

# Structural brain correlates of sensorimotor gating in antipsychotic-naive men with first-episode schizophrenia

Trine Bjørg Hammer, MD, PhD; Bob Oranje, PhD; Arnold Skimminge, PhD;  
Bodil Aggernæs, MD, PhD; Bjørn H. Ebdrup, MD, PhD; Birte Glenthøj, MD, DMSc;  
William Baaré, PhD

Hammer, Oranje, Aggernæs, Ebdrup, Glenthøj — Center for Neuropsychiatric Schizophrenia Research & Center for Clinical Intervention and Neuropsychiatric Schizophrenia Research, Psychiatric Center Glostrup, Copenhagen University Hospital, Glostrup, Denmark; Skimminge, Baaré — Danish Research Centre for Magnetic Resonance, Copenhagen University Hospital Hvidovre, Hvidovre, Denmark

**Background:** Prepulse inhibition (PPI) of the startle reflex is modulated by a complex neural network. Prepulse inhibition impairments are found at all stages of schizophrenia. Previous magnetic resonance imaging (MRI) studies suggest that brain correlates of PPI differ between patients with schizophrenia and healthy controls; however, these studies included only patients with chronic illness and medicated patients. Our aim was to examine the structural brain correlates of PPI in antipsychotic-naive patients with first-episode schizophrenia. **Methods:** We performed acoustic PPI assessment and structural MRI (1.5 and 3 T) in men with first-episode schizophrenia and age-matched controls. Voxel-based morphometry was used to investigate the association between PPI and grey matter volumes. **Results:** We included 27 patients and 38 controls in the study. Patients had lower PPI than controls. The brain areas in which PPI and grey matter volume correlated did not differ between the groups. Independent of group, PPI was significantly and positively associated with regional grey matter volume in the right superior parietal cortex. Prepulse inhibition and grey matter volume associations were also observed in the left rostral dorsal premotor cortex, the right presupplementary motor area and the anterior medial superior frontal gyrus bilaterally. Follow-up analyses suggested that the rostral dorsal premotor cortex and presupplementary motor area correlations were driven predominantly by the controls. **Limitations:** We used 2 different MRI scanners, which might have limited our ability to find subcortical associations since interscanner consistency is low for subcortical regions. **Conclusion:** The superior parietal cortex seems to be involved in the regulation of PPI in controls and antipsychotic-naive men with first-episode schizophrenia. Our observation that PPI deficits in schizophrenia may be related to the rostral dorsal premotor cortex and presupplementary motor area, brain areas involved in maintaining relevant sensory information and voluntary inhibition, warrants further study.

## Introduction

Disturbances in attention and information processing are thought to play a pivotal role in the symptomatology of schizophrenia.<sup>1</sup> Patients with schizophrenia are generally unable to focus their attention and are easily distracted by irrelevant stimuli.<sup>2</sup> This incapacity to filter out irrelevant stimuli has been hypothesized to cause an overload of sensory information, which may underlie positive symptoms.<sup>1</sup>

An often-used measure of automatic, preconscious information processing is the prepulse inhibition (PPI) of the startle reflex paradigm. The PPI paradigm is based on an indi-

vidual's startle reflex (i.e., the response to a sudden and intense stimulus).<sup>3</sup> In humans, the startle reflex is usually measured as the amplitude of the electromyographic response of the orbicularis oculi muscle. If the startle-eliciting stimulus is preceded (30–500 ms) by a weak stimulus (i.e., the prepulse), the startle response is usually reduced. This is called sensorimotor gating, or PPI.<sup>4</sup>

Deficient PPI is not only found in patients at all stages and states of schizophrenia,<sup>5–7</sup> but also in those with schizotypal disorder,<sup>7</sup> psychosis-prone individuals<sup>6</sup> and clinically unaffected relatives of patients with schizophrenia.<sup>7</sup> Consequently, this deficit is regarded as an endophenotype for

**Correspondence to:** T.B. Hammer, Center for Neuropsychiatric Schizophrenia Research, University Psychiatric Center Glostrup, Ndr. Ringvej 29-67, DK-2600 Glostrup, Denmark; tbh@cnsr.dk

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schizophrenia related to neurodevelopmental and/or genetic factors.<sup>8</sup>

Animal studies have found PPI to be mediated by the brainstem and regulated by forebrain structures through a cortico–striatal–pallido–thalamic network (for example, see the reviews by Fendt and colleagues,<sup>9</sup> Swerdlow and colleagues,<sup>10</sup> and Geyer and colleagues<sup>11</sup>). Human studies investigating neural brain correlates of PPI are sparse, and conclusions drawn from their findings are constrained by methodological differences (e.g., auditory or tactile PPI paradigms, functional magnetic resonance imaging [fMRI] or structural MRI, inclusion or exclusion of women), methodological limitations<sup>12</sup> and by discrepancies in the results across the studies.<sup>13–19</sup> Nevertheless, PPI has generally been found to be associated with regions within the forebrain: the thalamus; striatum; globus pallidus; hippocampus; insula; and the temporal, parietal and frontal cortices.<sup>13–19</sup>

