Brain Imaging and Behavior (2020) 14:1350–1360
https://doi.org/10.1007/s11682-019-00040-8

ORIGINAL RESEARCH

Aberrant resting-state functional connectivity of salience network in first-episode schizophrenia

Huan Huang[1,2] · Zeng Botao[3] · Yuchao Jiang[4] · Yingying Tang[2] · Tianhong Zhang[2] · Xiaochen Tang[2] · Lihua Xu[2] · Junjie Wang[2] · Jin Li[2] · Zhenying Qian[2] · Xu Liu[2] · Huiling Wang[1] · Cheng Luo[4] · Chunbo Li[2,5,6] · Jian Xu[7] · Donald Goff[8] · Jijun Wang[2,5,6]

Published online: 28 January 2019

Abstract

The disruption of salience network (SN) has been consistently found in patients with schizophrenia and thought to give rise to specific symptoms. However, the functional dysconnectivity pattern of SN remains unclear in first-episode schizophrenia (FES). Sixty-five patients with FES and sixty-six health controls (HC) were enrolled in this study and underwent resting-state functional magnetic resonance imaging (rs-fMRI). The eleven regions of interest (ROIs) within SN were derived from the peaks of the group independent component analysis (gICA). Seed-based whole-brain functional connectivity (FC) analyses were performed with all SN ROIs as the seeds. Both hyper- and hypo-connectivity of SN were found in the FES. Specifically, the increased FC mainly existed between the SN and cortico-cerebellar sub-circuit and prefrontal cortex, while the reduced FC mainly existed within cortico-striatal-thalamic-cortical (CSTC) sub-circuit. Our findings suggest that FES is associated with pronounced dysregulation of SN, characterized prominently by hyperconnectivity of SN-prefrontal cortex and cerebellum, as well as hypoconnectivity of CSTC sub-circuit of the SN.

Keywords First-episode schizophrenia · Resting-state functional magnetic resonance imaging · Salience network · Functional connectivity

Introduction

The underlying etiology of schizophrenia remains largely unclear. It is generally thought to involve alterations in neural circuits that support sensory, cognitive, and emotional processes (Stephan et al. 2009; Pettersson-Yeo et al. 2011), which could better explain the range of various impairments seen in this illness. Resting-state functional magnetic resonance imaging (rs-fMRI) is an excellent non-invasive tool to explore brain in vivo; it is easy to implement and has been

Electronic supplementary material The online version of this article (https://doi.org/10.1007/s11682-019-00040-8) contains supplementary material, which is available to authorized users.

✉ Jian Xu
xujianr@163.com

✉ Jijun Wang
jijunwang27@163.com

[1] Department of Psychiatry, Renmin Hospital of Wuhan University, Wuhan 430060, China

[2] Shanghai Key Laboratory of Psychotic Disorders, Shanghai Mental Health Center, Shanghai Jiaotong University School of Medicine, Shanghai 200030, China

[3] Department of Psychiatry, Qingdao Mental Health Center, Qingdao, China

[4] Key Laboratory for NeuroInformation of Ministry of Education, School of Life Science and Technology, University of Electronic Science and Technology of China, Chengdu 610054, China

[5] CAS Center for Excellence in Brain Science and Intelligence Technology (CEBSIT), Chinese Academy of Science, Beijing, China

[6] Brain Science and Technology Research Center, Shanghai Jiao Tong University, Shanghai 200030, China

[7] Department of Neurology, Nantong University Affiliated Mental Health Center, Nantong 226005, Jiangsu, China

[8] Department of Psychiatry, New York University Langone Medical Center, One Park Ave, Room 8-212, New York, NY 10116, USA

widely applied to assess the relationships of spontaneous low-frequency (<0.1 Hz) neural activity between different regions of the brain during rest in the clinical population. The main inference of functional connectivity (FC) is that, if two brain regions have highly temporal correlated blood-oxygen-level-dependent (BOLD) signals then they are more likely to be communicating with one another and therefore are within the same functional network. The growing evidence from rs-fMRI studies has indicated that schizophrenia has been regarded as a disorder with deficits involving large-scale brain functional networks, and salience network (SN) are increasingly considered as having a fundamental role in the pathophysiology of the disorder (Peters et al. 2016; Wang et al. 2016a; Palaniyappan and Liddle 2012; Palaniyappan et al. 2012; Kapur 2003).

The SN is a large-scale paralimbic-limbic functional network anchored to the anterior insula (AI) and dorsal anterior cingulate cortex (dACC), as well as the anterior prefrontal cortex (APFC), the supramarginal gyrus (SMG), the striatum/basal ganglion, the thalamus and the cerebellum (Dosenbach et al. 2007; Seeley et al. 2007; Dosenbach et al. 2006; Menon 2015). Meanwhile, a network called cingulo-opercular network (CON) also comprises the AI, anterior prefrontal cortex, dorsal ACC, and thalamus (Damoiseaux et al. 2006). As we have seen, the SN and CON overlap anatomically, and it remains unclear whether they constitute separate or entities or are merely different descriptions of the same network (Menon 2015). However, most studies regard SN and CON as the same network that anchors on AI and ACC(P. C. Tu et al. 2012; Han et al. 2018), and so does this study. By performing a graph analysis, Tu et al. conclude that the network is composed of a core cortical network interconnected with cortico-striatal-thalamic-cortical (CSTC) and cortico-cerebellar sub-circuits(P. C. Tu et al. 2012). The SN has been shown to be involved in detecting, processing, and integrating internal and external salient information, as this network is commonly activated across a wide range of cognitive and affective tasks (Sridharan et al. 2008; Uddin et al. 2010). In addition, SN was also found to play a crucial role in controlling interactions between default mode network (DMN) and central executive network (CEN); it acts as a "dynamic switch", which initiates transient control signals that engage the CEN and disengage the DMN to mediate the cognitive control process when a salient external event is detected (Dosenbach et al. 2007).

Given that the SN is a unique hub for driving inter-network interactions, it is perhaps not surprising that dysfunction of the network is increasingly believed to underlie the observed clinical symptoms and cognitive impairments in schizophrenia. Indeed, previous neuroimaging studies have provided preliminary evidence that abnormalities of SN are associated with schizophrenia. A recent meta-analysis found that, of large-scale brain networks, SN plays the crucial role in this disorder. Its impaired connectivity with other functional networks may underlie the core difficulty of patients to differentiate self-representation (inner world) and environmental salience processing (outside world) (Dong et al. 2017). A number of imaging studies have illustrated structural and functional alterations of SN in schizophrenia. The most consistent findings include the most significant reduction in gray matter volume (Glahn et al. 2008), as well as reduced task-related activation(P. Tu et al. 2010), of bilateral insula and ACC in schizophrenia. Nevertheless, the findings regarding the SN connectivity abnormalities are variable across studies and yielded mixed and inconsistent results. For example, some studies have found reduced connectivity of SN in schizophrenia(P. C. Tu et al. 2012); whereas other studies have reported a mixed pattern of increased and decreased connections within the SN (White et al. 2013).

