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Schizophrenia with auditory hallucinations: A voxel-based morphometry study

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Abstract

Many studies have shown widespread but subtle pathological changes in gray matter in patients with schizophrenia. Some of these studies have related specific alterations to the genesis of auditory hallucinations, particularly in the left superior temporal gyrus, but none has analysed the relationship between morphometric data and a specific scale for auditory hallucinations. The present study aims to define the presence and characteristics of structural abnormalities in relation with the intensity and phenomenology of auditory hallucinations by means of magnetic resonance voxel-based morphometry (MR-VBM) method applied on a highly homogeneous group of 18 persistent hallucinatory patients meeting DSM-IV criteria for schizophrenia compared to 19 healthy matched controls. Patients were evaluated using the PSYRATS scale for auditory hallucinations. Reductions of gray matter concentration in patients to controls were observed in bilateral insula, bilateral superior temporal gyri and left amygdala. In addition, specific relationships between left inferior frontal and right postcentral gyri reductions and the severity of auditory hallucinations were observed. All these areas might be implicated in the genesis and/or persistence of auditory hallucinations through specific mechanisms. Precise morphological abnormalities may help to define reliable MR-VBM biomarkers for the genesis and persistence of auditory hallucinations.

Keywords: Auditory hallucinations; Magnetic resonance imaging; PSYRATS; Schizophrenia; Voxel-based morphometry

1. Introduction

Pathological changes in the gray matter (GM) have been shown in patients with schizophrenia by means of magnetic resonance (MR) imaging techniques. These morphologic changes are widespread (Honea et al., 2005) but subtle and with a variable level of evidence and expression (Shenton et al., 2001). Region of interest (ROI) analyses, which are landmark-based manual procedures, have been the gold standard for structural MR examinations. While their advantages include anatomical validity, definition of landmarks in native space and quantitative measures of the voxels in the regions under study, they are time consuming and are of varying reliability. Focused ROI procedures are mainly useful in situations where the expected changes are small and the involved areas are clearly defined. Even more, multi-ROI studies are tedious to perform, and need many inter- and intra-subject reliability tests, which are not thoroughly performed. For this reason, several automated whole-brain techniques have been developed.

Voxel-based morphometry (VBM) (Ashburner and Friston, 2000; Good et al., 2001) is a fast and reliable technique, although less sensitive than focused ROI procedures. VBM provides the relative changes in the concentration of GM within

Abbreviations: AAL, Automated Anatomical Labeling; AH, Auditory Hallucinations; BPRS, Brief Psychiatric Rating Scale; CSF, Cerebrospinal Fluid; DSM-IV, Diagnostic and Statistical Manual of Mental Disorders, Forth Edition; FDR, False Discovery Rate; GM, Gray Matter; MR, Magnetic resonance; PSYRATS, Psychotic Symptom Rating Scale; RFT, Random Field Theory; ROI, Region of Interest; SnPM, Statistical non-Parametric Mapping; SPM, Statistical Parametric Mapping; VBM, Voxel-based morphometry; WM, White Matter.

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each voxel. Therefore, brain areas shown to be abnormal with VBM analysis represent specific points of maximal changes instead of defined affected regions.

Both VBM and ROI measurement studies have tried to correlate volumetric changes with auditory hallucinations (AH) in patients with schizophrenia (Tables 1 and 2). Most of these studies have looked for specific relationship between superior temporal gyrus reductions and AH severity (Barta et al., 1990; Flaum et al., 1995; Levitan et al., 1999; Rajarethinam et al., 2000; Matsumoto et al., 2001; Onitsuka et al., 2004). Left Heschl's gyrus reduction has also been specifically related to AH (Sumich et al., 2005). Structural reductions associated with AH have also been identified in other areas as the left anterior cingulate (Noga et al., 1995), left anterior hippocampus (Rajarethinam et al., 2001), left middle temporal gyrus (Onitsuka et al., 2004), and also a leftward asymmetry of the sylvian fissure (Shapleske et al., 2001).

Also, voxel-based studies have found a possible relationship between different structural abnormalities and AH. Shapleske et al. (2002) compared prominent auditory verbal hallucinatory and non-hallucinatory patients and described only one significant cluster with a deficit in GM including the left insula and adjacent medial temporal lobe. A significant correlation was found between size of the left insula cluster and hallucinations. Milev et al. (2003) prospectively studied 123 psychotic patients during a period of 5 years using an automated stereotaxic-based parcellation method showing a bilateral temporal lobe GM reduction associated with the persistence of AH. Using a similar method, Shin et al. (2005) studied 25 drug-naïve schizophrenic patients showing that the hallucinatory group had right temporal white matter (WM), and frontal and temporal GM excess. Gaser et al. (2004) found, using deformation-based morphometry, that the severity of AH was significantly correlated with volume loss in the left Heschl's gyrus and in the left supramarginal gyrus, as well as middle and inferior right prefrontal gyri. Finally, Neckelmann et al. (2006) have recently published a VBM study in which the superior (transverse) temporal gyrus, left thalamus, and left and right cerebellum density were negatively correlated with hallucinations.

