tomography (PET) study employing the $5HT_2A$ specific radioligand fluorine 18-labelled alanserin in a subset of the present cohort of first-episode schizophrenia patients treated with quetiapine, higher $5HT_2A$ occupancy was associated with a reduction in positive symptoms. Moreover, the optimal clinical effect associated with $5HT_2A$ occupancy was obtained with occupancies between 60% and 70%, corresponding to moderate doses of 336–538 mg/d (Rasmussen et al. 2010). These observations could suggest that in doses higher than $\sim 538$ mg/d (70% $5HT_2A$ occupancy) the antipsychotic effect of quetiapine may predominately be mediated through dopaminergic blockade. Until now, longitudinal studies investigating dose-dependent effects of antipsychotic monotherapy on striatal volumes are largely absent (for an exception see Glenthoj et al. 2007). Hypothetically, dose-dependent involvement of serotonergic and dopaminergic systems, respectively, may induce differential striatal alterations.

Investigations of the relationship between progressive striatal brain changes and symptom improvement have yielded inconsistent results. The aforementioned caudate volume reduction associated with clozapine treatment was significantly related to positive and general symptom improvement (Scheepers et al. 2001). In contrast, left-sided caudate and putamen volume increases have been associated with positive symptom improvement after 4 wk treatment with either FGAs or SGAs (Taylor et al. 2005).

Adminsitration of SGAs early in the course of schizophrenia may protect against progressive global grey-matter (GM) loss and ventricular enlargement which in turn may improve symptoms and the functional outcome (Hulshoff Pol & Kahn, 2008; Lieberman et al. 2008). The hippocampus, in particular, is a plastic region which is highly susceptible to neuropsychiatric stress (Geuze et al. 2005). Both in chronic as well as in first-episode schizophrenia patients hippocampal volume reductions have been consistently found (Shenton et al. 2001; Steen et al. 2006; Vita et al. 2006). This has prompted the question as to whether antipsychotic compounds, and in particular SGAs, may protect against hippocampal volume loss (Angelucci et al. 2000; Bai et al. 2003; Halim et al. 2004). In preclinical studies quetiapine has been shown to increase brain-derived neurotrophic factor (BDNF) expression (Park et al. 2006; Xu et al. 2002), to reduce neurodegeneration induced by global cerebral ischaemia (Bi et al. 2009; Yan et al. 2007), and to reverse stress-induced suppression of neurogenesis (Luo et al. 2005) in hippocampus.

The present study encompasses follow-up MRI data on a cohort of antipsychotic-naive, first-episode schizophrenia patients after 6 months treatment with quetiapine. Twenty-two patients and 28 matched healthy control subjects (HC) participated. At baseline the patients ($n = 38$) compared to HC ($n = 43$) displayed $a$-priori hypothesized volume reductions in the caudate nucleus and the hippocampus, albeit no enlargement of the ventricles (Ebdrup et al. 2010). In the current study, we used tensor-based morphometry (TBM) to explore striatal volume changes after $\sim$ 6 months of treatment. We explicitly tested if high quetiapine doses were associated with relative striatal volume increases compared to low doses. Moreover, we investigated whether quetiapine had a preserving effect on hippocampus and ventricle volumes. Finally, associations between structural changes and psycho-pathology were explored.
Method

The study was conducted in accordance with the Declaration of Helsinki II and approved by the Ethics Committee of the Capital Region, Copenhagen (H-KF-01-78/97). After complete description of the study to the subjects, written informed consent was obtained.

Participants

Twenty-two patients and 28 HC participated in the follow-up study. Demographic and clinical characteristics are given in Table 1. All participants were re-examined with MRI and psychopathology was reassessed with the Positive and Negative Syndrome Scale (PANSS; Kay et al. 1987) by trained raters (intraclass correlation = 0.92). After the baseline examinations, the patients were treated with quetiapine according to their clinical need (mean = 538; S.D. = 265 mg/d) in an intended treatment period of 6 months (mean = 7.3, S.D. = 1.0 months).

Baseline data on 38 patients and 43 age-, gender- and parental socioeconomic status- (P-SES) matched HC has been reported elsewhere (Ebdrup et al. 2010). Briefly, patients were recruited from the Capital Region of Copenhagen, Denmark. Inclusion criteria were: a diagnosis of schizophrenia, no prior exposure to antipsychotic medication, age between 18–45 yr, and no medical or neurological comorbidity. DSM-IV