In fact, the patients with schizophrenia recruited in most studies were clinically stable, chronic, and antipsychotic medicated, therefore, the study of patients during their first episode schizophrenia (FES) would be of high importance to identify SN neural signatures of the illness by limiting the influence of illness burden and medication exposure (Mikolas et al. 2016; Guo et al. 2015b). Also, studies of early biomarkers of illness may contribute to early diagnosis and intervention, as well as possible preventive measures. Additionally, most of these studies applied prior-determined regions of interest (ROI) analyses based on previous studies and/or anatomical knowledge, so results must be highly limited to the chosen ROIs, and to the spatial definition of the ROIs. Obviously, a data-driven ROI-defined approach would be better to avoid such bias.

In the current study, a relatively large sample of patients with FES($n = 65$) was enrolled and underwent rs-fMRI. We applied a data-driven independent component analysis (ICA) to define the SN ROIs and compared the resting-state FC differences of SN ROIs between the FES patients and the healthy controls. Further, we also explored the relationships between FC abnormalities of SN and clinical variables in FES, to identify putative neural correlates of clinical variables in schizophrenia.

Methods and materials

Participants

Sixty-five patients with FES were recruited from the Shanghai Mental Health Center (SMHC), Shanghai, China, from March 2013 to October 2014. The patients were diagnosed with schizophrenia or schizophreniform psychosis by trained clinical psychiatrists using the SCID-I/P (Structural Clinical Interview for DSM-IV-TR, Patient's version) and met the following inclusion criteria: (1) 16–40 years of age; (2) right-handed; (3) at their first acute episode. Sixty-five patients with

no history of exposure to antipsychotics were recruited. Due to the time of patients and availability of MRI scanner, 11 of 65 patients were medicated with one or more second-generation antipsychotics by their clinicians according to routine clinical practice for less than one week (mean 3.1 days) prior to MRI scans. Daily antipsychotic medication dosage was converted to chlorpromazine equivalents (mg/d) (Leucht et al. 2015; Leucht et al. 2014). Nine patients were treated with risperidone (mean 4.82 ± 2.11 mg/d), seven with olanzapine (mean 14.22 ± 3.35 mg/d), six with aripiprazole (mean 16.55 ± 6.31 mg/d), two with amisulpride (mean 989.47 mg/d and 587.23 mg/d) and one with quetiapine (mean 200.00 mg/d).

Sixty-six healthy control subjects (HC) were recruited from the local community through advertisements and were matched to the FES group by gender, age, and education. The main exclusion criteria for HC were a history of any psychiatric disorder. We also excluded subjects with a family history of psychosis in their first-degree relatives to reduce the potential effect of similar genetic backgrounds.

Further exclusion criteria, for both the FES group and HC group, included current or a history of neurological disorders, a history of substance abuse or dependence, a history of mental retardation, and any contraindications for MRI scanning.

The Ethics Committee of SMHC approved the study protocol. The written, informed consent of all subjects was obtained after receiving a complete description of the study.

Clinical assessments

All patients underwent clinical assessment of psychopathology using the 24-item Brief Psychiatric Rating Scale (BPRS) Expanded Version and the Scale for Assessment of Negative Symptoms (SANS) (Andreasen 1989) at the time of screening. Duration of untreated psychosis (DUP) and clinical symptom ratings were obtained by a trained psychiatrist before MRI scan.

Imaging data acquisition

Whole-brain imaging data for all subjects were acquired on a 3.0 T Siemens Verio MRI scanner (Erlangen, Germany) with a 32-channel head coil. The subjects were instructed to keep their eyes closed, relax, and lie as still as possible and not to focus their thoughts on anything in particular.

Functional MRI data were collected using a gradient echo planar imaging (EPI) sequence (repetition time [TR] = 3000 ms; echo time [TE] = 30 ms; flip angle = 90°; field of view [FOV] = 220 mm × 220 mm; matrix = 64 × 64; 45 slices; slice thickness = 4 mm; gap = 0.6 mm, resulting in 170 volumes to be obtained.

High-resolution structural data were obtained using a T1-weighted magnetization-prepared rapid acquisition gradient echo (MPRAGE) sequence (TR = 2300 ms, TE = 2.96 ms, flip angle = 7°, inversion time = 1100 ms, FOV = 256 mm × 256 mm, matrix = 256 × 256, 192 sagittal slices, voxel size = 1 mm × 1 mm × 1 mm).

Data preprocessing

Image data analysis was carried out using the CONN toolbox v.17.f (http://www.nitrc.org/projects/conn) (Whitfield-Gabrieli and Nieto-Castanon 2012) running on MATLAB (Mathworks, Inc., Natick, MA, USA). The first 10 time points of functional data were discarded to allow stability of the longitudinal magnetization. The fMRI data were preprocessed using Statistical Parametric Mapping (SPM12) (Welcome Department of Cognitive Neurology, University College, London, UK), including functional realignment and unwrap, slice-timing correction, functional direct segmentation and normalization to Montreal Neurological Institute (MNI) space; structural segmentation and normalization to MNI space; functional spatial smoothing with a 6-mm full width at half maximum (FWHM) kernel. All images were transformed into standard stereotaxic space and resampled at 2 × 2 × 2 mm^3 voxel size. In addition, outliers were identified as volumes with a composite movement greater than 0.5 mm or more than three standard deviations away from the mean image intensity using ART toolbox(http://www.nitrc.org/projects/artifact_detect). Spurious sources of noise (such as physiological effects) estimated by the anatomical component base noise reduction strategy (aCompCor) (Behzadi et al. 2007). Then in the denoising step, a combination of aCompCor, scrubbing (identified outliers by ART), motion regression was used to remove unwanted confounding effects from the BOLD signal. After that, default band-pass filtering was performed with a frequency window of 0.008–0.09 Hz. The average number of removed functional outliers was 7.38 ± 8.78 in the FES patients and 9.14 ± 10.57 in the healthy controls.

Independent component analysis (ICA) and regions of interest (ROIs) selection

The data-driven Group-ICA provides the appropriate representation of the structured components (i.e. networks of highly functionally-connected areas across the entire brain) and therefore is chosen for identifying the SN here. We used Calhoun's group-level ICA approach (Calhoun et al. 2001) implemented in the CONN toolbox to perform group-ICA for rs-fMRI data across all subjects(patients and controls). The fastICA was used for the estimation of independent spatial components and twenty spatial independent components (ICs) were decomposed from the preprocessed and denoised images (Table 1).