Many of the aforementioned brain regions supposedly involved in the regulation of PPI have been shown to be affected in patients with schizophrenia. Studies using MRI techniques with region-of-interest (ROI)-based volume measurements have, among others, found grey matter volume changes in the thalamus; basal ganglia; hippocampus; and the temporal, parietal and frontal cortices in medicated patients with chronic schizophrenia.<sup>20</sup> These findings are generally corroborated by findings from studies using voxel-based morphometry.<sup>21–23</sup> Similarly, although more restrained, grey matter volume changes have been reported in patients with early-episode schizophrenia<sup>24,25</sup> and even in individuals at ultra-high risk for psychosis.<sup>26</sup>

Several studies have shown associations between behaviour and cognition and structural properties (e.g., grey matter volume in specific brain regions).<sup>27</sup> The lower PPI in patients could be due to structural abnormalities (reflected in grey matter volume changes) in brain areas contributing to the regulation of PPI (i.e., a quantitative difference between patients and healthy controls), or it could be due to PPI being regulated by less effective areas or networks in patients than in controls (i.e., a qualitative difference). This latter hypothesis is, for instance, supported by studies showing impaired cognition in patients with schizophrenia to be related to abnormal brain networks.<sup>28,29</sup>

To our knowledge, only 2 studies have addressed the association between PPI and grey matter volume. One study included healthy men and women.<sup>18</sup> The other study included medicated men with chronic schizophrenia.<sup>17</sup> Although both studies found PPI to correlate with grey matter volume in areas within the cortico–striatal–pallido–thalamic network, none of the areas found in the study on healthy individuals were confirmed in the patient study. Unfortunately, it is not possible to determine whether the discrepancies between the results of these 2 studies were related to the disease, disease progress, sex differences or medication effects.

The aim of the present study was to examine the structural brain correlates of PPI in patients and healthy controls without the influence of sex, age, antipsychotic medication and chronicity. To this end, we included antipsychotic-naïve men with first-episode schizophrenia and age-matched healthy

controls. We expected PPI to be associated with grey matter volume in the striatum; globus pallidus; thalamus; hippocampus; and the insular, temporal, frontal and parietal cortices. Moreover, we expected these associations to differ in patients and healthy controls.

## Methods

### *Participants*

We recruited 2 cohorts of participants, comprising medication-naïve men with first-episode schizophrenia and age-matched controls each, for inclusion in the study. Cohort A was assessed from 1998 to 2002 and cohort B from 2005 to 2008. The inclusion criteria were identical, and the same measures were obtained from the 2 cohorts; however, the procedures differed as described in the MRI methods section that follows.

We recruited in- and outpatients with schizophrenia-like symptoms from hospitals around Copenhagen, Denmark. All patients were admitted to treatment for the first time and had never received any antipsychotic medication. A schizophrenia diagnosis according to International Classification of Diseases (ICD)-10 criteria was confirmed using the Schedule for Clinical Assessment in Neuropsychiatry (SCAN), version 2.0 or 2.1.<sup>30</sup> Exclusion criteria were coercive measures, a history of mental retardation, organic brain damage or organic psychosis. We assessed psychopathology using the Positive and Negative Syndrome Scale (PANSS).<sup>31</sup> Patients with a history of alcohol or cannabis abuse were not excluded, but urine samples were analyzed for the presence of the following drugs: amphetamines, opiates, cannabis, cocaine, methamphetamine and benzodiazepines. Healthy controls were excluded if they had any history of psychiatric disorder, somatic disease or a history of drug or alcohol abuse, or if their first-degree relatives had a history of psychiatric illness. Healthy controls were screened using the SCAN version 2.0 to exclude major psychopathology. Women were excluded from the current study because PPI fluctuates through the menstrual cycle.<sup>32</sup>

All participants underwent a comprehensive examination program, including psychopathological ratings, psychophysiological<sup>5,33,34</sup> and neuropsychological examinations,<sup>35,36</sup> single-photon emission computed tomography (SPECT)<sup>37</sup> or positron emission tomography,<sup>38,39</sup> structural MRI<sup>40,41</sup> and fMRI.<sup>42</sup> We report only on the combined PPI and structural MRI data.

The ethical committees of Copenhagen and Frederiksberg approved the study. We obtained written informed consent from all participants before inclusion and after oral and written explanation of the study aims and procedures. The individual appraisal of participants' capacity to consent was done by trained psychiatrists and was based on formal contact with the patients during the introduction to the study and continually throughout the study.