Noteworthy, results are not completely in agreement. Clinical heterogeneity is probably one of the main reasons for the discrepancies. Studying large and homogeneous patient samples through VBM analyses may reduce this bias (Kubicki et al., 2002; Shapleske et al., 2002). On the other hand, most ROI studies have looked at the superior temporal gyrus and therefore, other potentially relevant areas have not been routinely evaluated. Although global analysis can be performed by whole-brain studies, only five such studies have been focused on AH and data are not completely in agreement (Tables 1 and 2). Finally, the available data show that some of the phenomenological dimensions of AH have specific neural substrates (Stephane et al., 2003). This may be another source of disagreement and gives support to the recommendation of using more accurate scales to measure unreality symptoms (Sumich et al., 2005).

The present study explores the relationship between AH and structural brain abnormalities through a VBM study. The examination was performed on a highly homogeneous group of schizophrenic patients with persistent hallucinations by using a specific clinical scale for assessing multiple dimensions of AH.

Table 1
Summary of MR imaging studies reporting positive findings in schizophrenia and AH

Author/year	Sample	Clinical evaluation	MRI technique	Association with auditory hallucinations
Barta et al. (1990)	15 M medicated Schiz/15 HC	Retrospective SAPS	ROIs	Left STG reduction
Flaum et al. (1995)	166 Schiz spectrum patients	CASH	ROIs	Left STG reduction
Levitan et al. (1999)	30 Schiz with history of AH	SAPS	ROIs	Left anterior STG reduction
Rajarethinam et al. (2000)	20 M Schiz/20 HC	BPRS	ROIs	Left anterior STG reduction
Matsumoto et al. (2001)	40 recent-onset Schiz/40 HC	PANSS	ROIs	Right STG reduction
Rajarethinam et al. (2001)	20 M Schiz/20 HC	BPRS	ROIs	Left anterior hippocampus reduction
Shapleske et al. (2001)	74 M Schiz (30 without AH and 44 with strong history of AH)/32 HC	SAPS (for sample characterization)	ROIs	Sylvian fissure asymmetry (the longer the left Sylvian fissure, the more severe)
Shapleske et al. (2002)	72 M Schiz (41 with prominent AH and 31 without AH)	SAPS	Voxel-based method similar to VBM	Left insula and right medial temporal lobe reduction; right inferior parietal lobe and white matter in left temporal–parietal connecting tracts excess
Milev et al. (2003)	123 Schiz, Schizophreniform or Schizoaffect	Outcome at a mean of five years later	Automated stereotaxic based parcellation method	Bilateral temporal lobe gray matter volumes reduction (with persistence of AH)
Gaser et al. (2004)	85 (56 non-hallucinating and 29 hallucinating) Schiz	SAPS	Deformation-based morphometry	Left transverse temporal gyrus of Heschl, right middle/inferior prefrontal gyri and left inferior supramarginal gyrus reduction
Onitsuka et al. (2004)	23 M, chronic Schiz/28 HC	SAPS	ROIs	Left STG and MTG
Shin et al. (2005)	25 drug-naïve Schiz (17 with and 8 without AH)	SCID (for sample characterization)	Automated stereotactic-based parcellation method	Right temporal white matter, frontal gray matter and temporal gray matter excess
Sumich et al. (2005)	25 first-episode psychotic patients	PANSS	ROIs	Left Heschl's gyrus reduction
Neckelmann et al. (2006)	12 Schiz/12 HC	BPRS	VBM	Left STG gyrus, bilateral cerebellum and left thalamus reduction

Abbreviations: M, males; Schiz, patients with schizophrenia; HC, healthy controls; STG, superior temporal gyrus; MTG, middle temporal gyrus.

Table 2
Summary of MR imaging studies reporting negative or mixed findings in schizophrenia and AH

Author/year	Sample	Clinical evaluation	MRI technique	Association with auditory hallucinations
Delisi et al. (1994)	85 first-episode Schiz/40 HC	SADS, chart review	ROIs	Not with size or lateralization of planum temporale or STG
Zipursky et al. (1994)	22 M, chronic Schiz/20 HC	BPRS	ROIs	Not with STG or hippocampus
Noga et al. (1995)	14 Schiz/14 HC	Retrospective SAPS	ROIs	Left anterior cingulated reduction, not significant after Bonferroni correction
Marsh et al. (1997)	56 M, severely ill, Schiz inpatients/52 HC	BPRS	ROIs	Not with STG
Havermans et al. (1999)	30 Schiz (15 with chronic AH and 15 without AH)/17 HC	BPRS (for sample characterization)	ROIs	Not with STG and other temporal lobe structures
Rossell et al. (2001)	71 M (42 with and 29 without history of AH) Schiz/33 HC	SAPS (for sample characterization)	ROIs	Not with corpus callosum areas

Abbreviations: M, males; Schiz, patients with schizophrenia; HC, healthy controls; STG, superior temporal gyrus; MTG, middle temporal gyrus.