<table>
<thead>
<tr>
<th>Time-point</th>
<th>Lo_quee (n = 13)</th>
<th>Hi_quee (n = 9)</th>
<th>Pt_all (n = 22)</th>
<th>HC (n = 28)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (yr)</td>
<td>B 26.2 (5.7)</td>
<td>27.8(5.1)</td>
<td>26.2 (5.4)</td>
<td>28.4 (6.0)</td>
</tr>
<tr>
<td>Gender (male/female)</td>
<td>– 8/5</td>
<td>7/2</td>
<td>15/7</td>
<td>21/7</td>
</tr>
<tr>
<td>Handedness (right/left)</td>
<td>– 13/0</td>
<td>8/1</td>
<td>21/1</td>
<td>26/2</td>
</tr>
<tr>
<td>P-SES (high/moderate/low)</td>
<td>– 7.6/0/0</td>
<td>2/5/2</td>
<td>9/11/2</td>
<td>19/7/27</td>
</tr>
<tr>
<td>MR scan interval (months)</td>
<td>– 7.3 (1.1)</td>
<td>7.2 (0.8)</td>
<td>7.3 (1.0)</td>
<td>7.7 (2.3)</td>
</tr>
<tr>
<td>Quetiapine (mean dose)</td>
<td>F 352.3 (103.3)</td>
<td>807.2 (178.3)</td>
<td>538.4 (265.7)</td>
<td>–</td>
</tr>
<tr>
<td>Benzodiazepines (yes/no)*</td>
<td>L 7/6</td>
<td>9/0</td>
<td>16/6</td>
<td>–</td>
</tr>
<tr>
<td>Antidepressants (yes/no)b</td>
<td>F 0/13</td>
<td>3/6</td>
<td>3/19</td>
<td>–</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>B 73.9 (21.0)</td>
<td>76.4 (10.3)</td>
<td>74.9 (17.1)</td>
<td>75.8 (9.6)</td>
</tr>
<tr>
<td>PANSS positive score</td>
<td>B 19.2 (3.0)</td>
<td>20.7 (5.3)</td>
<td>19.8 (4.0)</td>
<td>–</td>
</tr>
<tr>
<td>PANSS negative score</td>
<td>F 14.5 (3.7)</td>
<td>15.2 (4.6)</td>
<td>14.8 (4.0)</td>
<td>–</td>
</tr>
<tr>
<td>PANSS total score</td>
<td>B 21.9 (6.4)</td>
<td>21.1 (7.8)</td>
<td>21.6 (6.8)</td>
<td>–</td>
</tr>
<tr>
<td>Intracranial volume (ICV)</td>
<td>B 1509.3 (142.7)</td>
<td>1612.2 (240.0)</td>
<td>1555.4 (190.4)</td>
<td>1576.5 (105.9)</td>
</tr>
<tr>
<td>Total brain volume (TBV)</td>
<td>B 1226.1 (128.0)</td>
<td>1284.6 (128.0)</td>
<td>1254.9 (159.5)</td>
<td>1277.4 (100.8)</td>
</tr>
<tr>
<td>Total grey matter (GM)</td>
<td>B 753.8 (73.3)</td>
<td>779.4 (113.1)</td>
<td>764.3 (90.1)</td>
<td>778.1 (63.1)</td>
</tr>
<tr>
<td>Total white matter (WM)</td>
<td>B 472.2 (60.7)</td>
<td>517.1 (90.3)</td>
<td>490.6 (75.7)</td>
<td>499.3 (43.5)</td>
</tr>
</tbody>
</table>

Lo_quee, Patients treated with quetiapine <538 mg/d; Hi_quee, patients treated with quetiapine ≥538 mg/d; Pt_all, all patients; HC, healthy control subjects; B, Baseline; F, Follow-up; L, Lifetime; P-SES, parental socioeconomic status; PANSS, Positive and Negative Syndrome Scale.

Volumes of ICV, TBV, GM, WM and CSF are in cm³ (S.D.).

* Number of patients having any prescription of benzodiazepines.

* Number of patients in antidepressant treatment. At follow-up the three patients were treated with: selective serotonin re-uptake inhibitors (n = 2); noradrenergic and specific serotonergic antidepressant (n = 1).

* Number of patients with any lifetime diagnosis of substance abuse. None of the patients abused in the investigation period.

* DUI, Duration of untreated illness at baseline.
diagnoses were based on the Schedules for Clinical Assessment in Neuropsychiatry (SCAN) version 2.1 (Wing et al. 1990). Diagnoses were confirmed by regular clinical contacts during the treatment period. Benzodiazepines were allowed to reduce agitation and anxiety. Use of antidepressants was registered. None of the patients received anti-cholinergic medication. Duration of untreated illness (DUI) was defined as the time between the first unspecific symptoms related to psychosis and the date of the MRI scan. Symptoms had to be associated with a decline in a previous stable level of functioning. DUI data were collected as a best-estimate approach (Keshavan et al. 1998) with information from the SCAN interview, clinical records and relatives when possible. Of the 38 patients included at baseline, nine fulfilled lifetime DSM-IV criteria for substance abuse. At follow-up, 5/22 patients had a lifetime history of substance abuse, but none abused in the treatment period. Further, at follow-up all subjects had a negative urine screening for substance intake. Extrapyramidal side-effects (EPS) were evaluated with The Extrapyramidal Symptom Rating Scale (ESRS; Chouinard & Margolese, 2005).

HC were recruited from the community. Both patients and HC had normal physical and neurological examinations and MRI scans were without pathology.