The built-in spatial match template computed the spatial correlation between each group-level map and the CONN's default network masks (ICA maps from HCP, Fig. S1) were

Table 1 Demographic and clinical data for all participants

	FES ($n = 65$)	HC($n = 66$)	Statistical test
Gender (male/female)	25/39	29/34	$F = 0.0657$, $P = 0.417^{a;n.s.}$
Age (years)	25.83 ± 7.48	25.06 ± 6.41	$t = 0.633$, $P = 0.528^{b;n.s.}$
Years of education (years)	12.40 ± 3.20	12.68 ± 2.72	$t = -0.543$, $P = 0.588^{b;n.s.}$
FD (mm)	0.057 ± 0.031	0.068 ± 0.044	$t = -1.651$, $P = 0.101^{b;n.s.}$
Number of outliers	7.38 ± 8.78	9.14 ± 10.57	$t = -1.031$, $P = 0.304^{b;n.s.}$
Percentage of outliers	(4.62 ± 5.49)%	(5.71 ± 6.61)%	$t = -1.031$, $P = 0.304^{b;n.s.}$..
Duration of untreated pychosis (weeks)	35.8 ± 39.6	–	–
BPRS			
Total	46.25 ± 8.23	–	–
Positive	15.26 ± 4.44	–	–
Negative	7.81 ± 2.87	–	–
SANS	20.38 ± 12.21	–	–

FES, first-episode schizophrenia; *HC*, healthy control; *FD*, framewise displacement; *BPRS*, Brief Psychiatric Rating Scale; *SANS*, Scale for Assessment of Negative Symptoms

[a] chi-square test

[b] two-sample *t* test

[n.s.] not significant ($P > 0.05$)

used to identify networks of interest. The component comprising bilateral AI and dACC showed the best match to the SN template (r = 0.354795). After further evaluating on the basis of congruence between its spatial distribution and the SN identified by previous studies (Sridharan et al. 2008; P. C. Tu et al. 2012), we chose it as our network of interest (Fig. 1). Then we defined ROIs in the most important hubs within the network of interest (SN component) based on the peaks of the ICA clusters. The extracted peak MNI coordinates of each ROI were showed in Table 2, including dACC, bilateral AI, bilateral APFC, bilateral SMG, striatum (mainly caudate), and the cerebellum. Consequently, 6 mm radius spherical ROIs were centered on these peaks and used as seeds for whole brain seed-to-voxel functional connectivity analysis. The ROI selection procedures are widely used in functional connectivity studies (Sridharan et al. 2008; Uddin et al. 2011).

Whole brain seed-to-voxel functional connectivity analysis

The rs-fMRI time series of each ROI were computed by averaging all the voxels within each seed at each time. Pearson's correlation coefficients were calculated between the time series of each seed and that of all other voxels in the whole brain. Subsequently, Fisher's r-to-z transform was applied for increasing normality.

Statistical analysis

Two-sample t-tests were performed to examine SN's FC differences between groups. Age, gender, years of education, and FD were included in the analysis as covariates of non-interest. Results were examined at a combined threshold of voxel-wise $p < 0.001$ uncorrected and cluster-level $p < 0.05$ family-wise error (FWE) corrected for multiple comparisons.

To further examine the correlations between FC of SN and clinical variables (BRPS total score, BRPS subscale scores, and DUP), mean FC z-values were extracted from the clusters with significant group differences in the patients. As most variables did not meet the normality distribution assessed by Kolmogorov-Smirnov tests, Spearman correlations analyses (two-sided) were performed between these variables. The correlative relationship was considered to be significant at $P < 0.05$ (uncorrected for multiple comparisons).

Results

Demographic data

The detailed demographic characteristics, clinical information of 65 patients and 66 controls are shown in Table 1. No difference was found in age ($p = 0.528$), gender ($p = 0.417$), years of education ($p = 0.588$), head motion measures ($p = 0.101$) between the FES group and the HC group.

Functional connectivity analyses

As shown in Fig. 2 and Table 3, significant FC differences were found in several clusters from the ROI seeds of salience network to whole-brain other voxels. Compared to the HC group, the FES group demonstrated significantly reduced

functional connectivity between dACC and thalamus and caudate head, as well as between right AI and bilateral putamen. In contrast, significantly increased functional connectivity was found between left AI and prefrontal cortex (PFC), right APFC and right fusiform gyrus, and between right caudate and right PFC in the FES group compared to the HC group. Additionally, when the ROI seed was located on the right cerebellum, between-group comparison exhibited significantly increased FC with the right insula in the FES group.

Correlation with clinical variables

As displayed in Fig. 3, regarding the clinical symptoms, a positive correlation was found between the negative symptoms (SANS) and the FC of right cerebellum and right insula ($r = 0.288$, $p = 0.020$, uncorrected, two-sided) in the FES group. Additionally, the FC of the right cerebellum and right insula were found positively correlated with the DUP ($r = 0.253$, $p = 0.042$, uncorrected two-side). For the DUP, we also conducted the additional correlation analysis after excluding the outliers and five extremes (Supplementary materials).

Discussion

In this study, we investigated the FC of SN in the patients with FES. The SN was robustly derived by gICA of rs-fMRI data. Using eleven ROIs within the SN, we found that SN functional dysconnectivity in schizophrenia was obvious, mainly involving in CSTC circuit, cortico-cerebellar, and PFC areas. The patients with FES showed a significant hypoconnection between cortical and subcortical regions, as well as a hyperconnection between cortical regions and cerebellum within the SN. In the following correlation analyses, the reduced CSTC FC did not correlate with any clinical variables, representing a state independent dysfunction for CSTC in FES. On the other hand, significant positive associations were noted between cerebellum hyperconnectivity and negative symptoms and DUP in schizophrenia patients. Our results provide compelling evidence for the critical role of SN in the pathogenesis of schizophrenia.

The most notable connectivity abnormalities observed in the present study suggests a robust cortical-subcortical disconnection of SN during the resting state in schizophrenia, which is similar to previous evidence(P. C. Tu et al. 2012; P. C. Tu et al. 2015). We found that, compared with the HC group, the FES group showed reduced FC between the dACC and the bilateral thalamus and caudate head, and between rAI and bilateral putamen. The above-mentioned subcortical regions are commonly regarded as components of SN in the healthy and show positive

Table 2 Coordinates of salience network ROIs derived from group ICA of resting state fMRI data

Salience network ROIs	Peak (MNI)			z value
	x	y	z	
Dorsal Anterior cingulate cortex (dACC)	-2	32	26	5.86
Left ainterior insula (lAI)	−30	20	6	3.86
Right ainterior insula (rAI)	30	20	6	3.50
Left anterior prefrontal cortex (lAPFC)	−24	48	30	5.19
Right anterior prefrontal cortex (rAPFC)	26	50	26	4.81
Left supramarginal Gyrus (lSMG)	−56	−48	36	1.78
Right supramarginal Gyrus (rSMG)	60	−40	30	1.33
Left caudate (lCaudate)	−14	16	−4	2.11
Right caudate (rCaudate)	16	18	6	1.82
Left cerebellum posterior lobe (lCereb)	−44	−50	−46	1.75
Right cerebellum posterior lobe (rCereb)	48	−58	−36	2.15

ICA, independent component analysis; *MNI*, Montreal Neurological Institute; *ROI*, region of interest