### *MRI methods*

High-resolution 3-dimensional (3-D)  $T_1$ -weighted, sagittal magnetization-prepared rapid-gradient echo scans of the

whole head were acquired in the unit of magnetic resonance imaging, University of Copenhagen, Hvidovre Hospital, Hvidovre, Denmark. Cohort A was examined using a 1.5 T Siemens Vision scanner. The  $T_1$ -weighted sequence was as follows: echo time (TE) 4 ms, repetition time (TR) 9.7 ms, flip angle  $12^\circ$ , field of view (FOV) 250 mm, matrix  $256 \times 256$ ,  $0.98 \times 0.98 \times 1$  mm voxels, 170 slices. Cohort B was examined on a 3.0 T Siemens Magnetom Trio scanner. The  $T_1$ -weighted sequence parameters were as follows: TE 3.93 ms, TR 1540 ms, inversion time 800 ms, flip angle  $9^\circ$ , FOV 256 mm, matrix  $256 \times 256$ ,  $1 \times 1 \times 1$  mm voxels, 192 slices.

### Image processing

All acquired images were visually inspected to ensure sufficient quality. Images acquired on the 3 T scanner were corrected for spatial distortions owing to nonlinearity in the gradient system of the scanner.<sup>43</sup> This step was not necessary for images acquired on the 1.5 T scanner. All images were processed using the VBM8 toolbox in SPM8 (<http://dbm.neuro.uni-jena.de/vbm/>; Wellcome Trust Centre for Neuroimaging) with default settings. The VBM8 toolbox incorporates a number of image processing techniques. The processing began with a segmentation algorithm based on a maximum a posteriori technique and a partial volume estimation method,<sup>44,45</sup> including estimation of parameters for affine transformation to Montreal Neurological Institute (MNI) space. Furthermore, we applied 2 denoising methods: a spatially adaptive nonlocal means denoising filter<sup>46</sup> and a Markov random field method.<sup>44</sup> The  $T_1$ -images were warped to the VBM8 template of 550 healthy adult controls using DARTEL through use of the calculated segmentations. Note that the VBM8 template itself also was created using DARTEL. Using the flow fields that parameterize the deformations, grey and white matter and cerebrospinal fluid images were warped into MNI space. Images were modulated with the Jacobian determinant of the applied deformation fields to correct for local volume changes following the high dimensional intersubject warping. We calculated Jacobian determinants from the nonaffine components of the applied deformation fields only, such that the resulting tissue maps were corrected for differences in head size. Finally, we smoothed the resulting warped and modulated tissue images using an 8 mm full-width at half-maximum Gaussian kernel. The quality of the images generated at each processing stage was visually checked.

### PPI assessments

All participants were screened for hearing deficits with 40 dB(A) stimuli at 500, 1000 and 6000 Hz, and no hearing deficits were found. Participants were asked to not smoke or to drink any caffeinated beverages in the hour preceding the testing. Participants were seated in an armchair and instructed to sit still and fix their gaze at a point straight ahead of them. The eye-blink component of the acoustic startle response was measured by recording electromyographic activity from the right orbicularis oculi muscle. The acoustic stimuli were presented binaurally.

We used 2 slightly different PPI paradigms, which have been described in detail elsewhere.<sup>5,33,34</sup> Briefly, we used para-

digms A<sup>5,34</sup> to assess all participants from cohort A. It consisted of 73 trials of pulse alone and prepulse-pulse trials. Pulse alone (PA) stimuli were 40 ms of 116 dB(A) broad frequency spectrum noise. Prepulse-pulse trials were PA trials preceded by a prepulse stimulus (20 ms, 85 dB(A) broad frequency spectrum noise). The prepulse stimulus was presented with a 30, 60 or 120 ms stimulus onset asynchrony (SOA) before the PA stimulus. The first trial was a PA trial, which was followed by 6 blocks of 12 trials. Each block consisted of 3 PA trials and 3 of each prepulse-pulse trial type (PPI30, SOA = 30 ms; PPI60, SOA = 60 ms; PPI120, SOA = 120 ms) presented in a pseudorandomized order. We used paradigm B<sup>33</sup> to assess all participants from cohort B. It consisted of 66 trials presented in 3 experimental blocks. Blocks 1 and 3 consisted of 8 PA trials (20 ms broad frequency spectrum noise of 115 dB(A)). Block 2 consisted of 10 PA trials and 40 prepulse-pulse trials. The prepulse stimuli had an intensity of 76 or 85 dB(A) and consisted of 20 ms of broad frequency spectrum noise. The SOAs were either 60 or 120 ms. Each of the 4 prepulse-pulse combinations contained 10 trials and were presented in a pseudorandomized order. In both paradigms, PPI was expressed as  $[1 - (\text{mean startle amplitude to prepulse-pulse trials}) / (\text{mean startle amplitude to PA trials})] \times 100\%$ .