Medication

As described in the Introduction, PET data from our group have indicated that the antipsychotic effect of quetiapine is associated with up to 70% 5-HT2A occupancy corresponding to ~538 mg/d (Rasmussen et al. 2010). We hypothesized that higher quetiapine doses act primarily through D2 blockade. The mean quetiapine dose in this study was 538 mg/d and was suitable for testing if high doses would be associated with relative striatal volume increases compared to low doses. Accordingly, to test for medication effects the patients were a-priori split into a high quetiapine (Hi_{que}) (≥538 mg) (n = 9) and a low quetiapine (Lo_{que}) (<538 mg) (n = 13) group. Cumulative quetiapine intake was also calculated (mean = 111.8 g, s.d. = 62.5), but since this correlated strongly with the mean dose (ρ = 0.97) only data on the mean dose are presented here.

Image acquisition and processing

Details of the MRI acquisition protocol and processing of the images have been presented elsewhere (Edbrup et al. 2010). Briefly, high-resolution 3D T1-weighted, sagittal, magnetization-prepared rapid gradient echo (MPRAGE) scans and 2D T2-weighted, axial, turbo-spin echo scans were acquired of the whole head at two time-points on a Siemens Magnetom Trio 3 T MR scanner (Siemens, Germany) with an eight-channel head coil. Images were gradient un-warped (Jovicich et al. 2006) and processed using the VBM5 toolbox (http://dbm.neuro.uni-jena.de/vbm/vbm5-forspm5/) in SPM5 (Wellcome Department of Cognitive Neurology, University College London, UK). T2-weighted images were used to create brain masks in native space. Brain-masked GM and white-matter (WM) tissue maps were used for high-dimensional inter-subject registration by means of DARTEL (‘diffeomorphic anatomical registration through exponentiated lie algebra’; Ashburner, 2007). The brain-masked GM, WM, and CSF images were warped into average image space (DARTEL space) and modulated with the Jacobian determinant (JD) of the applied deformation fields to correct for local volume changes following the inter-subject warping.

To determine volumetric changes from baseline to follow-up, follow-up bias-corrected and intensity-normalized T1 images in average image space were warped to their baseline counterparts using high-dimensional intra-subject warping (Ashburner et al. 2000). To evaluate volume changes over time the resulting intra-subject JD image was transformed with the natural logarithm and warped into DARTEL space. The volume of each tissue class at follow-up was calculated by the following formula:

\[\text{GM}_{\text{DARTEL}}^* (1 - \exp \{ \log (\text{JD}_{\text{intra-subject, DARTEL}}) \})\]

The volume change from baseline to follow-up was calculated by subtraction of baseline and follow-up images.

Regions of interest (ROIs)

ROI masks were created using the average of DARTEL-warped MPRAGE images of all subjects to test our specific hypotheses (Edbrup et al. 2010). Used delineation landmarks are referenced: striatum (comprised of masks of caudate nucleus, nucleus accumbens, putamen) (Glenthoj et al. 2007), hippocampus (Baare et al. 2001), ventricles (comprised of lateral and third ventricle) (Cahn et al. 2002).

Volumetric brain measures

Total brain volume (TBV) and intracranial volume (ICV) were estimated by integrating and adding image-intensity values of modulated and warped GM and WM (TBV), and CSF (ICV) images. ROI volumes were acquired to investigate whether local voxel-wise
changes were reflected in whole ROI volume changes. ROI volume estimates were derived by integrating image intensity values of modulated and warped images within the specific ROI (for details see Ebdrup et al. 2010). Global and regional volume estimates are given in Tables 1 and 3, respectively.

Statistical analyses

Statistical Package for the Social Sciences software (SPSS Inc., USA) was used to analyse demographic and volumetric data. The distribution of continuous data was tested for normality with the Shapiro–Wilk test. Age and volumetric changes in ROIs (caudate nucleus, nucleus accumbens, putamen, hippocampus, ventricles) were not normally distributed, thus group comparisons were performed non-parametrically with the Mann–Whitney U test. Handedness and gender differences were tested with Fisher’s exact test and socioeconomic status with Pearson’s χ² test. Potential outliers were identified with Grubbs’ outlier test (Grubbs, 1969). Differences in PANSS scores between patient groups were tested with the Student’s t test.

In the voxel-wise analyses we tested how groups differed on volumetric changes over time (baseline – follow-up) and in volumes at follow-up. We first tested for differences between all patients (Ptall) and HC. Subsequently, we tested for medication effects using the following contrasts: Hique vs. HC, Loque vs. HC, and Hique vs. Loque. TBV change served as a covariate. Since patients and HC did not differ regarding age, gender, and MR-scan interval analyses were initially performed without these covariates. Post-hoc, these variables were entered as covariates to assess their possible effects. When contrasting Hique vs. Loque groups, both TBV change and percentage weight change were entered as covariates, because patients significantly gained weight compared to HC. General linear models were estimated non-parametrically using randomize, part of FSL (http://www.fmrib.ox.ac.uk/fsl/randomise/index.html), using 10,000 permutations. Small volume correction, applying ROI masks, was used to test a priori hypotheses. A false discovery rate threshold of 0.05 was used to correct for multiple comparisons.