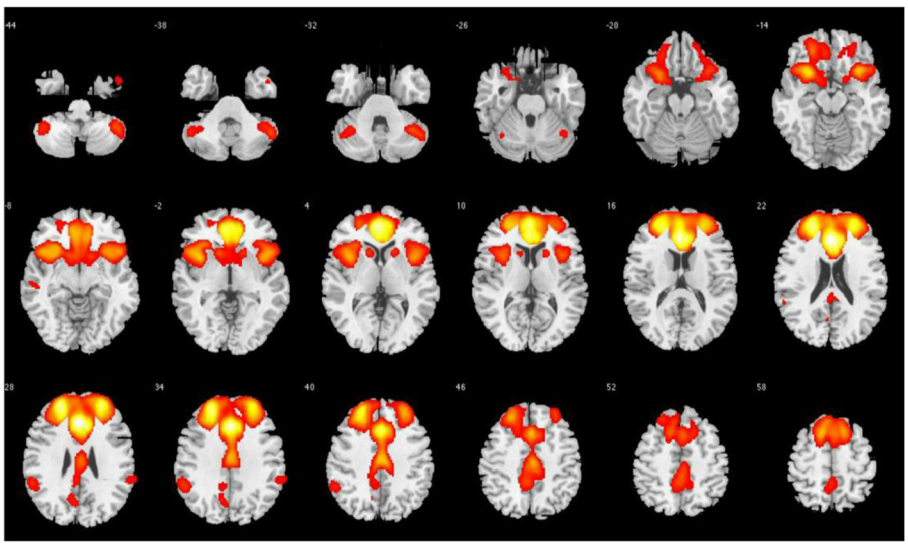

Fig. 1 The salience network (SN) identified from group ICA analysis of the resting-state fMRI data of all subjects ($n = 131$)

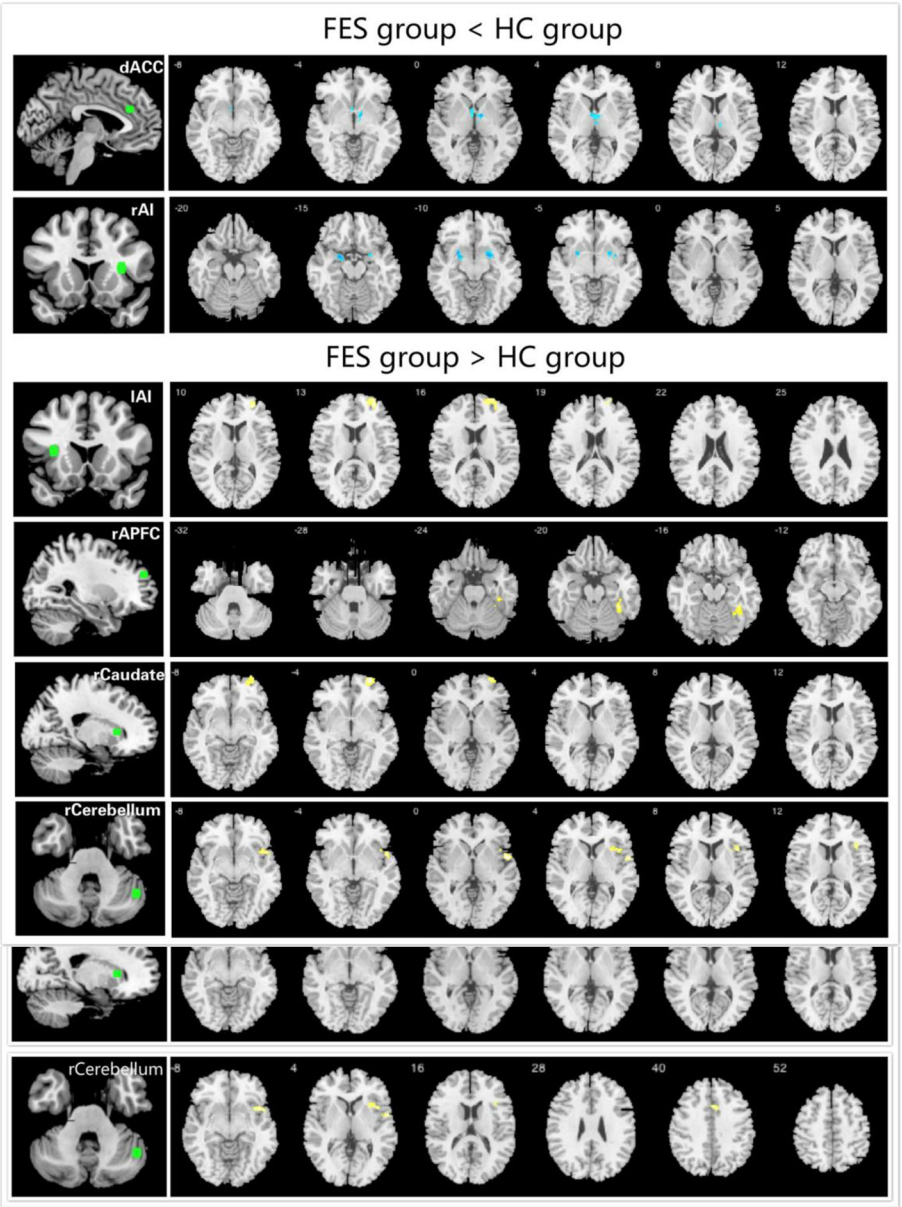

Fig. 2 Abnormal functional connectivity of salience network ROIs in the FES group compared with the HC group. Brain regions where functional connectivity of dorsal ACC (dACC), right anterior insula (rAI), left anterior insula (lAI), right anterior prefrontal cortex (rAPFC), right caudate (rCaudate), and right cerebellum posterior lobe (rCereb) showed significant differences between patients and control subjects (a combined threshold of voxel-wise $p < 0.001$, uncorrected and cluster-level $p < 0.05$ family wise error corrected for multiple comparisons). The yellow regions indicate where functional connectivity was increased in patients; blue regions indicate where functional connectivity was reduced in patients. FES, first episode schizophrenia; HC, healthy control

FC with dACC and AI; these regions are mainly located in the CSTC sub-circuit of SN, so that they are able to cooperate effectively when processing information flows. Unlike the healthy controls who showed positive dACC-thalamus and rAI-putamen connections, the patients showed weaker or even absent connections. These findings extend those of Tu et al., confirming that the significant disconnection CSTC circuit seen in schizophrenia not only exists in the chronic stage but also in the first episode. Reduced CSTC connectivity within the SN during information processing in schizophrenia suggests a pathophysiological disturbance in the system that effects changes in contextually relevant functional brain state (Palaniyappan and Liddle 2012; Sridharan et al. 2008), and might be a state independent trait in schizophrenia and represent a risk phenotype in patients with FES and their relatives (Fornito et al. 2013).

Another important finding of our study is that, in the patients, we observed an increased right cerebellar FC of cortical insular regions within the SN, and the increased FC of the right cerebellum was positively associated with negative symptoms and DUP. Despite the traditional role in motor control and perception (Paulin 1993), the cerebellum has been proposed to be involved in cognitive and affective functions (Gordon 2007; De Smet et al. 2013), and performance monitoring (Peterburs and Desmond 2016). Structural and functional abnormalities of the cerebellum have been widely investigated in schizophrenia (Zhuo et al. 2017; Liu et al. 2011; Chen et al. 2013; Guo et al. 2015a). Here, we found increased FC between cortical regions and cerebellum (mainly right Crus II) within the SN, which are inconsistent with frequently reported reduced connectivity of cerebellum.