We hypothesized reductions of GM in these patients, correlating with the AH severity. If statistically validated, a reliable morphometric biomarker for persistent AH in this homogeneous phenomenological group of schizophrenic patients will be obtained.

2. Methods

2.1. Subjects

Thirty seven subjects were selected for this study, including 18 patients with DSM-IV schizophrenia and persistent AH and 19 healthy control subjects. Patients' ages ranged from 21 to 42 years (mean 35.71±6.11) while the control group ranged from 20 to 48 years (mean 33.11±7.61). Both groups were also matched by sex (all males), ethnic group (all Caucasian), laterality (all right handed), and educational level (all had a secondary school qualification). All participants gave written permission in order to participate in the study, which was approved by the local ethics committee. Exclusion criteria included previous electroconvulsive therapy or severe head trauma, present or past criteria for drug abuse (except for tobacco and cannabis), carrying a metallic prosthesis or suffering from a hearing impairment. All patients were in maintenance phase and met the following selection criteria for persistent hallucinations:

a) Voices were not modified in any way by treatment over the course of a year.
b) At least two antipsychotic drugs had been tried, at doses equivalent to 600 mg/day of chlorpromazine, in the last year.

Patients were assessed with the Psychotic Symptom Rating Scale (PSYRATS) for AH (Haddock et al., 1999) and the Brief Psychiatric Rating Scale (BPRS) (Ventura et al., 1993) by a trained evaluator, just before data acquisition. One of these patients had to be discarded because of having completely disorganized behaviour, thinking and speech with an unmeasurable range of hallucinations. The PSYRATS scale rates several characteristics of AH in 11 domains on a five-point scale: frequency, duration, location, loudness, beliefs about the origin of voices, amount of negative content, degree of negative content, amount of distress, intensity of distress, disruption to life and controllability of voices. The BPRS consists of 18 items, each to be rated in a 7-point scale of severity ranging from 'not present' to 'extremely severe'. Table 3 shows clinical and demographical variables.

Table 3
Data from both groups of subjects

	Control subjects (n=19)	Schizophrenic patients (n=17)
Age (years)	33.11±7.61	35.71±6.11
Age first hallucinations (years)	–	18.82±7.76
GAF [a]	–	41.29±9.51
BPRS [b]	–	53.41±8.92
PSYRATS [c]	–	30.56±4.26

Data are displayed as mean±SD.
[a] Global Assesment of Function.
[b] Brief Psychiatric Rating Scale.
[c] Psychotic Symptom Rating Scale.

2.2. Scanning protocol

Three dimensional high-resolution whole-brain images were obtained with spoiled gradient-echo T1-weighted sequence on a 1.5 T MR magnet (Intera, Philips Medical Systems, Best, The Netherlands). The acquisition protocol parameters were: 96 axial slices covering the whole brain, TR=7 ms, TE=1.9 ms, flip angle=8°, 1.25 mm slice thickness with no inter-slice gap, acquisition matrix=256×256 and field of view=220 mm, achieving a voxel size of 0.86×0.86×1.25 mm.

Table 4
Areas with GM density reduction in schizophrenic patients vs. control subjects showed by VBM analysis

Coordinates (mm)			Label	L/R	t value	p(cluster)
X	Y	Z				
−43	12	−10	Insula	L	5.84	0.00
45	16	−9	Insula	R	5.10	0.00
57	−35	8	Superior temporal gyrus	R	3.87	0.11
−23	4	−22	Amygdala	L	3.56	0.03
−37	8	−18	Superior temporal gyrus	L	3.51	0.04

p<0.05 FDR corrected k=135. Corrected p values are shown at cluster-level.