The contrasts tested in volumetric analyses were identical to those tested in the voxel-wise analyses. Effects of TBV changes were regressed out. When contrasting Hique vs. Loque groups, the ROI volumes were corrected for both TBV change and percentage weight change. Age, gender and MR-scan interval were entered as covariates post-hoc. Moreover, TBV and weight-corrected volumetric changes and the corresponding follow-up ROI volumes were used to test for linear effects (using Spearman’s rank correlation coefficient) of medication and change in psychopathology. Change in psychopathology (baseline – follow-up) was calculated separately for positive, negative and total PANSS scores. Regional percentage volume change was calculated as ROI volume change divided by total brain change.

To control for any lifetime substance abuse diagnosis (n = 5), all analyses were also performed excluding these patients. All tests were two-tailed, and the significance level was set to p < 0.05.

Results

Attrition

Sixteen patients were not included at follow-up. Reasons for attrition were: clinically inadequate effect (n = 2), intolerable side-effects (sedation, weight gain, rise in liver enzymes) (n = 4), pregnancy (n = 2) or refusal to participate at follow-up (n = 5). Three patients had stated that after the baseline examinations they were not willing to undergo treatment with quetiapine. The 16 excluded patients had significantly smaller baseline ventricles (Z = −2.10, p = 0.044) compared to the patients who participated at follow-up. However, there were no baseline differences in demographic measures (p values > 0.38), PANSS scores (p values > 0.23), or in other volumetric brain measures (p values > 0.36).

Baseline

At baseline Hique patients had significantly higher lifetime exposure to benzodiazepines than Loque patients (Fisher’s exact test, p = 0.046). There were no other significant differences on baseline demographic, clinical and volumetric measures between the patient subgroups (Tables 1 and 3).

Follow-up

Demographics

The interval between baseline and follow-up MR scans did not differ between any of the patient groups and HC (Z = −0.51 p = 0.61). Patients gained significantly more weight than did HC (Z = −3.43 p = 0.001). The mean weight gain in patients was 7.8 kg (s.d. = 8.4), corresponding to 11.1% (s.d. = 12.4). There was no association between quetiapine dosage and percentage weight gain (ρ = 0.26, p = 0.24). Hique patients tended to use more benzodiazepines than Loque patients (Fisher’s exact test, p = 0.055). None of the patients
developed EPS. Follow-up PANSS scores were missing for one patient.

Global brain volumes

No significant global brain tissue volume (TBV, GM, WM) changes were observed between Ptall × HC, Hique × Loque, Hique × HC, and Loque × HC (p values >0.15).

Striatum

The voxel-wise analyses revealed significant bilateral volume loss in Ptall compared to HC in the caudate nucleus and putamen (Table 2). The volumetric analyses paralleled the voxel-wise results. The striatal volume was significantly reduced over time in Ptall compared to HC (Z = -2.21, p = 0.027). In post-hoc analyses of striatal subregions separately Ptall compared to HC had significant caudate (Z = -2.62, p = 0.009), putamen (Z = -1.99, p = 0.046), but not accumbens (Z = -1.72, p = 0.017) volume loss over time.

At follow-up Ptall had significantly smaller striatal volumes than did HC (Z = -2.15, p = 0.032). Post-hoc analyses of the striatal structures separately, revealed smaller caudate (Z = -2.76, p = 0.006) and accumbens (Z = -2.52, p = 0.012), but not putamen follow-up volumes (Z = 1.04, p = 0.30).

Striatum, dose-dependent medication effect

In the voxel-wise analyses bilateral striatal volume loss was significant in Loque, but not in Hique patients, Table 2. Results of the voxel-wise analyses