Table 3 Regions with abnormal functional connectivity with ROIs of SN in FES patients compared with healthy controls

Cluster Location	Peak(MNI)			The z values of FC:Mean ± SD		Number of voxel	T Value[a]
	x	y	z	FES	HC		
FES < HC							
Seed: Dorsal anterior cingulate cortex(dACC)							
Thalamus and caudate head	10	−2	−2	0.074 ± 0.085	0.171 ± 0.100	143	−4.71
Seed: Right anterior insula(rAI)							
Left putamen	−24	0	−14	0.062 ± 0.104	0.160 ± 0.097	124	−4.65
Righe putamen	20	2	−10	0.081 ± 0.112	0.175 ± 0.099	158	−4.46
FES > HC							
Seed: Left anterior insula(lAI)							
Right prefrontal cortex	26	54	12	0.082 ± 0.178	−0.054 ± 0.143	206	4.41
Seed: Right anterior prefrontal cortex(rAPFC)							
Right fusiform gyrus	36	−44	−18	0.054 ± 0.091	−0.035 ± 0.108	146	5.06
Seed: Right caudate							
Right prefrontal cortex	28	64	−2	0.038 ± 0.114	−0.056 ± 0.098	230	4.67
Seed: Right cerebellum							
Right insula extending to superior temporal gyrus	52	2	2	0.045 ± 0.099	−0.056 ± 0.105	285	4.44

ROI, regions of interest; *SN*,salience network; *MNI*, Montreal Neurological Institut; *FES*, first episode schizophrenia; *HC*, healthy control

[a] A positive T value represents an increased functional connectivity and a negative T value represents a decreased functional connectivity (a combined threshold of voxel-wise $p < 0.001$ uncorrected and cluster-level $p < 0.05$ family wise error)

The prevailing opinion of overall reduced connectivity in schizophrenia is mainly drawn from findings with chronic and/or medicated patients. When the participants include drug-naive and early-stage patients, increased connectivity could be displayed (Wang et al. 2016b). Therefore, it is assumed that changes of cerebellar connectivity may meet the inverted-U hypothesis. The neural substrate underlying negative symptoms in schizophrenia remains poorly understood. Previous studies reveal the role of the abnormalities of frontal and temporal regions (Hovington and Lepage 2012; Ince and Ucok 2018), as well as cortico-striatal disconnection within the SN in negative symptoms(P. C. Tu et al. 2012). Our finding indicated that the enhanced right cerebellar FC to cortical regions may also be considered as another potential mechanism for negative symptoms. The exploratory correlation results also indicated that DUP was associated with increased cerebellar FC in the FES. However, the relationship needs further studies to confirm since it is some kind of unstable when the extremes and outliers were excluded.

Along with the above-mentioned main findings, significant between-group FC differences were also found between SN seeds and some other regions. The FES group showed increased FC between PFC and SN regions (left AI and right caudate), as well as between the right APFC and right fusiform gyrus. Among the regions implicated in the pathophysiology of schizophrenia, the PFC has always been of interest (Zhou et al. 2015). The PFC plays an essential role in the organization and control of goal-directed thought and behavior (Szczepanski and Knight 2014). Our results link the SN dysconnectivity with PFC dysconnectivity in schizophrenia, suggesting that the increased FC between these regions might contribute to the failure of effective functional integration and

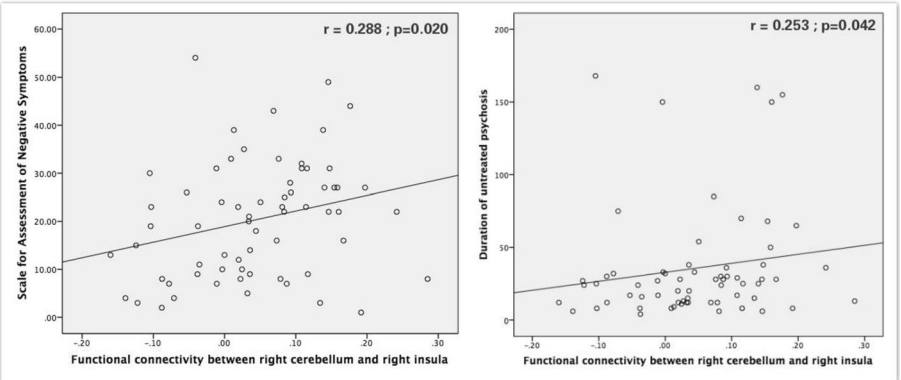

Fig. 3 Correlations between abnormal functional connectivity and clinical vari

separation when processing the goal-directed information (Palaniyappan et al. 2013).

Numerous neuroimaging studies suggest that distributed brain regions are organized into several coordinated large-scale networks and show reliable patterns of connectivity. The disruption of the coordinated activity of these brain networks is crucial for various pathological psychiatric conditions (Whitfield-Gabrieli and Ford 2012). Specifically, the attention has been mainly focused on disturbances in DMN, CEN, and SN, which are supposed to be impaired in schizophrenia (Whitfield-Gabrieli and Ford 2012; Moran et al. 2013; Jiang et al. 2017). The DMN, which includes the medial prefrontal cortex (MPFC), posterior cingulate cortex (PCC) and bilateral angular, plays an important role in internally oriented self-related mental processes (Fox et al. 2005), while the CEN, which includes the bilateral dorsolateral prefrontal cortex (DLPFC) and inferior parietal lobule (IPL), is crucial for manipulating external stimuli for goal-oriented activities (Seeley et al. 2007). Based on a triple network model (Menon 2011), a well-balanced anticorrelation between the DMN and the CEN appears crucial for effective information processing and this anticorrelation is mediated by the SN (Menon and Uddin 2010; Sridharan et al. 2008). Unlike previous studies (Manoliu et al. 2014; Moran et al. 2013; Wotruba et al. 2014), we did not find significant aberrant FC between SN and key regions within these two networks. A recent research applied support vector machine (SVM) to examine whether the rsFC of the three major networks could distinguish the FES from the healthy population. SN was identified as a classifier with an accuracy of 73.0%, and the accuracy was not related to the medication dose or psychotic symptoms, whereas the FC within the DMN or the CEN did not yield classification accuracies above chance level (Mikolas et al. 2016). Therefore, we assume that the functional abnormalities of the SN may be the most significant feature in the early course of schizophrenia beyond DMN or CEN.

As the most predominant hubs of SN, AI, and dACC share unique cytoarchitecture that helps us better understand the possible role of the SN in the pathogenesis of schizophrenia. In the human brain, dACC and AI contain a specialized class of large, bipolar projection neurons known as von Economo neurons (VENs) (Allman et al. 2010). These neurons have wider axons and appear late in evolutional history. Although the explicit function of VEN remains unknown, it has been suggested that they can enable signals to travel rapidly from the AI and dACC to other brain regions for further cognitive and emotional procedures. Density reduction (Brune et al. 2010) and ultrastructural alterations (Krause et al. 2017) of VEN in the ACC have been reported in Schizophrenia. In addition, Brune et al. found that the VEN density in the right was positively correlated with the age at onset and inversely correlated with the duration of illness in schizophrenia but not in bipolar disorder (Brune et al. 2010). These findings suggest that VENs in the ACC are involved in the pathological mechanism of schizophrenia and also highlight the vulnerability of VEN in neurodevelopmental and perhaps neurodegenerative processes for schizophrenia patients.

