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Abnormal amygdala connectivity in patients with primary insomnia: Evidence from resting state fMRI

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ABSTRACT

Background: Neurobiological mechanisms underlying insomnia are poorly understood. Previous findings indicated that dysfunction of the emotional circuit might contribute to the neurobiological mechanisms underlying insomnia. The present study will test this hypothesis by examining alterations in functional connectivity of the amygdala in patients with primary insomnia (PI).
Methods: Resting-state functional connectivity analysis was used to examine the temporal correlation between the amygdala and whole-brain regions in 10 medication-naive PI patients and 10 age- and sex-matched healthy controls. Additionally, the relationship between the abnormal functional connectivity and insomnia severity was investigated.
Results: We found decreased functional connectivity mainly between the amygdala and insula, striatum and thalamus, and increased functional connectivity mainly between the amygdala and premotor cortex, sensorimotor cortex in PI patients as compared to healthy controls. The connectivity of the amygdala with the premotor cortex in PI patients showed significant positive correlation with the total score of the Pittsburgh Sleep Quality Index (PSQI).
Conclusions: The decreased functional connectivity between the amygdala and insula, striatum, and thalamus suggests that dysfunction in the emotional circuit might contribute to the neurobiological mechanisms underlying PI. The increased functional connectivity of the amygdala with the premotor and sensorimotor cortex demonstrates a compensatory mechanism to overcome the negative effects of sleep deficits and maintain the psychomotor performances in PI patients.

1. Introduction

Insomnia is considered to be the most common sleep disorder. Occasional episodes of insomnia symptoms are reported in half of the all adults while chronic insomnia is prevalent in 10–15% of the adult population. Despite its wide prevalence and broad medical impact, little is known about the neurobiological mechanisms underlying insomnia.

Clinical and psychometric studies show that patients with primary insomnia (PI) are associated with inhibited and repressive personality traits [1]. Kales proposed that PI patients tend to handle stress and conflicts through internalization of emotions, which leads to a state of constant emotional arousal and resultant physiological activation, such as increased heart rate, elevated rectal temperature and increased body movements before sleep onset [2]. Moreover, during the sleep onset period at night, PI patients frequently experience intrusive thoughts, which were described as worrisome and negatively toned. Since intrusive thoughts lead to emotional arousal, PI patients face difficulties in sleep initiation or in returning to sleep after awakenings. On the basis of these findings, it is supposed that heightened emotional reactivity contributes to the aetiology of insomnia [1].

In addition, insomnia is commonly comorbid with emotional disorders such as mood and anxiety disorders. Almost all the patients with depression had comorbid insomnia, and insomnia is also one of the most common symptoms of anxiety disorders. Moreover, depression and anxiety disorders are the most common primary diagnoses in patients suffering from insomnia. A number of longitudinal studies indicated that insomnia is predictive of future depression and bidirectionally related to anxiety [1]. These findings suggest that dysfunctional emotional reactivity

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might mediate the relationship between insomnia and emotional disorders.

Nofzinger et al. [3] evaluated the regional brain metabolism of the PI patients and healthy controls using [18F]-fluoro-2-deoxyglucose positron emission tomography (PET). It was found that PI patients exhibited smaller declines in relative glucose metabolism from wakefulness to sleep in regions associated with cognition and emotion, including the amygdala, hippocampus, insular, anterior cingulate cortex and prefrontal cortices, and in wake-promoting regions including the ascending reticular activating systems, hypothalamus and thalamus. This study gave direct evidence of heightened emotional reactivity in PI patients. Recently, Cano et al. [4] developed a rat model, with behavioural and electrophysiological similarities to stress-induced insomnia in humans, to investigate the neural circuitry of insomnia. During the period of insomnia, they found co-activation of the limbic system (including the amygdala, infralimbic cortex, cingulate cortex, lateral septum, hippocampus and the bed nucleus of the stria terminalis), cerebral cortex, parts of the arousal and autonomic systems, as well as the sleep-promoting regions. Taken together, these two important studies and the theoretical considerations outlined above suggest that dysfunction of the emotional circuit might contribute to the neurobiological mechanisms underlying insomnia.

Previous studies have confirmed that the amygdala plays a key role in emotion and is considered to be the key centre in the emotional circuit [5,6]. Dysfunction of the amygdala has been implicated in the pathophysiology of emotional disorders. Hyperactivation of the amygdala has been found as well in patients with various emotional disorders such as post-traumatic stress disorder, social anxiety disorder, specific phobia and unipolar depression [7]. Furthermore, a number of evidence suggest that the amygdala is an important modulator of sleep and plays a key role in the regulation of wakefulness during stressful events. Anatomically, the amygdala has afferent and efferent connections with sleep-regulating regions, such as the basal forebrain, preoptic area of the anterior hypothalamus, brainstem reticular formation, and solitary tract nucleus [8,9]. Lesions of the amygdala in rhesus monkeys resulted in increased sleep consolidation and total sleep time. Activation of the amygdala contributes to maintain wakefulness during stressful events [9]. These findings indicate that dysfunction in the amygdala-based neural network may be a neurobiological mechanism for insomnia.