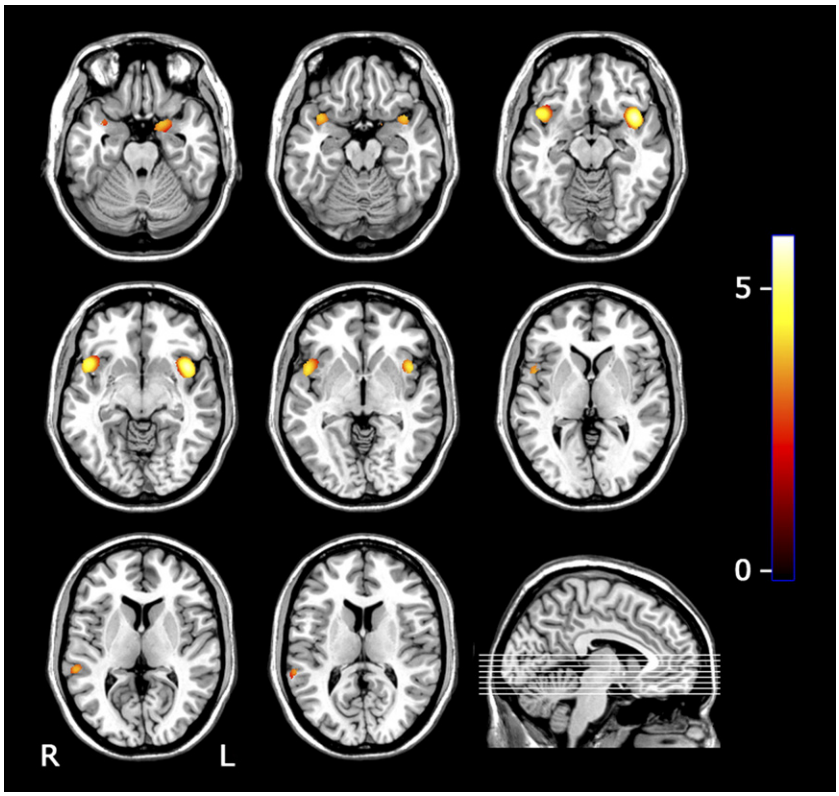

Fig. 1. Areas of GM density reduction in schizophrenic patients with persistent auditory hallucinations vs. control subjects reported by VBM analysis. $p<0.05$ FDR corrected $k=135$.

2.3. Image processing

Statistical Parametric Mapping (SPM2, Wellcome Institute, London, United Kingdom) was used to perform the image processing. The tests were carried out using MATLAB 7.1 (The MathWorks, Natick, MA, USA). Statistically significant anatomical differences between patients and healthy subjects were measured using the optimized VBM protocol (Ashburner and Friston, 2000).

Custom templates were created to minimise the bias induced by using standard anatomical templates in the normalisation processes. The creation of custom templates process involved the normalisation of each raw image with the standard ICBM 152 template applying a 12-parameter affine transformation to

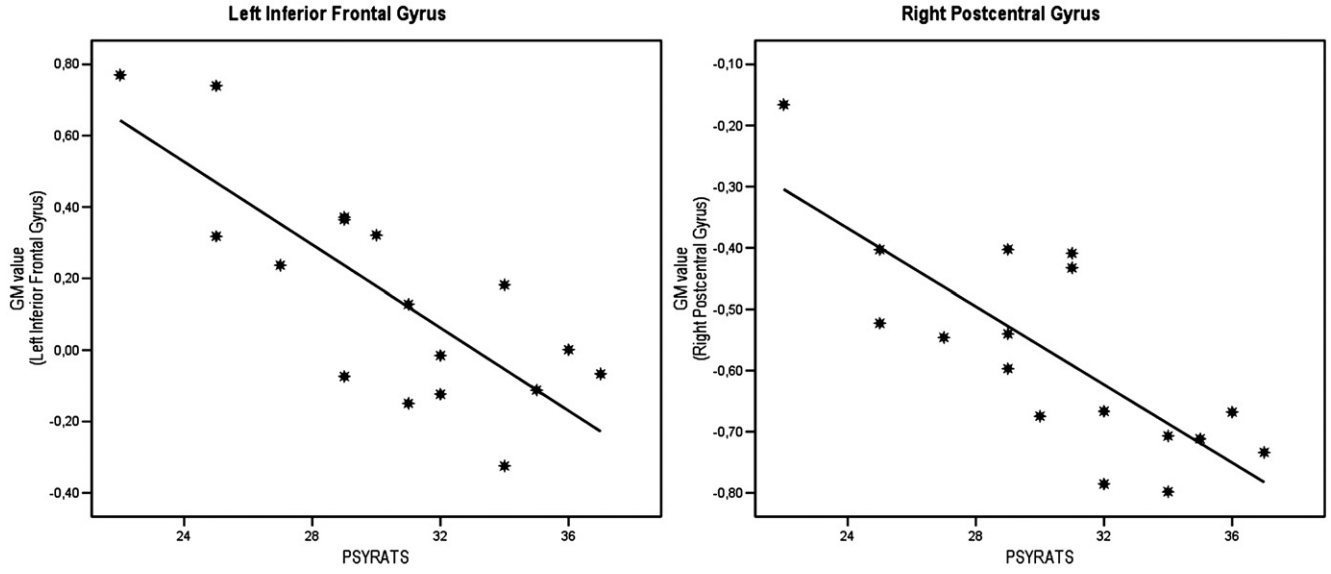

Fig. 2. Scatterplot of GM values (arbitrary units) negatively correlated with PSYRATS scale in schizophrenic patients with persistent auditory hallucinations in the left inferior frontal gyrus ($r^2=0.60$) and in the right postcentral gyrus ($r^2=0.59$). $p<0.05$ FDR corrected $k=37$.