Common to both paradigms was the PPI60 and PPI120 with a 85 dB(A) prepulse. However, only the PPI120/85 dB(A) trial type was used in the PPI and MRI analysis, since this trial type consistently elicits higher PPI than the PPI60/85 dB(A) trial type (for example, see Mackeprang and colleagues,<sup>5</sup> Aggernaes and colleagues,<sup>33</sup> Braff and colleagues<sup>47</sup>). Furthermore, numerous studies have found PPI120/85 dB(A) to be lower in patients with schizophrenia than in healthy controls.<sup>5,48,49</sup>

We combined PPI derived from paradigms A and B with MRI scans acquired on the 1.5 and 3 T scanners, respectively.

### Statistical analysis

We used SPSS for Windows, version 18.0, for statistical analysis of demographic and clinical variables and PPI. The  $\alpha$  level for significance was set at  $p < 0.05$ , 2-tailed. Age, PPI120 and PANSS scores were normally distributed (Kolmogorov-Smirnov test). Group differences in age were tested using independent-sample Student *t* tests. Differences in handedness were tested with the Fisher exact test. The same tests were performed to assess differences between participants in cohorts A and B. To investigate differences in PPI between groups (patients, healthy controls) and PPI paradigms, we performed an analysis of variance with PPI120 as a dependent factor and group and paradigm/cohort as between-subject factors. We analyzed correlations between PPI and psychopathology with Pearson correlation tests.

### Associations between PPI and grey matter volumes

We performed the voxel-wise statistical analyses in SPM8. The  $\alpha$  level for significance was set at  $p < 0.05$ , family-wise error (FWE)-corrected. To avoid type II errors, we also considered voxels surviving a significance level of  $p < 0.001$ , uncorrected for multiple comparisons with an extent threshold,

empirically determined by SPM8 to be of interest. Liebermann and Cunningham<sup>50</sup> recommended a combined intensity and cluster size threshold since it has been demonstrated to provide a good balance between risk of type I and type II errors. Only voxels with more than 10% grey matter volume were included to diminish possible partial volume effects at the borders between grey and white matter and between grey matter and cerebrospinal fluid.

Since we hypothesized PPI to be associated with grey matter in the striatum; thalamus; hippocampus; and insular, frontal, temporal and parietal cortices, we applied a mask only including these regions to restrict our statistical search area and thereby the amount of multiple comparisons. The mask was created with the MARINA software (Masks for Region of INterest Analysis, version 0.6.1, Bertram Walter Bender Institute of Neuroimaging, University of Giessen).<sup>51</sup>

We used flexible factorial design to test for PPI120 and grey matter volume associations. Group and scanner type (and thus paradigm type and cohort) were included as between-subject factors, and PPI120, age and scanner drift (number of days passed since the first scan of the study) were included as covariates. An interaction was set for group and PPI120. Four statistical parametric maps were generated: 1 for the positive correlation between PPI120 and grey matter volume in both patients and controls, 1 for the negative correlation between PPI120 and grey matter volume in both patients and controls, and 2 for the interactions between patients and controls. With respect to the interactions between patients and controls, we expected that any effects would be found in areas where PPI and grey matter volume were associated in healthy controls. Therefore, when testing for interaction effects, we restricted our statistical search area to those brain regions where controls exhibited PPI120–grey matter volume correlations ( $p < 0.05$ , no extent threshold).

Planned follow-up single-group analyses were performed to examine PPI120–grey matter volume correlations separately for patients and controls. The design matrices included PPI120, scanner type, age and scanner drift as covariates. For each group, 2 statistical parametric maps were generated: 1 for the positive correlation and 1 for the negative correlation between PPI120 and grey matter volume.

### Correlations between volumetric measures of the associated brain areas and PPI

Grey matter volume estimates were extracted from the 5 clusters of voxels that showed PPI120–grey matter volume correlations in patients and controls by integrating the grey matter intensities within each cluster. The grey matter volume estimates used regression analysis to examine how much of the total variance in PPI (dependent factor) could be explained by the joint grey matter volume estimates. Finally, we calculated correlations between PPI120 and grey matter volume estimates for the individual anatomic areas separately using Pearson correlations.

To address a possible scanner effect, we used flexible factorial design to test for a main effect of scanner and scanner  $\times$  PPI interaction effects. Group and scanner type were included as between-subject factors, and PPI120, age and scan-

ner drift were included as covariates. An interaction was set for scanner and PPI120. Besides addressing possible scanner effects, the aforementioned analyses were also sensitive for possible anatomic differences between cohorts.

## Results

### *Demographic and clinical characteristics of participants*

In all, 32 medication-naive men with first-episode schizophrenia and 42 healthy men underwent a PPI assessment and structural MRI (scanning to PPI assessment interval was mean 13.7 [range 0–118] d; 2 patients and 4 healthy controls had an interval of more than 1 month). After assessment, we excluded 2 patients and 1 control because they were non-responders on the PPI paradigm (startle amplitude  $< 10 \mu\text{V}$ ); 1 patient and 2 controls because they had outlying PPI values (standard deviation [SD]  $> 2.5$  from the mean); and 2 patients and 1 control because of artifacts on their MRI scans. Thus, 27 patients and 38 healthy controls were included in the final analysis. Of these, 11 patients and 12 controls were in cohort A, and 16 patients and 26 controls were in cohort B. Demographic and clinical characteristics of participants are listed in Table 1.