In contrast to our findings, two previous rs-fMRI studies reported no significant abnormalities of SN in patients with schizophrenia when comparing the FC within the regions of the SN (Woodward et al. 2011; Repovs et al. 2011). They both used the SN seeds as defined by Dosenbach in a previous study (Dosenbach et al. 2007), so the location of seed regions may be the main cause for the negative results. The inconsistency may also be partly attributed to the small sample sizes, different patient inclusions, and different image processing toolboxes. Compared to previous studies, our study investigated the detailed between-group SN resting-state functional connectivity differences in the FES and HC. The results highlight that network hubs are distinguishingly affected in schizophrenia, suggesting the different importance of two sub-circuits in SN and their roles in the pathophysiology of schizophrenia.

Several limitations should be considered when interpreting the results of this study. The first limitation is the relatively modest sample size, although the sample size was quite comparable with other fMRI studies that investigated the patients with FES. Larger sample sizes would be more powerful for the certainty of the SN abnormalities. Moreover, although only eleven patients in our study were treated with antipsychotics, the potential influence of medications contributing to the findings may not be ignored. Another potential limitation is the downside of group-ICA approach, which gives a hybrid SN map that is somewhere between the two groups included in our study (i.e., the group-ICA derived SN might not represent the network in either of the groups very well, and it might be in a way slightly different to the network described in the literature), which can make interpretation of group difference results challenging. In addition, we used less strict multiple comparison correction thresholds when reporting the seed-to-voxel functional connectivity analysis. Therefore, the present findings need to be validated by following researches.

In conclusion, the findings of the present study highlighted the abnormal functional connectivity of the SN in the patients with FES, characterized most prominently by hyperconnectivity of SN-prefrontal cortex and cerebellum, as well as hypoconnectivity of CSTC sub-circuit. Our results suggest a potentially implicated role for the SN in the pathophysiology of schizophrenia.

Author contributions DG and JW designed the current study. HH drafted the manuscript. HH, BZ, YT, TZ, LX, JW, JL, ZQ, JX, CL, and JW performed the experiments. HH, BZ, YJ, YT, TZ, LX, HW, CL, JX, and JW analyzed the data. HH, YJ, HW, JX, and JW revised the the manuscript. All of the authors read and approved the final manuscript.

Funding This work was supported by grants from Ministry of Science and Technology of China (2016YFC1306803), National Natural Science Foundation of China (81671329, 81671332), Program of Shanghai

Academic/Technology Research Leader (16XD1402400), Shanghai Science and Technology Committee (16JC1420200, 17ZR1424700), National Key Clinical Disciplines at Shanghai Mental Health Center (OMA-MH, 2011–873), Shanghai Key Laboratory of Psychotic Disorders (13dz2260500), Shanghai Jiao Tong University Foundation (14JCRY04, YG2014MS40), SHSMU- ION Research Center for Brain Disorders (2015NKX001, 15ZH2015, W35XT), Medicine Engineering Intersection Program of Shanghai Jiaotong University (YG2015ZD12) and Shanghai Hospital Development Center (16CR2015A, 16CR3017A). Projects of medical and health development in Shandong province (2017WS115), Projects of medical and health development in Qingdao city (2016WJZD068), Doctoral Innovation Fund from Shanghai Jiaotong University School of Medicine (BXJ201639).

Compliance with ethical standards

Conflict of interest The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Ethical approval The Ethics Committee of SMHC approved the study protocol(2012-45C1).

Informed consent The written, informed consent of all subjects was obtained after receiving a complete description of the study.

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

References

Allman, J. M., Tetreault, N. A., Hakeem, A. Y., Manaye, K. F., Semendeferi, K., Erwin, J. M., Park, S., Goubert, V., & Hof, P. R. (2010). The von Economo neurons in frontoinsular and anterior cingulate cortex in great apes and humans. *Brain Structure & Function, 214*(5–6), 495–517. https://doi.org/10.1007/s00429-010-0254-0.

Andreasen, N. C. (1989). The scale for the assessment of negative symptoms (SANS): Conceptual and theoretical foundations. *The British Journal of Psychiatry, 155*(Suppl 7), 49–58.

Behzadi, Y., Restom, K., Liau, J., & Liu, T. T. (2007). A component based noise correction method (CompCor) for BOLD and perfusion based fMRI. *Neuroimage, 37*(1), 90–101. https://doi.org/10.1016/j.neuroimage.2007.04.042.

Brune, M., Schobel, A., Karau, R., Benali, A., Faustmann, P. M., Juckel, G., et al. (2010). Von Economo neuron density in the anterior cingulate cortex is reduced in early onset schizophrenia. *Acta Neuropathologica, 119*(6), 771–778. https://doi.org/10.1007/s00401-010-0673-2.

Calhoun, V. D., Adali, T., Pearlson, G. D., & Pekar, J. J. (2001). A method for making group inferences from functional MRI data using independent component analysis. *Human Brain Mapping, 14*(3), 140–151.

Chen, Y. L., Tu, P. C., Lee, Y. C., Chen, Y. S., Li, C. T., & Su, T. P. (2013). Resting-state fMRI mapping of cerebellar functional dysconnections involving multiple large-scale networks in patients with schizophrenia. *Schizophrenia Research, 149*(1–3), 26–34. https://doi.org/10.1016/j.schres.2013.05.029.

Damoiseaux, J. S., Rombouts, S. A., Barkhof, F., Scheltens, P., Stam, C. J., Smith, S. M., et al. (2006). Consistent resting-state networks across healthy subjects. *Proceedings of the National Academy of Sciences of the United States of America, 103*(37), 13848–13853. https://doi.org/10.1073/pnas.0601417103.

De Smet, H. J., Paquier, P., Verhoeven, J., & Marien, P. (2013). The cerebellum: Its role in language and related cognitive and affective functions. *Brain and Language, 127*(3), 334–342. https://doi.org/10.1016/j.bandl.2012.11.001.

Dong, D., Wang, Y., Chang, X., Luo, C., & Yao, D. (2017). Dysfunction of large-scale brain networks in schizophrenia: A meta-analysis of resting-state functional connectivity. *Schizophr Bull*. https://doi.org/10.1093/schbul/sbx034.

Dosenbach, N. U., Fair, D. A., Miezin, F. M., Cohen, A. L., Wenger, K. K., Dosenbach, R. A., et al. (2007). Distinct brain networks for adaptive and stable task control in humans. *Proceedings of the National Academy of Sciences of the United States of America, 104*(26), 11073–11078. https://doi.org/10.1073/pnas.0704320104.

Dosenbach, N. U., Visscher, K. M., Palmer, E. D., Miezin, F. M., Wenger, K. K., Kang, H. C., et al. (2006). A core system for the implementation of task sets. *Neuron, 50*(5), 799–812. https://doi.org/10.1016/j.neuron.2006.04.031.