Table 5
Areas negatively correlated between PSYRATS and GM values in schizophrenic patients

Coordinates (mm)			Label	L/R	*t* value	*p*(cluster)
X	*Y*	*Z*				
−49	18	−3	Inferior frontal gyrus	L	4.93	0.04
68	−7	19	Postcentral gyrus	R	3.94	0.11

$p<0.05$ FDR corrected $k=37$. Corrected p values are shown at cluster-level.

place the data in a common stereotactic space (Good et al., 2001). The normalised images were then segmented to obtain GM maps, which were averaged and smoothed with an 8-mm Gaussian smoothing filter obtaining a whole-brain template and a specific GM template.

To perform the optimized VBM analysis, each original T1-weighted MR image was normalised with the created whole-brain template and again segmented to obtain GM tissue maps. At this stage, segmentation involved a cleaning process that removed non-useful tissue such as scalp, skull and dural venous sinus (Good et al., 2001). Non-linear spatial normalisation (Ashburner and Friston, 1999) parameters between GM segmented images and GM template maps were then estimated and applied in order to warp the original T1 images. Finally, warped GM images were segmented and smoothed by a 12-mm Gaussian kernel.

2.4. Statistical analysis

Statistical measurements were performed with the Statistical non-Parametric Mapping (SnPM) (Nichols and Holmes, 2002) software, which provides a suitable framework for analyses when conditions as low degrees of freedom (small size of the sample) make the variance estimation noisy and, therefore, the assumptions of the Random Field Theory (RFT) about field continuity not feasible.

The SnPM software takes into consideration the multiple comparison problem and provides a smoothed variance estimation procedure to inherently deal with this issue (Holmes et al., 1996). A significance criterion was established by using a $p<0.05$, with correction for multiple comparisons following the False Discovery Rate (FDR) methodology (Genovese et al., 2002).

In order to measure the morphometric differences and evaluate the anatomical correlations with BPRS and PSYRATS scales, two independent statistical models were generated. The model constructed for assessing structural inter-group variability included a group condition (patients vs. controls) and two nuisance variables (age and total intracranial volume). Total intracranial volume was computed from the GM, WM and cerebrospinal fluid (CSF) maps obtained after segmenting the original raw images. GM, WM and CSF tissue volumes were calculated by integrating the probability value of each voxel with the voxel size. These three values were added to derive the total intracranial volume. Statistical parametric maps were obtained by performing permutation tests over the GM maps, using one-tailed contrasts in order to measure differences among groups. The number of permutations was fixed to 30,000. This value was chosen in order to obtain a random sub-sample of all possible permutations with a confidence interval of 95% for a given threshold of $p<0.05$. Additionally, a spatial filter was applied to eliminate spurious findings by considering only those clusters with a minimum size of $k=135$ voxels (expected number of voxels per cluster).

To perform the correlation analysis between PSYRATS and BPRS variables with structural variability, another statistical model was built including the GM images of patients and the PSYRATS and BPRS scores. A correlation test was performed to evaluate the relationship between illness duration and brain volume of each patient. As results were not significant ($r^2=0.00$; $p=0.90$), these variables were not included in the model. Estimation parameters were adjusted looking for linear regressions