Patients and controls in the 2 cohorts did not differ significantly in age or handedness. Moreover, the patients in cohorts A and B did not differ in benzodiazepine prescribed or substance abuse. None of the participants tested positive on urine screening for substance use, and none had clinically important brain pathology. The 2 patient cohorts did not differ on PANSS negative and positive scores; however, the PANSS general score was significantly lower in cohort A than B (29.9 v. 40.0, respectively; Table 1). Finally, most likely as a result of their lower PANSS general score, there was a trend for a lower PANSS total score among patients from cohort A compared with those from cohort B (72.4 v. 81.6, respectively; Table 1).

### *Prepulse inhibition*

We found a significant effect of group, indicating that patients had significantly lower PPI than healthy controls ( $F_{1,61} = 4.06$ ,  $p = 0.048$ ). We did not observe an effect of paradigm/cohort ( $F_{1,61} = 0.17$ ,  $p = 0.68$ ), nor did we find an interaction between paradigm/cohort and group ( $F_{1,61} = 0.04$ ,  $p = 0.84$ ). No significant correlations were found between PANSS positive, negative, general or total scores and PPI120 (all  $p > 0.16$ ).

### *Associations between PPI and grey matter volume*

Results of the voxel-wise analysis are presented in Table 2. There was no significant interaction between group and PPI120, indicating that patients and controls did not differ with respect to PPI120–grey matter volume associations. However, independent of diagnosis, PPI120 was significantly and positively associated with grey matter volume in a cluster in the right superior parietal lobe ( $p = 0.006$ , FWE-corrected). Moreover, although not significant at an FWE-corrected level, clusters of interest (i.e., voxel level significance of  $p < 0.001$ ,

extent threshold 88 voxels) were observed in the right superior parietal lobe, the right presupplementary motor area, the left rostral dorsal premotor cortex and the anterior medial superior frontal lobe bilaterally (Fig. 1).

Planned follow-up analyses revealed that in healthy controls, when analyzed separately, PPI120 was positively associated with grey matter volume in the right superior parietal lobe, presupplementary motor area and the left rostral dorsal premotor cortex. Separate analyses of patients showed that PPI was positively associated with grey matter volume in the right

superior parietal lobe only. Further lowering the voxel-level threshold to 0.01 (no extent threshold) revealed additional positive associations between PPI120 and bilateral anterior medial superior frontal grey matter in patients and healthy controls. No negative associations were observed in the analyses.

In addition, both data sets were analyzed separately (independent of group,  $p < 0.005$ , uncorrected). All associations found in the analyses of the combined data set were confirmed. Details are presented in Appendix 1, Table S1, available at [cma.ca/jpn](http://cma.ca/jpn).

**Table 1: Demographic and clinical characteristics and prepulse inhibition of medication-naïve men with first-episode schizophrenia and age-matched healthy controls\***

Characteristic	Patients with schizophrenia			Controls		
	Cohort A†	Cohort B†	<i>p</i> value	Cohort A†	Cohort B†	<i>p</i> value
<b>Methodological features</b>						
No. participants	11	16	—	12	26	—
Scanner type	1.5 T	3 T	—	1.5 T	3 T	—
PPI paradigm‡	A	B	—	A	B	—
<b>Demographic and clinical data</b>						
Age, mean (SD) yr	29.3 (1.9)	25.7 (1.2)	0.19	28.4 (5.1)	26.6 (5.3)	0.34
Handedness, right/left	9/2	16/0	0.16	9/3	25/1	0.09
Current substance abuse, yes/no§	3/7	2/14	0.37	0/12	0/9	—
Benzodiazepine, yes/no¶	1/10	5/11	0.18	0/12	0/9	—
Antipsychotics, yes/no**	0/11	1/15	0.62	0/12	0/9	—
Smoking status, yes/no††	3/6	10/4	—	Unknown	3/23	—
<b>Psychopathology PANSS, mean (SD) score</b>						
Positive	21.1 (4.1)	19.9 (4.1)	0.46	—	—	—
Negative	21.4 (4.5)	21.7 (5.1)	0.87	—	—	—
General	29.9 (4.6)	40.0 (8.4)	0.021	—	—	—
Total	72.4 (10.8)	81.6 (14.2)	0.08	—	—	—
<b>Percentage PPI, mean (SD)</b>						
PPI120/85 dB(A)	62.1 (7.0)	60.9 (6.4)	0.91	75.0 (6.8)	71.4 (3.6)	0.61