Functional connectivity analysis using resting-state functional magnetic resonance imaging (fMRI) has been previously used to identify broadly connected networks of brain regions. Recent studies using resting-state fMRI have shown that the amygdala is extensively connected with cortical regions including the orbital and medial prefrontal cortex, cingulate cortex, parahippocampal gyrus, insula, and sub-cortical regions, including the hippocampus, striatum, thalamus and hypothalamus [10,11]. These results are consistent with known anatomical connections in nonhuman primates [5]. Abnormalities in resting-state connectivity of the amygdala have been identified in generalized anxiety disorder, posttraumatic stress disorder, bipolar disorder and schizophrenia. Use of a resting-state approach may therefore provide a useful tool for studying changes in the amygdala-based neural network in PI patients.

Based on the functional characters of the amygdala as well as the above-mentioned previous studies, we hypothesize that PI patients would exhibit altered resting-state functional connectivity of the amygdala. In the present study, we used a seed-based correlation analysis to explore resting-state functional connectivity of the amygdala in PI patients.

2. Materials and methods

2.1. Subjects

Ten medication-naive PI outpatients from our institution and ten healthy subjects, matched for age and sex, were included in this study. The study was approved by our Institutional Review Board (IRB) and written informed consent was obtained from each subject.

Subjects retained for a screening visit were interviewed and examined by two sleep neurologists. The interview included the Mini International Neuropsychiatric Interview (MINI), State-Trait Anxiety Inventory (STAI), Beck Depression Inventory-II (BDI-II), and the Pittsburgh Sleep Quality Index (PSQI). Subjects showing any polysomnographic (PSG) evidence of other sleep disorders, such as a sleep apnea syndrome (i.e., thermistors monitored apnea–hypopnea index >5) and periodic leg movements (i.e., periodic leg movement index >10) were excluded from the study.

Inclusion criteria for PI patients were as follows: PSQI higher than 5; actual PI according to the Diagnostic and Statistical Manual of Mental Disorders (4th edition, DSM-IV) criteria for PI; absence of other psychiatric diseases as evidenced by the MINI; absence of PSG evidence of other sleep disorders; no history of psychopharmacological treatment for insomnia.

Inclusion criteria for healthy subjects were as follows: PSQI lower than 5; absence of psychiatric diseases as evidenced by the MINI; absence of PSG evidence of sleep disorders.

Exclusion criteria for both groups were as follows: score higher than 50 on either of the 2 scales of the STAI (STAI-State or STAI-Trait); score higher than 13 on the BDI-II; evidence of neurological or other physical diseases such as respiratory, cardiac, renal, hepatic and endocrinal diseases as assessed by clinical history, physical examination or routine laboratory tests performed during the screening visit; any medication that might affect sleep or regional cerebral function within 14 days; irregular sleep schedules associated with shift work, frequent travel, or personal preference (as indicated by a weekly variation >3 h in bedtime or wake time, or time in bed duration <5.5 or >10 h per night).

2.2. MRI data acquisition

Scanning was performed on a 3.0 T MRI system (Siemens Trio Tim; Siemens Medical System, Erlangen, Germany) and with an 8-channel phased array head coil. Foam paddings and headphones were used to limit head motion and reduce scanning noise. Structural images were obtained by using a three-dimensional magnetization prepared rapid acquisition gradient echo (3D MPRAGE) sequence with the following parameters: repetition time (TR)/echo time (TE) = 1600/2.25 ms, flip angle = 9°, 192 sagittal slices, voxel size = 1 mm × 1 mm × 1 mm. Functional data were acquired using a T2* gradient echo echo-planar imaging (EPI) pulse sequence (TR/TE = 2000/31 ms, 30 axial slices, voxel size = 3.75 mm × 3.75 mm × 4.0 mm, 0.8 mm inter-slice gap, 90° flip angle, 64 × 64 matrix size in 240 mm × 240 mm field of view [FOV]). This acquisition sequence generated 210 volumes. Subjects were instructed to keep their eyes closed, relax and remain awake during the resting state scan.