<table>
<thead>
<tr>
<th>Region</th>
<th>Subregion</th>
<th>Contrast</th>
<th>Side</th>
<th>Z score</th>
<th>p value</th>
<th>MNI coordinates (x, y, z)</th>
<th>Cluster size</th>
</tr>
</thead>
<tbody>
<tr>
<td>Striatum</td>
<td>Cau</td>
<td>Ptall × HC</td>
<td>Left</td>
<td>3.66</td>
<td>0.003</td>
<td>-15, 8, 22</td>
<td>2680</td>
</tr>
<tr>
<td></td>
<td>Cau</td>
<td>Right</td>
<td></td>
<td>3.01</td>
<td>0.004</td>
<td>19, 18, 15</td>
<td>2590</td>
</tr>
<tr>
<td></td>
<td>Put</td>
<td>Left</td>
<td></td>
<td>3.35</td>
<td>0.003</td>
<td>-30, -8, 10</td>
<td>2110</td>
</tr>
<tr>
<td></td>
<td>Put</td>
<td>Right</td>
<td></td>
<td>3.56</td>
<td>0.003</td>
<td>24, 1, 12</td>
<td>2180</td>
</tr>
<tr>
<td></td>
<td>Hique × HC</td>
<td>-</td>
<td></td>
<td>-</td>
<td>-</td>
<td>n.s.</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Cau</td>
<td>Loque × HC</td>
<td>Left</td>
<td>3.51</td>
<td>0.004</td>
<td>-16, 18, 11</td>
<td>5530</td>
</tr>
<tr>
<td></td>
<td>Cau</td>
<td>Right</td>
<td></td>
<td>3.36</td>
<td>0.004</td>
<td>18, 14, 13</td>
<td>3020</td>
</tr>
<tr>
<td></td>
<td>Put</td>
<td>Left</td>
<td></td>
<td>3.20</td>
<td>0.003</td>
<td>-26, -7, 9</td>
<td>2230</td>
</tr>
<tr>
<td></td>
<td>Put</td>
<td>Right</td>
<td></td>
<td>3.67</td>
<td>0.004</td>
<td>26, 2, 10</td>
<td>2610</td>
</tr>
<tr>
<td>Hiquest</td>
<td>-</td>
<td>Ptall × HC</td>
<td>Left</td>
<td>3.04</td>
<td>0.006</td>
<td>-22, -34, -4</td>
<td>3140</td>
</tr>
<tr>
<td>Hiquest</td>
<td>-</td>
<td>Right</td>
<td></td>
<td>3.09</td>
<td>0.006</td>
<td>15, -38, -1</td>
<td>2070</td>
</tr>
<tr>
<td>Hiquest</td>
<td>-</td>
<td>Right</td>
<td></td>
<td>2.44</td>
<td>0.02</td>
<td>14, -10, -17</td>
<td>75</td>
</tr>
<tr>
<td>Hiquest</td>
<td>-</td>
<td>Hique × HC</td>
<td>Left</td>
<td>3.39</td>
<td>0.00</td>
<td>-33, -31, -10</td>
<td>2970</td>
</tr>
<tr>
<td>Hiquest</td>
<td>-</td>
<td>Right</td>
<td></td>
<td>3.73</td>
<td>0.007</td>
<td>23, -33, -5</td>
<td>1500</td>
</tr>
<tr>
<td>Hiquest</td>
<td>-</td>
<td>Right</td>
<td></td>
<td>2.18</td>
<td>0.03</td>
<td>15, -12, -17</td>
<td>47</td>
</tr>
<tr>
<td>Hiquest</td>
<td>-</td>
<td>Loque × Hique</td>
<td>-</td>
<td>-</td>
<td>n.s.</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Hiquest</td>
<td>-</td>
<td>Hique × Loque</td>
<td>-</td>
<td>-</td>
<td>n.s.</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Ventricles</td>
<td>-</td>
<td>Ptall × HC</td>
<td>-</td>
<td>-</td>
<td>n.s.</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Hiquest</td>
<td>-</td>
<td>Hique × HC</td>
<td>-</td>
<td>-</td>
<td>n.s.</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Loque</td>
<td>-</td>
<td>Hique × HC</td>
<td>-</td>
<td>-</td>
<td>n.s.</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Loque</td>
<td>-</td>
<td>Loque × Hique</td>
<td>-</td>
<td>-</td>
<td>n.s.</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

Results of small volume correction with the study-specific masks applied to the different contrasts. Loque, Patients treated with quetiapine <538 mg/d; Hique, patients treated with quetiapine ≥538 mg/d; Ptall, all patients; HC, healthy control subjects; Cau, caudate nucleus; Put, putamen; MNI, Montreal Neurological Institute coordinates (cluster sizes are in mm³).

The displayed contrasts test for volume loss between the groups; however, in the ventricles, enlargement is tested. The opposite contrasts were also tested, but all were non-significant (data not shown). Contrasts were performed with total brain volume (TBV) change as a covariate. However, the contrast Hiq × Loque was performed with both TBV change and percentage weight change as covariates. All significant contrasts are displayed (p<0.05). Non-significant contrasts are assigned n.s. Entering age, gender and MR scan interval as covariates did not change the results. Exclusion of the five subjects with a lifetime substance abuse diagnosis did not alter the results. p values are false discovery rate-corrected (p<0.05).
compared to HC. Direct comparison between Hi\textsubscript{que} and Lo\textsubscript{que} patients was not significant (Table 2).

The volumetric analyses paralleled the voxel-wise results (Table 3). Lo\textsubscript{que} patients had significant striatal volume loss over time as compared to HC ($Z = -2.22$, $p = 0.027$). Hi\textsubscript{que} and HC did not differ in total striatal volume change ($p = 0.23$). Comparison between Hi\textsubscript{que} and Lo\textsubscript{que} patients was not significant ($Z = -0.50$, $p = 0.62$). In post-hoc analyses the volume loss in Lo\textsubscript{que} patients was accounted for by significant volume loss in caudate ($Z = -2.55$, $p = 0.010$) and putamen ($Z = -1.99$, $p = 0.047$), but not in accumbens ($Z = -1.37$, $p = 0.17$).

At follow-up Lo\textsubscript{que} ($Z = -2.10$, $p = 0.036$), but not Hi\textsubscript{que} ($Z = -1.24$, $p = 0.22$) patients had significantly smaller striatal volumes compared to HC. In direct comparison the patient groups did not differ significantly ($Z = -1.30$, $p = 0.19$). In post-hoc analyses the follow-up volume reduction in the Lo\textsubscript{que} group appeared in the caudate ($Z = -2.35$, $p = 0.019$) and in the accumbens ($Z = -2.47$, $p = 0.014$), but not in the putamen ($Z = -1.18$, $p = 0.24$).