ICD = International Classification of Diseases; PANSS = Positive and Negative Syndrome Scale;<sup>31</sup> PPI = prepulse inhibition; SD = standard deviation.  
 \*No significant differences were observed between the 2 patient populations (except for PANSS general score) or between the 2 healthy control populations.  
 †We recruited 2 cohorts of participants, each comprising medication-naïve men with first-episode schizophrenia and age-matched. Cohort A was assessed from 1998 to 2002 using a 1.5 T scanner and cohort B was assessed from 2005 to 2008 using a 3 T scanner. The 2 cohorts did not differ significantly with regard to the ratio of patients and controls (Fisher exact test,  $p = 0.6$ ).  
 ‡Paradigm A has been described elsewhere by Mackeprang and colleagues<sup>5</sup> and Hammer and colleagues;<sup>34</sup> paradigm B has been described elsewhere by Aggernaes and colleagues.<sup>33</sup>  
 §Substance abuse as determined by ICD-10.  
 ¶Prescription of benzodiazepine.  
 \*\*Treatment with antipsychotics.  
 ††Smoking status was not available for all participants.

**Table 2: Structural brain correlates of prepulse inhibition with a stimulus onset asynchrony of 120 ms in medication-naïve men with first-episode schizophrenia and healthy controls\***

Anatomic area	Lobe	Side	Controls and patients, $n = 65$ †					Controls, $n = 38$ ‡					Patients, $n = 27$ §				
			MNI coordinates			Cluster size	<i>t</i> value	MNI coordinates			Cluster size	<i>t</i> value	MNI coordinates			Cluster size	
			<i>x</i>	<i>y</i>	<i>z</i>			<i>x</i>	<i>y</i>	<i>z</i>			<i>x</i>	<i>y</i>	<i>z</i>		
Superior (7A)	Parietal	R	33	-66	58	5.8¶	589	33	-60	57	4.9	140	30	-66	60	5.6	220
	Parietal	R	9	-67	51	3.8	116										
Pre-SMA	Frontal	R	4	23	61	3.9	321	4	24	64	5.11	508					
rPMd	Frontal	L	-21	3	72	4.3	237	-21	3	67	3.77	110					
Medial superior	Frontal	R/L	0	52	15	3.9	122										

L = left; MNI = Montreal Neurological Institute; R = right; rPMd = rostral dorsal premotor cortex; SMA = supplementary motor area.  
 \*Statistical parametric maps were thresholded at  $p < 0.001$  with an extent threshold, empirically determined by SPM8. Only positive correlations were found. All analyses were corrected for scanner type, age and scanner drift.  
 †Extent threshold of  $k_e = 88$  voxels.  
 ‡Extent threshold of  $k_e = 75$  voxels.  
 §Extent threshold of  $k_e = 81$  voxels.  
 ¶ $p = 0.006$ , family-wise error-corrected.

**Grey matter volume estimates and their correlations with PPI**

The cumulated grey matter volume estimates of the areas that were positively associated with PPI120 in the main analysis — the right superior parietal cortex, right presupplementary motor area, left rostral dorsal premotor cortex and bilateral anterior medial superior frontal lobe — explained 67% of the variance in PPI among healthy controls ( $R^2 = 0.67$ ,  $F_{4,33} = 16.83$ ,  $p < 0.001$ ) and 47% of the variance in PPI among patients ( $R^2 = 0.47$ ,  $F_{4,22} = 4.85$ ,  $p = 0.006$ ).

The Pearson correlation for PPI120 and grey matter volume estimates in the right superior parietal cortex in patients was 0.67 ( $p < 0.001$ ). The Pearson correlations for PPI120 and grey matter volume estimates in the right superior parietal cortex, right presupplementary motor area and rostral dorsal premotor cortex in healthy controls were 0.75 ( $p < 0.001$ ), 0.62 ( $p < 0.001$ ) and 0.45 ( $p = 0.005$ ), respectively.

*Supplementary analyses***Effect of scanner and scanner  $\times$  PPI interactions**