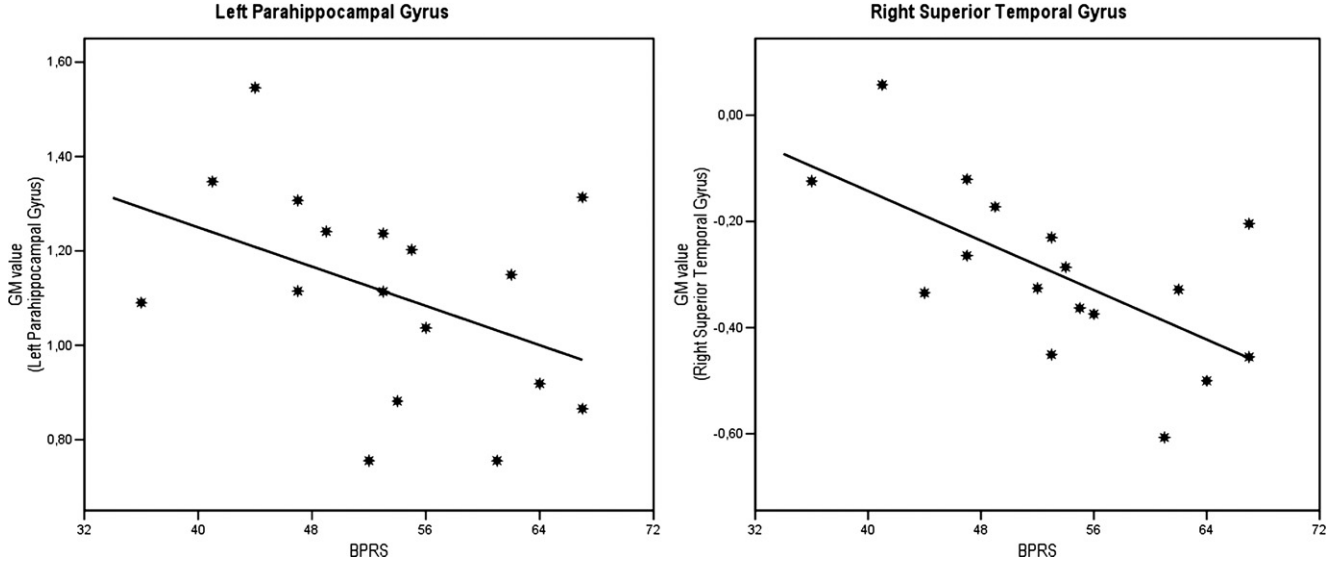

Fig. 3. Scatterplot of GM values (arbitrary units) negatively correlated with BPRS scale in schizophrenic patients with persistent auditory hallucinations in the left parahippocampal gyrus ($r^2=0.17$) and in the right superior temporal gyrus ($r^2=0.40$). $p<0.05$ FDR corrected $k=37$.

between the PSYRATS and BPRS scales related to GM signal changes. The significance threshold was fixed at $p<0.05$ FDR corrected and the cluster filter was set to $k=37$ voxels (expected number of voxels per cluster). The model was estimated against 30,000 permutations.

Both, morphometric and correlation results were labelled with the Automated Anatomical Labeling (AAL) software (Tzourio-Mazoyer et al., 2001). Areas-identifying coordinates were determined by the maximum Student-*t* value in the corresponding brain area.

3. Results

3.1. Optimized VBM method

GM density reduction in patients vs. controls ($patients_{GM}<controls_{GM}$) was found in the areas shown in Table 4 and represented in Fig. 1. The results showed a significant ($p<0.05$ FDR Corrected, $k=135$) GM decreased concentration in insula (bilateral), superior temporal gyrus (bilateral) and amygdala (left). $Patients_{GM}>controls_{GM}$ comparison (GM concentration increases in patients vs. controls) did not produce any significant results.

3.2. PSYRATS and BPRS regression

Significant ($p<0.05$ FDR Corrected, $k=37$) negative correlations between each PSYRATS and BPRS scales related to focal GM density reduction were observed in patients. PSYRATS variable was negatively correlated in the left inferior frontal gyrus ($r^2=0.60$; $p=0.04$) and in the right postcentral gyrus ($r^2=0.59$; $p=0.11$) (Fig. 2, Table 5), while negative BPRS correlation affected the left parahippocampal ($r^2=0.17$; $p=0.21$) and the right superior temporal gyrus ($r^2=0.40$; $p=0.03$) (Fig. 3, Table 6) regions.

3.3. Exploratory analyses: specific PSYRATS items and self-related areas

Exploratory correlations were also performed between three PSYRATS domains (items #2 or duration, #5 or beliefs about the origin of voices, and #11 or controllability of voices) which we hypothesized as of particular relevance, and GM values in insula. Significant ($p<0.05$ FDR corrected, $k=37$) negative correlations between 'duration' and bilateral insula and between 'controllability of voices' and left insula GM values were observed. Significant ($p<0.05$ FDR corrected, $k=37$) positive correlations were also obtained between 'beliefs about the origin of voices' and bilateral insula GM values.

Table 6
Areas with negative correlation between BPRS and GM values in schizophrenic patients

Coordinates (mm)			Label	L/R	*t* value	*p*(cluster)
X	*Y*	*Z*				
−17	−14	−25	Parahippocampal gyrus	L	4.29	0.21
70	−29	8	Superior temporal gyrus	R	3.70	0.03

$p<0.05$ FDR corrected $k=37$. Corrected *p* values are shown at cluster-level.