The percentage striatal volume loss (corrected for total brain change) in the different groups were: Pt\textsubscript{all} = 0.8%, Lo\textsubscript{que} = 1.0%, Hi\textsubscript{que} = 0.5%, HC = -0.6% (Fig. 1).

In Pt\textsubscript{all} no significant linear correlations were observed between quetiapine dose and striatal volume loss or follow-up volume, respectively ($p$ values > 0.46).

**Ventricles**

Neither the voxel-wise (Table 2) nor the volumetric analyses revealed ventricle differences between any of the groups ($p$ values > 0.27). Exploratory analyses testing for group differences for third and lateral ventricle volumes separately were not significant. In Pt\textsubscript{all} no linear correlation between mean quetiapine dose and ventricle volumes were observed ($p$ values > 0.19).

**Hippocampus**

Significant bilateral hippocampal volume loss in Pt\textsubscript{all} compared to HC was revealed both by the voxel-wise analyses (Table 2) and the volumetric analyses ($Z = -3.23$, $p = 0.002$) (Table 3). At follow-up Pt\textsubscript{all} had significantly smaller hippocampal volumes than HC ($Z = -2.13$, $p = 0.033$).

**Hippocampus, dose-dependent medication effect**

In the voxel-wise analyses bilateral hippocampal volume loss was significant in Hi\textsubscript{que}, but not in Lo\textsubscript{que} patients compared to HC. Direct comparison between Hi\textsubscript{que} and Lo\textsubscript{que} patients was not significant (Table 2).

The volumetric analyses paralleled the voxel-wise results by showing significant hippocampal volume loss in Hi\textsubscript{que} ($Z = -3.22$, $p = 0.001$), and trend-level volume loss in Lo\textsubscript{que} ($Z = -1.93$, $p = 0.053$) patients compared to HC. There was no difference between the two patient groups ($Z = -1.37$, $p = 0.17$).

At follow-up Hi\textsubscript{que} ($Z = -2.90$, $p = 0.004$), but not Lo\textsubscript{que} ($Z = -0.76$, $p = 0.45$) patients had smaller hippocampal volumes compared to HC; however, this difference was not significant ($Z = -0.97$, $p = 0.33$) between the patient groups.

The percentage hippocampal volume loss (corrected for total brain change) in the different groups were: Pt\textsubscript{all} = 0.6%, Lo\textsubscript{que} = 0.3%, Hi\textsubscript{que} = 1.0%, HC = -0.4% (Fig. 2).

In Pt\textsubscript{all} no linear correlations were observed between quetiapine dose and hippocampal volume loss or follow-up volume, respectively ($p$ values > 0.21).
Whole brain
Exploratory voxel-wise whole-brain analyses did not reveal additional regional GM, WM or CSF volume loss between groups.

Psychopathology
From baseline to follow-up patients improved significantly on positive ($t_{20} = 5.74, p < 0.001$) and total symptoms ($t_{20} = 2.88, p = 0.009$), but not on negative symptoms ($t_{20} = 1.56, p = 0.14$). There were no significant correlations between mean quetiapine doses and baseline, follow-up or changes in PANSS positive, negative and total scores ($p$ values > 0.15).

A positive correlation between baseline positive symptoms and striatal volume loss over time ($\rho = 0.44, p = 0.042$) was found (Fig. 3a). Post-hoc analyses revealed positive correlations with volume loss in all three striatal structures separately (caudate: $\rho = 0.51, p = 0.016$; accumbens: $\rho = 0.52, p = 0.013$; putamen: $\rho = 0.43, p = 0.047$). Further, we found a positive correlation between baseline positive symptoms and hippocampal volume loss over time ($\rho = 0.51, p = 0.016$) (Fig. 3b). Finally, ventricle increase over time was associated with less improvement in negative symptoms ($\rho = -0.46, p = 0.038$) (Fig. 4). The correlations did not survive a correction for multiple comparisons. No other linear correlations between volumetric changes or follow-up volumes, and positive, negative or total symptoms were observed.

Adding age, gender and MR interval as covariates did not change the above results significantly. Similarly, exclusion of the five subjects with lifetime substance abuse diagnosis did not significantly alter the results.

Discussion
In this longitudinal TBM study we investigated medication and psychopathological effects on brain...
structure in 22 initially antipsychotic-naive, first-episode schizophrenia patients after treatment with quetiapine for approximately 6 months. The main finding was that patients had significant bilateral striatal and hippocampal volume loss during the treatment period. The striatal volume loss was most pronounced in patients treated with low quetiapine doses whereas the volume loss was less apparent in patients treated with high doses. Conversely, hippocampal volume loss appeared more pronounced in patients treated with high quetiapine doses than in patients treated with low doses. However, the dose-dependent differences were only apparent in patient subgroups when compared to HC. Clinically, higher baseline positive symptoms were associated with more striatal and hippocampal volume loss over time. Although patients’ ventricles did not change significantly over time, ventricular increases were associated with less improvement on negative symptoms. No significant changes in global brain measures (TBV, GM, WM) were observed.