Fornito, A., Harrison, B. J., Goodby, E., Dean, A., Ooi, C., Nathan, P. J., Lennox, B. R., Jones, P. B., Suckling, J., & Bullmore, E. T. (2013). Functional dysconnectivity of corticostriatal circuitry as a risk phenotype for psychosis. *JAMA Psychiatry, 70*(11), 1143–1151. https://doi.org/10.1001/jamapsychiatry.2013.1976.

Fox, M. D., Snyder, A. Z., Vincent, J. L., Corbetta, M., Van Essen, D. C., & Raichle, M. E. (2005). The human brain is intrinsically organized into dynamic, anticorrelated functional networks. *Proc Natl Acad Sci U S A, 102*(27), 9673–9678. https://doi.org/10.1073/pnas.0504136102.

Glahn, D. C., Laird, A. R., Ellison-Wright, I., Thelen, S. M., Robinson, J. L., Lancaster, J. L., Bullmore, E., & Fox, P. T. (2008). Meta-analysis of gray matter anomalies in schizophrenia: Application of anatomic likelihood estimation and network analysis. *Biological Psychiatry, 64*(9), 774–781. https://doi.org/10.1016/j.biopsych.2008.03.031.

Gordon, N. (2007). The cerebellum and cognition. *European Journal of Paediatric Neurology, 11*(4), 232–234. https://doi.org/10.1016/j.ejpn.2007.02.003.

Guo, W., Liu, F., Chen, J., Wu, R., Zhang, Z., Yu, M., Xiao, C., & Zhao, J. (2015a). Resting-state cerebellar-cerebral networks are differently affected in first-episode, drug-naive schizophrenia patients and unaffected siblings. *Scientific Reports, 5*, 17275. https://doi.org/10.1038/srep17275.

Guo, W., Liu, F., Liu, J., Yu, L., Zhang, J., Zhang, Z., Xiao, C., Zhai, J., & Zhao, J. (2015b). Abnormal causal connectivity by structural deficits in first-episode, drug-naive schizophrenia at rest. *Schizophrenia Bulletin, 41*(1), 57–65. https://doi.org/10.1093/schbul/sbu126.

Han, S. W., Eaton, H. P., & Marois, R. (2018). Functional fractionation of the Cingulo-opercular network: Alerting insula and updating cingulate. *Cereb Cortex*. https://doi.org/10.1093/cercor/bhy130.

Hovington, C. L., & Lepage, M. (2012). Neurocognition and neuroimaging of persistent negative symptoms of schizophrenia. *Expert Review of Neurotherapeutics, 12*(1), 53–69. https://doi.org/10.1586/ern.11.173.

Ince, E., & Ucok, A. (2018). Relationship between persistent negative symptoms and findings of Neurocognition and neuroimaging in schizophrenia. *Clinical EEG and Neuroscience, 49*(1), 27–35. https://doi.org/10.1177/1550059417746213.

Jiang, Y., Duan, M., Chen, X., Chang, X., He, H., Li, Y., Luo, C., & Yao, D. (2017). Common and distinct dysfunctional patterns contribute to triple network model in schizophrenia and depression: A preliminary study. *Prog Neuropsychopharmacol Biol Psychiatry, 79*(Pt B), 302–310. https://doi.org/10.1016/j.pnpbp.2017.07.007.

Kapur, S. (2003). Psychosis as a state of aberrant salience: A framework linking biology, phenomenology, and pharmacology in schizophrenia. *The American Journal of Psychiatry, 160*(1), 13–23. https://doi.org/10.1176/appi.ajp.160.1.13.

Krause, M., Theiss, C., & Brune, M. (2017). Ultrastructural alterations of Von Economo neurons in the anterior cingulate cortex in schizophrenia. *Anat Rec (Hoboken), 300*(11), 2017–2024. https://doi.org/10.1002/ar.23635.

Leucht, S., Samara, M., Heres, S., Patel, M. X., Furukawa, T., Cipriani, A., Geddes, J., & Davis, J. M. (2015). Dose equivalents for second-generation antipsychotic drugs: The classical mean dose method. *Schizophrenia Bulletin, 41*(6), 1397–1402. https://doi.org/10.1093/schbul/sbv037.

Leucht, S., Samara, M., Heres, S., Patel, M. X., Woods, S. W., & Davis, J. M. (2014). Dose equivalents for second-generation antipsychotics: The minimum effective dose method. *Schizophrenia Bulletin, 40*(2), 314–326. https://doi.org/10.1093/schbul/sbu001.

Liu, H., Fan, G., Xu, K., & Wang, F. (2011). Changes in cerebellar functional connectivity and anatomical connectivity in schizophrenia: A combined resting-state functional MRI and diffusion tensor imaging study. *Journal of Magnetic Resonance Imaging, 34*(6), 1430–1438. https://doi.org/10.1002/jmri.22784.

Manoliu, A., Riedl, V., Zherdin, A., Muhlau, M., Schwerthoffer, D., Scherr, M., et al. (2014). Aberrant dependence of default mode/central executive network interactions on anterior insular salience network activity in schizophrenia. *Schizophrenia Bulletin, 40*(2), 428–437. https://doi.org/10.1093/schbul/sbt037.

Menon, V. (2011). Large-scale brain networks and psychopathology: A unifying triple network model. *Trends in Cognitive Sciences, 15*(10), 483–506. https://doi.org/10.1016/j.tics.2011.08.003.

Menon, V. (2015). Salience network. *Brain Mapping: An Encyclopedic Reference, 2*, 597–611. https://doi.org/10.1016/b978-0-12-397025-1.00052-x.

Menon, V., & Uddin, L. Q. (2010). Saliency, switching, attention and control: A network model of insula function. *Brain Structure & Function, 214*(5–6), 655–667. https://doi.org/10.1007/s00429-010-0262-0.

Mikolas, P., Melicher, T., Skoch, A., Matejka, M., Slovakova, A., Bakstein, E., Hajek, T., & Spaniel, F. (2016). Connectivity of the anterior insula differentiates participants with first-episode schizophrenia spectrum disorders from controls: A machine-learning study. *Psychological Medicine, 46*(13), 2695–2704. https://doi.org/10.1017/S0033291716000878.

Moran, L. V., Tagamets, M. A., Sampath, H., O'Donnell, A., Stein, E. A., Kochunov, P., et al. (2013). Disruption of anterior insula modulation of large-scale brain networks in schizophrenia. *Biological Psychiatry, 74*(6), 467–474. https://doi.org/10.1016/j.biopsych.2013.02.029.

Palaniyappan, L., & Liddle, P. F. (2012). Does the salience network play a cardinal role in psychosis? An emerging hypothesis of insular dysfunction. *Journal of Psychiatry & Neuroscience, 37*(1), 17–27. https://doi.org/10.1503/jpn.100176.

Palaniyappan, L., Simmonite, M., White, T. P., Liddle, E. B., & Liddle, P. F. (2013). Neural primacy of the salience processing system in schizophrenia. *Neuron, 79*(4), 814–828. https://doi.org/10.1016/j.neuron.2013.06.027.

Palaniyappan, L., White, T. P., & Liddle, P. F. (2012). The concept of salience network dysfunction in schizophrenia: From neuroimaging observations to therapeutic opportunities. *Current Topics in Medicinal Chemistry, 12*(21), 2324–2338.