We did not find a significant main effect of scanner ( $p < 0.05$ ,

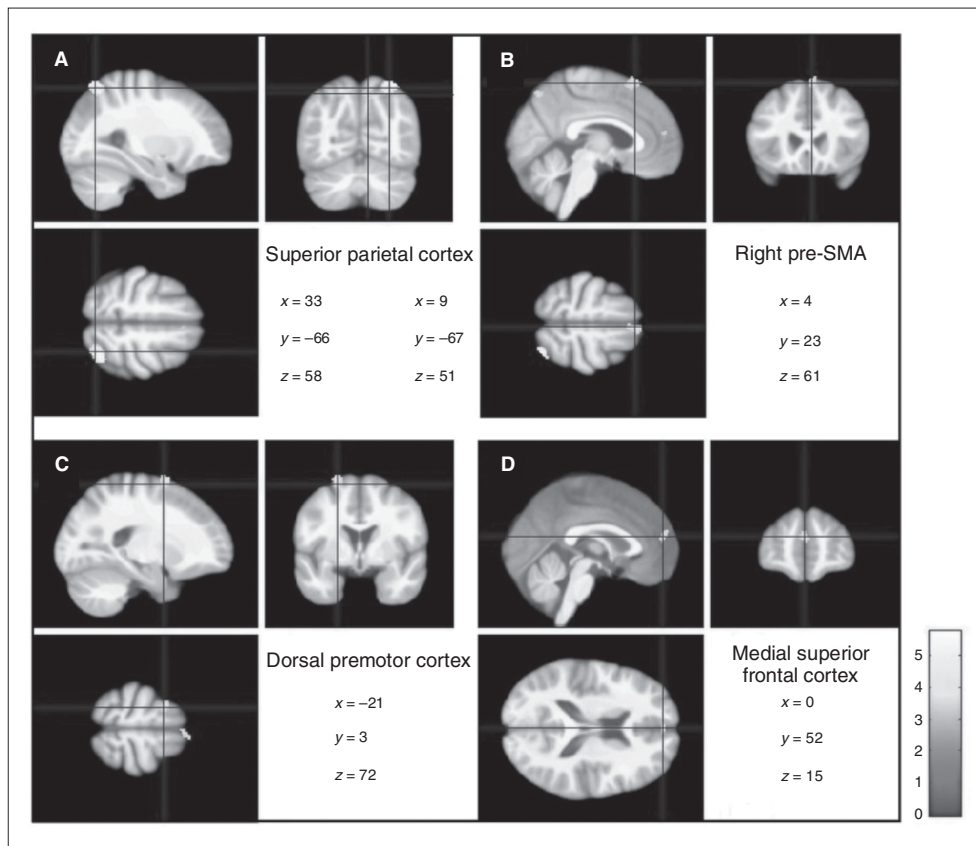
FWE-corrected); however, using a liberal voxel-level threshold ( $p < 0.01$ , uncorrected, no extent threshold) showed a possible scanner effect in various regions: the cerebellum, primary motor area, nucleus caudate and other subcortical regions. These regions did not overlap with brain areas showing PPI–grey matter volume correlations (Appendix 1, Figs. S1–S4). Likewise, we did not find a significant scanner  $\times$  PPI interaction effect ( $p < 0.05$ , FWE-corrected); however, using a liberal voxel threshold ( $p < 0.01$  uncorrected, no extent threshold) showed a possible scanner  $\times$  PPI interaction in various regions. Again, these regions did not overlap with the brain areas showing PPI–grey matter volume correlations (Appendix 1, Figs. S5–S8).

**Effect of substance abuse**

Five patients reported current substance abuse. Excluding these 5 patients did not significantly change the PPI results or the PPI–grey matter volume correlation results.

**Effect of extended scanning to PPI assessment interval**

Excluding the 6 participants with a scanning to PPI assessment



**Fig. 1:** Structural brain correlates of prepulse inhibition (PPI) with a stimulus onset asynchrony of 120 ms (PPI120) in patients with schizophrenia and healthy controls. Statistical parametric maps of the positive correlations between grey matter volume and PPI120 independent of diagnosis ( $p < 0.001$ , extended threshold  $k_E = 88$ ) show the (A) right superior parietal cortex, (B) right presupplementary motor area, (C) left rostral dorsal premotor cortex and (D) bilateral anterior medial superior parietal cortex. Neurological (right is right) convention is used. Results are projected on an average of all DARTEL warped magnetization-prepared rapid-gradient echo images.

interval of more than 1 month did not significantly change the PPI–grey matter volume correlation results.

## Discussion

To our knowledge, this is the first study in which the association between grey matter volume and PPI of the startle reflex was investigated in antipsychotic-naïve men with first-episode schizophrenia and healthy age-matched controls. Independent of group, PPI120 correlated significantly and positively with grey matter volume in the right superior parietal cortex and also positively with 3 areas in the frontal cortex: the right presupplementary motor area, left rostral dorsal premotor cortex and bilateral anterior medial superior frontal gyrus. Follow-up analyses could not confirm the presence of presupplementary motor area and rostral dorsal premotor cortex correlations in the patient group even when using a very liberal threshold of significance. The latter suggests that these frontal correlations may have been driven predominantly by the healthy controls, whereas the correlation within the superior medial frontal gyrus was driven by both patients and controls. We did not observe any brain structure–PPI correlates distinct among patients.