Quetiapine treatment has previously been associated with striatal volume loss in already medicated patients (Stip et al. 2008). However, in the present study patients were initially antipsychotic-naive and therefore quetiapine in itself may be associated with striatal volume reduction. One previous study in antipsychotic-naive patients reported no significant caudate volume reduction in 10 patients following 12 wk of quetiapine treatment (mean dose 494 mg/d) (Tauscher-Wisniewski et al. 2005). Nevertheless, they observed that the absolute caudate volume decreased over time. Their failure to observe significant effects might be related to the small subject sample and the relative short treatment period.

Fig. 3. Positive symptoms and striatal and hippocampal loss. Positive correlations between baseline Positive and Negative Syndrome Scale (PANSS) positive symptoms and (a) striatal ($\rho = 0.44, p = 0.042$), and (b) hippocampal ($\rho = 0.51, p = 0.016$) volume loss (in mm$^3$) over time. Volumes were corrected for changes in total brain volume (TBV) and percentage weight change. The correlations did not survive a correction for multiple comparisons. Note, that grey-matter loss is indicated with higher values on the y-axis. See text for details.

Fig. 4. Negative symptoms and ventricular volume increase. A negative correlation between improvement in negative symptoms [Positive and Negative Syndrome Scale (PANSS) negative baseline – follow-up] and ventricle increase (in mm$^3$) over time ($\rho = -0.46, p = 0.038$). Volumes were corrected for changes in TBV and percentage weight change. The correlation did not survive a correction for multiple comparisons. Note that symptom improvement is indicated with higher values on the x-axis and that ventricular increase is indicated with higher values on the y-axis. See text for details.
Our data suggest that high quetiapine doses may attenuate the striatal volume loss. Dose-dependent volumetric effects of quetiapine have not previously been described. Interestingly, clinical studies with risperidone have associated striatal volume increase with high doses of risperidone (6.05 mg/d) (Massana et al. 2005), whereas moderate (3.64 mg/d) (Glenthøj et al. 2007) and low (2.7 mg/d) (Lang et al. 2001) risperidone doses caused marginal increases or no striatal volume changes, respectively. According to a hypothesis put forward by Kapur & Seeman (2001), a transiently high striatal D<sub>2</sub> blockade may drive the antipsychotic effect in SGAs. We infer that our observation of relative striatal volume increases with high quetiapine doses could reflect this transiently high D<sub>2</sub> occupancy. Conversely, in low doses the clinical effect may be more related to 5-HT<sub>2</sub> occupancy. Conversely, in low doses the clinical effect may be more related to 5-HT<sub>2A</sub> occupancy as suggested by our previous PET data (Rasmussen et al. 2010). From the present study it is not possible to determine whether the observed striatal volume reduction is a consequence of the disease progress per se or of the 5-HT<sub>2A</sub> blockade. However, post-mortem studies have indicated that 5-HT<sub>2A</sub> receptor density in striatum is low (Dwivedi & Pandey, 1998) and that the striatal volume and neuron number decrease over the course of the illness regardless of chronic treatment with FGAs (Kreczmanski et al. 2007).

Since we did not asses D<sub>2</sub> blockade, inferences from the present data must be made cautiously. Nevertheless, the absence of linear correlations between quetiapine dose and striatal and hippocampal changes, respectively, supports the hypothesis of a threshold of 5-HT<sub>2A</sub>- and D<sub>2</sub>-mediated clinical effects. Although quetiapine doses of 400–450 mg have resulted in dopamine D<sub>2</sub> occupancies of ~60% 2 h post-dose (Kapur et al. 2000; Tauscher-Wisniewski et al. 2002) also lower occupancies of 44% and 30% 2 h post-450 mg quetiapine have been reported (Gefvert et al. 1998, 2001). Indisputably, a dose of 538 mg/d as applied in our study may not address the ‘true’ threshold. Larger sample sizes, preferably including PET data are required to confirm this possible dose-dependent effect of quetiapine and to identify a more precise threshold. Finally, the mechanisms behind striatal volume increase after sustained blockade of the D<sub>2</sub> receptors are still elusive, but up-regulation of the D<sub>2</sub> receptors (Burt et al. 1977; Seeman, 1987), axonal sprouting (Benes et al. 1983) and increased striatal blood flow (Corson et al. 2002; Miller et al. 1997) have been suggested.

Contrary to findings in preclinical studies, quetiapine did not seem to protect against hippocampal volume loss (Bi et al. 2009; Park et al. 2006). No linear correlation between mean dose and volume loss was observed and the hippocampal volume loss appeared most pronounced in the high-dose group compared to HC. Interestingly, this inverted dose-dependent association is supported by a preclinical study on rat hippocampi in which low doses of clozapine (0.5 mg/kg) increased markers of DNA synthesis in hippocampus whereas high doses (20 mg/kg) had no effect (Halim et al. 2004).