4. Discussion

4.1. Differences between patients and controls

GM reductions in bilateral insula and superior temporal gyrus, altogether with a left amygdala decrease have been detected in our highly homogeneous sample of patients with persistent AH compared to healthy controls. According to this, these areas seem to participate in the structural network involved in the persistence of AH. Some functional studies also support the multiplicity of areas involved in AH (Shergill et al., 2000). Most studies using the ROI approach (Tables 1 and 2) have centred on the superior temporal gyrus after the seminal paper by Barta et al. (1990), with other relevant brain areas being under recognized until the implementation of automated techniques.

A literature review does not show a straightforward relation between GM density alterations, either reductions or enlargements, and the severity or persistence of AH. Although the left superior temporal gyrus has repeatedly been involved in the severity of AH (Table 1), several negative reports (Table 2) have to be considered. Moreover, some studies have reported larger frontal and temporal GM volumes in patients with AH compared with patients without AH (Shin et al., 2005). It is to say that patients included in this study were first-episode psychotic patients and predominantly female (Shin et al., 2005). On the other hand, severity may not imply persistence. Thus, the only other morphometric study on patients with chronic auditory verbal hallucinations, including both genders, did not find volume reductions of temporal lobe structures compared with controls, neither in the hallucinators compared with the non-hallucinators patients (Havermans et al., 1999). The influence of the gender might help to clarify these controversies. In fact, superior temporal gyrus reductions may be more severe in men, while amygdala volumes may exhibit a reverse pattern with reductions in men and enlargements in women (Gur et al., 2000). Due to our series definition, sex effects cannot be analysed in our study. However, our bilateral findings for the superior temporal gyrus are somewhat concordant with a study by Matsumoto et al. (2001) that also gives support to the possible role of the right superior temporal gyrus reduction in the severity of AH.

According to our results, right and left insula and left amygdala may also be implicated on the persistence of AH. Shapleske et al. (2002) obtained results in agreement with these findings, since they found a significant cluster with a deficit in GM which included the left insula and adjacent medial temporal lobe when comparing hallucinators and non-hallucinators (Shapleske et al., 2002).

The insula is a key region of the emotional brain (Aleman and Kahn, 2005) and could also participate in conjunction with the amygdala to foster the chronicity of positive symptoms through emotion-mediated mechanisms. However this structure might be implicated through a different mechanism. One of the most accepted and an investigated cognitive model defends that AH appear because of defective verbal self-monitoring (Frith

and Done, 1988). Neural correlates supporting this model are now being investigated in imaging studies (Woodruff, 2004). The insula is involved in the perception of representations of the self (Farrer and Frith, 2002). Physiologically, a lesser sense of agency (experience of causing an action) would be associated to a reduced activation of both anterior insula (Farrer and Frith, 2002). Our finding on the insular GM reduction may be related to the persistence of AH through abnormalities in the sense of agency and, therefore, the self-monitoring. We obtained positive correlations for both insula GM values with the domain 'beliefs about the origin of voices'. However, the sign of correlations was unexpected (the more GM the higher external attribution) which raises questions on the role of insular cortex and the relationships between functional and structural findings. Thus, we cannot discard a coexistence of greater insular GM volumes with reduced activation, as we have shown for other brain areas with chronically hallucinating patients (Martí-Bonmatí et al., 2007). The rest of our exploratory correlations between specific AH domains and some brain areas also suggest a crucial role for the insula on the persistence of AH. A higher duration of voices was related to bilateral insular reductions while less controllability of voices was associated to left insula GM reductions. Finally, the insula region has also been related to the production of hallucinations in some functional studies (Shergill et al., 2004).

Although its exact role is still unclear, the amygdala is a key component in the emotional processing of patients with schizophrenia (Brunet-Gouet and Decety, 2006). Van Rijn et al. (2005) concluded in their review that amygdalar reduction is a risk factor for schizophrenia and they even proposed amygdala abnormalities as an endophenotype in schizophrenia. Aleman and Kahn (2005) have proposed a model supporting a role for amygdalar hyperactivation in the genesis of positive symptoms. In concordance with this model, in a previous functional MR imaging study with chronic hallucinators, we found a clear overactivation of the frontal lobe, temporal cortex, insula, cingulate, and amygdala in patients when hearing emotional words in comparison with controls (Sanjuán et al., 2007). We hypothesized that the functional overactivation in these patients could be directly related to the emotional response seen in positive symptoms. Aleman and Kahn (2005) also suggested that prolonged hyperactivation of the amygdala during psychotic states at the onset of schizophrenia could result in excitotoxicity due to excessive glutamatergic activity and hence lesions of the amygdala. This hypothesis could explain amygdalar volume reductions in chronically hallucinating patients. Notwithstanding, we only found a reduction in the left amygdala. In this sense, two meta-analyses have suggested greater left than right amygdalar activation to the cognitive processing of emotional stimuli (Wager et al., 2003; Baas et al., 2004). Moreover, the left amygdala seems to be more affected than the right amygdala in patients with schizophrenia (Honea et al., 2005). It has also been hypothesized a neurodevelopmentally mediated alteration of the left amygdala that would predispose to schizophrenia. Keshavan et al. (2002) investigated unaffected, young offspring of schizophrenia patients and reported reduced volumes of the left anterior and posterior amygdalo-hippocampal complex.