Paulin, M. G. (1993). The role of the cerebellum in motor control and perception. *Brain, Behavior and Evolution, 41*(1), 39–50.

Peterburs, J., & Desmond, J. E. (2016). The role of the human cerebellum in performance monitoring. *Current Opinion in Neurobiology, 40*, 38–44. https://doi.org/10.1016/j.conb.2016.06.011.

Peters, S. K., Dunlop, K., & Downar, J. (2016). Cortico-striatal-thalamic loop circuits of the salience network: A central pathway in psychiatric disease and treatment. *Frontiers in Systems Neuroscience, 10*, 104. https://doi.org/10.3389/fnsys.2016.00104.

Pettersson-Yeo, W., Allen, P., Benetti, S., McGuire, P., & Mechelli, A. (2011). Dysconnectivity in schizophrenia: Where are we now? *Neuroscience and Biobehavioral Reviews, 35*(5), 1110–1124. https://doi.org/10.1016/j.neubiorev.2010.11.004.

Repovs, G., Csernansky, J. G., & Barch, D. M. (2011). Brain network connectivity in individuals with schizophrenia and their siblings. *Biological Psychiatry, 69*(10), 967–973. https://doi.org/10.1016/j.biopsych.2010.11.009.

Seeley, W. W., Menon, V., Schatzberg, A. F., Keller, J., Glover, G. H., Kenna, H., Reiss, A. L., & Greicius, M. D. (2007). Dissociable intrinsic connectivity networks for salience processing and executive control. *The Journal of Neuroscience, 27*(9), 2349–2356. https://doi.org/10.1523/JNEUROSCI.5587-06.2007.

Sridharan, D., Levitin, D. J., & Menon, V. (2008). A critical role for the right fronto-insular cortex in switching between central-executive and default-mode networks. *Proceedings of the National Academy of Sciences of the United States of America, 105*(34), 12569–12574. https://doi.org/10.1073/pnas.0800005105.

Stephan, K. E., Friston, K. J., & Frith, C. D. (2009). Dysconnection in schizophrenia: From abnormal synaptic plasticity to failures of self-monitoring. *Schizophrenia Bulletin, 35*(3), 509–527. https://doi.org/10.1093/schbul/sbn176.

Szczepanski, S. M., & Knight, R. T. (2014). Insights into human behavior from lesions to the prefrontal cortex. *Neuron, 83*(5), 1002–1018. https://doi.org/10.1016/j.neuron.2014.08.011.

Tu, P., Buckner, R. L., Zollei, L., Dyckman, K. A., Goff, D. C., & Manoach, D. S. (2010). Reduced functional connectivity in a right-hemisphere network for volitional ocular motor control in schizophrenia. *Brain, 133*(Pt 2), 625–637. https://doi.org/10.1093/brain/awp317.

Tu, P. C., Hsieh, J. C., Li, C. T., Bai, Y. M., & Su, T. P. (2012). Cortico-striatal disconnection within the cingulo-opercular network in schizophrenia revealed by intrinsic functional connectivity analysis: A resting fMRI study. *Neuroimage, 59*(1), 238–247. https://doi.org/10.1016/j.neuroimage.2011.07.086.

Tu, P. C., Lee, Y. C., Chen, Y. S., Hsu, J. W., Li, C. T., & Su, T. P. (2015). Network-specific cortico-thalamic dysconnection in schizophrenia revealed by intrinsic functional connectivity analyses. *Schizophrenia Research, 166*(1–3), 137–143. https://doi.org/10.1016/j.schres.2015.05.023.

Uddin, L. Q., Supekar, K., Amin, H., Rykhlevskaia, E., Nguyen, D. A., Greicius, M. D., & Menon, V. (2010). Dissociable connectivity within human angular gyrus and intraparietal sulcus: Evidence from functional and structural connectivity. *Cerebral Cortex, 20*(11), 2636–2646. https://doi.org/10.1093/cercor/bhq011.

Uddin, L. Q., Supekar, K. S., Ryali, S., & Menon, V. (2011). Dynamic reconfiguration of structural and functional connectivity across core neurocognitive brain networks with development. *The Journal of Neuroscience, 31*(50), 18578–18589. https://doi.org/10.1523/JNEUROSCI.4465-11.2011.

Wang, C., Ji, F., Hong, Z., Poh, J. S., Krishnan, R., Lee, J., Rekhi, G., Keefe, R. S. E., Adcock, R. A., Wood, S. J., Fornito, A., Pasternak, O., Chee, M. W. L., & Zhou, J. (2016b). Disrupted salience network functional connectivity and white-matter microstructure in persons at risk for psychosis: Findings from the LYRIKS study. *Psychological Medicine, 46*(13), 2771–2783. https://doi.org/10.1017/S0033291716001410.

Wang, H., Guo, W., Liu, F., Wang, G., Lyu, H., Wu, R., Chen, J., Wang, S., Li, L., & Zhao, J. (2016a). Patients with first-episode, drug-naive schizophrenia and subjects at ultra-high risk of psychosis shared increased cerebellar-default mode network connectivity at rest. *Scientific Reports, 6*, 26124. https://doi.org/10.1038/srep26124.

White, T. P., Gilleen, J., & Shergill, S. S. (2013). Dysregulated but not decreased salience network activity in schizophrenia. *Frontiers in Human Neuroscience, 7*, 65. https://doi.org/10.3389/fnhum.2013.00065.

Whitfield-Gabrieli, S., & Ford, J. M. (2012). Default mode network activity and connectivity in psychopathology. *Annual Review of Clinical Psychology, 8*, 49–76. https://doi.org/10.1146/annurev-clinpsy-032511-143049.

Whitfield-Gabrieli, S., & Nieto-Castanon, A. (2012). Conn: A functional connectivity toolbox for correlated and anticorrelated brain networks. *Brain Connectivity, 2*(3), 125–141. https://doi.org/10.1089/brain.2012.0073.

Woodward, N. D., Rogers, B., & Heckers, S. (2011). Functional resting-state networks are differentially affected in schizophrenia. *Schizophrenia Research, 130*(1–3), 86–93. https://doi.org/10.1016/j.schres.2011.03.010.

Wotruba, D., Michels, L., Buechler, R., Metzler, S., Theodoridou, A., Gerstenberg, M., Walitza, S., Kollias, S., Rössler, W., & Heekeren, K. (2014). Aberrant coupling within and across the default mode, task-positive, and salience network in subjects at risk for psychosis. *Schizophrenia Bulletin, 40*(5), 1095–1104. https://doi.org/10.1093/schbul/sbt161.

Zhou, Y., Fan, L., Qiu, C., & Jiang, T. (2015). Prefrontal cortex and the dysconnectivity hypothesis of schizophrenia. *Neuroscience Bulletin, 31*(2), 207–219. https://doi.org/10.1007/s12264-014-1502-8.

Zhuo, C., Wang, C., Wang, L., Guo, X., Xu, Q., Liu, Y., et al. (2017). Altered resting-state functional connectivity of the cerebellum in schizophrenia. *Brain Imaging Behav*. https://doi.org/10.1007/s11682-017-9704-0.