Accumulating evidence suggests that ventricular enlargement occurs during the early course of schizophrenia (Steen et al. 2006; Van Haren et al. 2008; Vita et al. 2006). Although antipsychotic treatment may exert a preserving effect on ventricular volume (Lieberman et al. 2001), a recent review has suggested that antipsychotic treatment may on the contrary be associated with ventricular enlargement (Moncrieff & Leo, 2010). Similarly, the long-term effects of antipsychotic compounds on global brain volume are unclear (Moncrieff & Leo, 2010). The present results suggest that quetiapine may protect against ventricular enlargement and global brain loss in the early course of illness.

Treatment with quetiapine improved PANSS positive and total scores although negative symptoms did not improve significantly. The absence of correlations between mean quetiapine dose and any of the PANSS measures, allowed for independent investigations of mediation effects and psychopathology. On the other hand, this absence of an association between medication dose and psychopathology renders it controversial whether regional GM changes induced by antipsychotics are clinically beneficial (Navari & Dazzan, 2009; Smieskova et al. 2009). The clinical complexity of schizophrenia combined with the pharmacological complexity of quetiapine may partly explain why a dose–response relationship is not well-established (Kinon et al. 2004) and an optimal therapeutic range for quetiapine remains to be identified (Mauri et al. 2007).

MRI studies have previously associated striatal volume reductions with positive symptoms (Crespo-Facorro et al. 2007; Scheepers et al. 2001); however, the present study is the first to show that the severity of positive symptoms in the antipsychotic-naive state may predict progressive striatal volume loss. Interestingly, in subjects at high risk for developing psychosis, progressive hippocampal volume reductions have been associated with later transition to schizophrenia, although also reduced activation in prefrontal cortex, reduced neuronal density, and increased membrane turnover in frontal and cingulate cortex seem implicated in transition (Smieskova et al. 2009).
2010; Wood et al. 2008). Our finding of a positive correlation between baseline positive symptoms and progressive hippocampal loss could therefore suggest that the hippocampus is particularly sensitive to positive symptoms around and right after transition to psychosis. We speculate that this hippocampal sensitivity might reflect a higher level of stress accompanying a psychotic state possibly related to disturbances in the hypothalamic–pituitary–adrenal (HPA) axis (Phillips et al. 2006). Although we did not find ventricular enlargement over time a significant correlation between ventricular increase and less negative symptom improvement was observed. As such our data are consistent with the growing body of evidence which has associated negative symptoms and poor prognosis with ventricular enlargement (Hulshoff Pol & Kahn, 2008).

The present study is limited by several factors. First, the attrition was considerable during the 6-month period (42%). Notably, only 6/16 dropouts were directly related to quetiapine. Only ventricular volumes were significantly different (smaller) and no other demographic, clinical or structural differences between the dropouts and re-scanned patients were found. This suggests that our results were not biased by baseline differences. In our analyses of dose-dependent effects the patient subgroups are small and the observed differences appear in comparison with HC, whereas direct comparisons between the subgroups are non-significant. Nevertheless, the direction of the dose-dependent effect on striatum was a-priori hypothesized, rather than a result of a post-hoc comparison. Hence our failure to detect significant changes in direct comparison between the patient subgroups could be attributable to lack of power (a type II error). Moreover, a potential inaccuracy in our threshold of 538 mg/d would also reduce the differences between the patient groups. Ideally, a study of dose-dependent medication effects should apply a double-blind, randomized design; however, for ethical reasons this was not performed. The absence of correlations between our obtained clinical measures and medication dose indicate that the patient subgroups were not biased by illness severity. Finally, it should be emphasized that although clinically meaningful, our findings regarding structural changes and psychopathology appeared among several correlations and they would not survive a correction for multiple comparisons.

In conclusion we found that 6 months’ treatment with quetiapine in antipsychotic-naive, first-episode schizophrenia patients was associated with progressive bilateral volume loss in striatum and hippocampus. The progressive volumetric loss may be dose-dependent and clinically relevant. The mechanisms underlying associations between progressive brain changes and specific antipsychotic compounds and clinical symptoms warrant further clarification.

Acknowledgements
This study was sponsored by The Danish Medical Research Council, H:S (Copenhagen Hospital Cooperation), The Lundbeck Foundation, Gerda and Aage Haensch’s Foundation, Slagtermester Max Worzner og hustru Inger Worzner’s Foundation, The Danish Psychiatric Association and an unrestricted grant was received from AstraZeneca A/S, Denmark. [Trial Registration: Clinicaltrials.gov NCT00207064 http://clinicaltrials.gov/ct2/show/NCT00207064.]

Statement of Interest
None.

References
Angelucci F, Mathe AA, Aloe L (2000). Brain-derived neurotrophic factor and tyrosine kinase receptor TrkB in rat brain are significantly altered after haloperidol and risperidone administration. Journal of Neuroscience Research 60, 783–794.


Kerwin R (2007). When should clozapine be initiated in schizophrenia?: some arguments for and against earlier use of clozapine. CNS Drugs 21, 267–278.


Seeman P (1987). Dopamine receptors and the dopamine hypothesis of schizophrenia. Synapse 1, 133–152.


Taylor S, Christensen JD, Holcomb JM, Garver DL (2005). Volume increases in striatum associated with positive...
symptom reduction in schizophrenia: a preliminary observation. Psychiatry Research 140, 85–89.