4.2. Areas related to the severity of auditory hallucinations

We have observed specific relationships between left inferior frontal and right postcentral gyri reductions and the severity of AH. The left inferior frontal gyrus has previously been related to the pathogenesis of AH. At this point, it can be considered a replicated finding (McGuire et al., 1993; Shergill et al., 2004). Hunter and Spence (2005) suggested that the activation of frontal area could be related not only to the hallucinations but also to the intention to activate the motor area when the button is pressed in functional MRI studies. However, according to the results in this study, it could be considered that the inferior frontal gyrus area is thought to participate in the first stages of the hallucination by the genesis of auditory verbal contents.

Our finding on the postcentral gyrus seems more counterintuitive since this area is known to be primary somatosensory cortex. There is not a clear explanation for a possible relationship between postcentral gyrus and the severity of AH and reasoning on this issue has to be somewhat speculative. Generation of inner speech, which has been linked to the pathogenesis of AH (McGuire et al., 1995), has been associated with activation in the left inferior frontal, the right pre- and postcentral and both superior temporal gyri (Shergill et al., 2002). GM reductions in postcentral gyrus could imply alterations in the monitoring of inner speech but this hypothesis cannot be tested in our study.

Against our predictions, differences in the superior temporal gyrus density were present in group comparisons but did not appear specifically related to AH severity. Note that a type II error cannot be ruled out due to the sample size and the statistical procedures aimed to avoid false positives. Besides, as expected in a homogeneous sample, the variance in PSYRATS scores was low which has surely taken down the intensity of the expected correlations. Moreover, the transient character of AH poses another problem when searching for correlations with presumably more stable GM densities. The existence of chronic antipsychotic treatment, which is known to influence brain morphology, is a possible source of bias. Reductions related to the superior temporal gyrus can reverse after one year of treatment (Keshavan et al., 1998). As far as the sample composition is concerned, two previous studies used comparable samples and, interestingly, none has replicated the superior temporal gyrus findings on AH production (Havermans et al., 1999; Shapleske et al., 2002).

The comparability of studies could pose another major problem, both in regard to the neuroimaging technique and the sample composition. Several studies have compared ROI measurements with VBM (Wright et al., 1999). Volumes were similar for the caudate nuclei but not for temporal lobe measurements. However, Kubicki et al. (2002) showed consistencies for left superior temporal gyrus. Cluster analysis also showed similar results but could not reveal group differences in the medial temporal lobe selected with the ROI analysis. Job et al.'s (2002) results with VBM showed a relative rather than absolute replication of the ROI findings. Two recent studies have replicated previous ROI findings in global terms (Job et al., 2003; Moorhead et al., 2004). Finally, Giuliani et al. (2005) replicated the results of greater left inferior and right superior frontal

cortical GM deficits in schizophrenia. However VBM showed a significantly lower concentration of GM in the middle and superior temporal gyrus and did not lead to replication of the described changes.

4.3. Areas related to the severity of global symptoms

We have also found an association between symptom severity and both left parahippocampus and right superior temporal gyrus GM reductions. Although the parahippocampus has been mainly associated with cognitive abilities in patients with schizophrenia (Antonova et al., 2004), our finding is consistent with previous studies. Prasad et al. (2004) examined the parahippocampal gyrus morphology using structural MR imaging in 33 neuroleptic-naïve subjects with first-episode psychoses and 43 matched healthy subjects. The parahippocampal gyrus volume negatively correlated with total positive symptom, delusion and conceptual disorganization scores on BPRS. The association of right superior temporal gyrus volume reduction with symptom severity but not with AH severity was not expected. We do not have an explanation for this finding since this area is preferably related to AH.

Several study limitations and biases should be considered before establishing definitive conclusions. As all the patients in our series had AH to be included, a group of patients with schizophrenia who had never experienced AH was not within our sample. A study to clarify whether our observed findings are related specifically to the propensity to hallucinated, as opposed to the disorder of schizophrenia itself, should be investigated. Moreover, all patients were medicated with a wide range of drugs, both typical and atypical antipsychotics.

5. Conclusions

Our VBM study confirms the association between AH and structural abnormalities by analysing a highly homogeneous group of persistent hallucinatory patients. We believe this is clearly demonstrated by using a good phenomenological characterization of the AH symptom. Our results only partially confirmed the superior temporal gyrus as being crucial for AH persistence. Other areas, particularly the insula and amygdala, seem to be implicated through yet unclear pathological mechanisms.

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