Our finding of an association between PPI and grey matter volume in the cortical regions is in line with results from animal studies<sup>9–11</sup> and human psychophysiological studies that indicate top–down cortical modulation of PPI. The latter is indicated since levels of PPI increase when cortical-emanated functions (e.g., attention, emotion) are activated during a PPI paradigm.<sup>52,53</sup>

To our knowledge, only 2 MRI studies have previously reported on structural brain correlates of PPI.<sup>17,18</sup> Kumari and colleagues<sup>18</sup> examined healthy individuals only and found PPI–grey matter volume correlations in the striatum, hippocampus and inferior frontal gyrus. The present study did not confirm correlates in any of these 3 brain areas. In a later study, Kumari and colleagues<sup>17</sup> examined structural MRI in 42 chronically ill and medicated male patients with schizophrenia and, in contrast to the present study, found PPI to be associated with frontal cortical areas (i.e., left dorsolateral and right medial prefrontal cortex, right middle frontal cortex), but not the parietal cortex. Discrepancies between the findings of these 2 studies and the present study are most likely rooted in demographic, clinical (including medication) and methodological factors. Kumari and colleagues<sup>18</sup> included both healthy men and women, whereas only men were included in the present study. Sex differences are found not only in levels of PPI (where men usually score higher than women), but also in brain structure and brain activation during cognitive tasks.<sup>54–57</sup> Moreover, the level of PPI appears to fluctuate during the female menstrual cycle.<sup>32</sup> Furthermore, in contrast to Kumari and colleagues,<sup>17</sup> we investigated antipsychotic-naïve patients with first-episode schizophrenia, thereby diminishing possible effects of disease chronicity, hospital admissions and medication. Finally, we used a high-dimensional inter-subject warping method and adjusted for differences in brain sizes, thereby diminishing inter-subject anatomic variability.

Although future studies are needed to confirm our findings, the superior parietal cortex is known to be involved in detecting or ignoring irrelevant stimuli.<sup>58</sup> The presupplementary motor area and rostral dorsal premotor cortex are a part of the frontal premotor areas, but they are primarily involved in the more cognitive aspects of motor control (e.g., in the maintenance of relevant sensory information).<sup>59</sup> The functions of the area we found in the anterior medial superior frontal gyrus are not well described; however, midline–frontal areas in general are found to be involved in voluntary response inhibition.<sup>60</sup>

Although functions of relevance for schizophrenia (e.g., feeling of self-agency, mirror neuron properties) seem to originate from the presupplementary motor area and rostral dorsal premotor cortex, only a few studies on schizophrenia have focused on these structures. However, the volume,<sup>61</sup> integrity<sup>62</sup> and functions<sup>63</sup> emanating from the rostral dorsal premotor cortex and the presupplementary motor area have been found to be altered in patients with schizophrenia compared with healthy controls.

The grey matter volume of the 4 cortical areas (2 clusters in the parietal area and 3 clusters in the frontal areas) that we found to be associated with PPI explained 47% and 67% of the variance in PPI in antipsychotic-naïve patients and healthy controls, respectively. Likewise, Kumari and colleagues<sup>17</sup> found that brain regions associated with PPI explained about 40% of the variance in PPI in medicated patients with chronic schizophrenia and 62% of the variance in PPI in healthy men and women.<sup>18</sup> These observations led them to suggest that there may be a stronger structure–function correspondence in healthy people than in patients with schizophrenia.<sup>17</sup>

## Limitations

The present study has some limitations. Although the number of antipsychotic-naïve patients studied was considerable given the difficulties of recruiting such a patient group, we cannot rule out that we did not have enough statistical power to observe any group differences. In addition, 2 different MRI scanners were used. Analyses testing for possible scanner effects, and thus implicitly also testing for possible anatomic differences between cohorts, revealed that brain areas showing a main effect of scanner or a scanner × PPI interaction effect, which was only apparent using liberal levels of significance, did not overlap with brain areas showing PPI–grey matter volume correlations. It is therefore unlikely that the use of 2 different scanners has affected our findings. Nevertheless, the use of 2 different scanners might especially have limited our ability to find possible associations between PPI and subcortical regions. Indeed, interscanner consistency has frequently been found to be low for subcortical regions (e.g., thalamus, basal ganglia) but high for cortical regions.<sup>64,65</sup> Finally, the fact that PPI was derived from 2 somewhat different PPI paradigms could have affected the PPI–grey matter volume associations by introducing variability to the data. However, we used paradigm type (A or B) as a covariate in all analyses, and PPI levels in the 2 paradigms did not differ significantly. Significantly more patients than controls were

smokers, and although we tried to avoid the acute effect of nicotine by requesting that participants not smoke in the hour before the PPI assessment, we cannot rule out that some of the differences found between patients and healthy controls were due to differences in smoking status. However, because smoking is found to increase PPI, and since PPI was lower in the group that smoked the most (i.e., the patients), we think it unlikely that observed group differences resulted from the effects of nicotine.

## Conclusion

The present data indicate that the superior parietal cortex is involved in the regulation of PPI in both antipsychotic-naïve men with first-episode schizophrenia and healthy age-matched controls. Although the patients showed significantly lower levels of PPI than the controls, no significant group differences were found in the associations between grey matter volume and PPI. Our data did, however, suggest that the right presupplementary motor area and left rostral dorsal motor cortex grey matter values were more strongly associated with PPI in healthy controls than in patients. This could indicate a possible link between dysfunctions within the frontal premotor areas and PPI deficits in patients with schizophrenia.

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