2.3. fMRI data analysis processing

The first 10 volumes of the functional images were discarded for the signal equilibrium and participants' adaptation to the scanning noise. The slice timing, head motion correction, and spatial normalization with re-sampling to 3 mm × 3 mm × 3 mm were conducted using Statistical Parametric Mapping (SPM5, http://www.fil.ion.ucl.ac.uk/spm). No participant had head motion

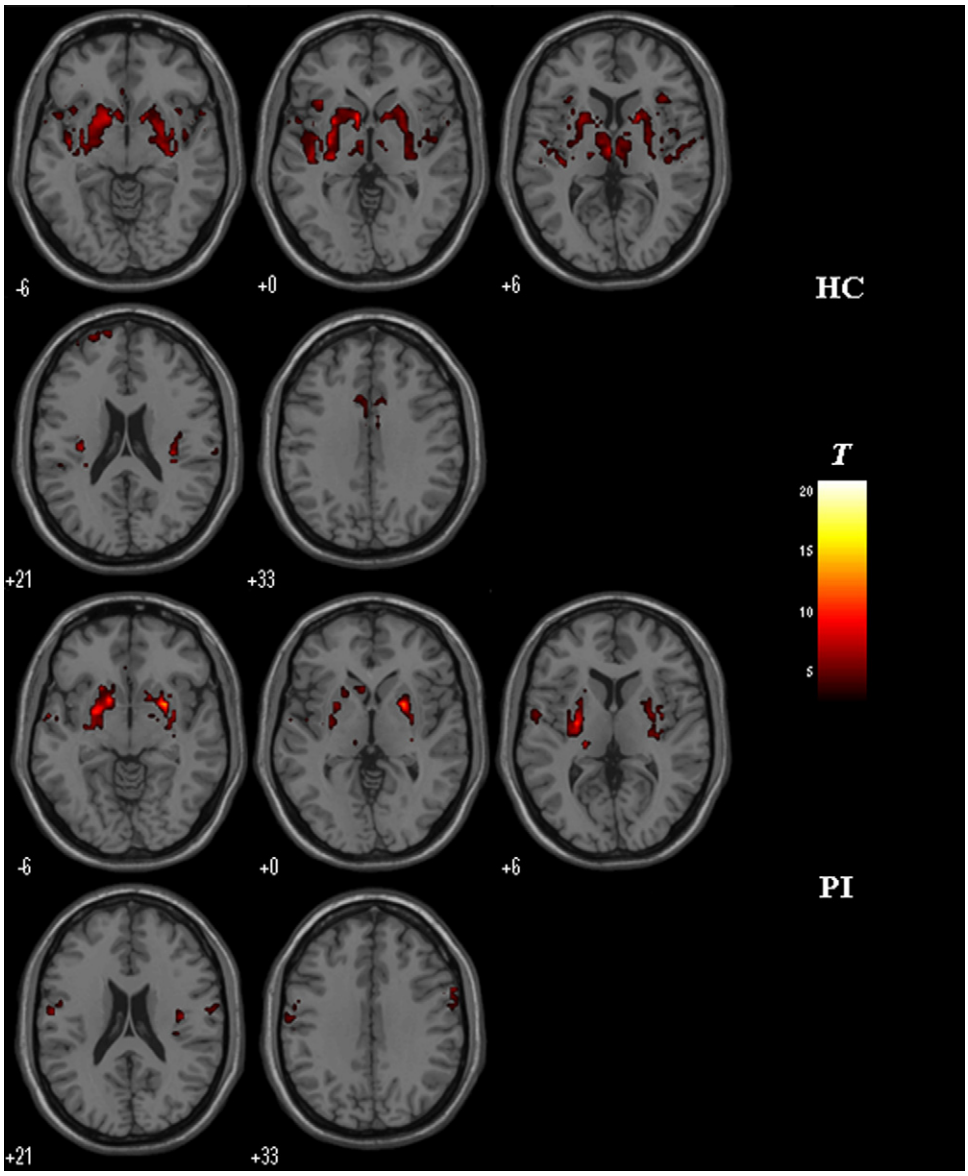

Fig. 1. Resting-state connectivity maps of the left amygdala in healthy controls and PI patients. A combined threshold was set at $p < 0.01$ and cluster size > 405 mm^3; this yields a corrected threshold of $p < 0.001$. HC: healthy controls; PI: primary insomnia.

of more than 1.5 mm maximum displacement in any of the *x*, *y*, or *z* directions, nor 1.5° of any angular motion throughout the course of scan. Resting-state fMRI data analysis toolkit (http://resting-fmri.sourceforge.net) was then used for removing the linear trend of time courses and for temporally band-pass filtering the data (0.01–0.08 Hz) [12]. The resulting data was spatially smoothed (4 mm full-width at half maximum [FWHM] Gaussian kernel). To further reduce the effects of confounding factors, we also used a linear regression process to further remove the effects of head motion and other possible sources of artifacts: (1) six motion parameters, (2) whole-brain signal averaged over the entire brain, (3) linear drift.

2.4. Functional connectivity analysis

The bilateral amygdala region of interest (ROI) was generated using the free software WFU_PickAtlas (http://www.ansir.wfubmc.edu) [13]. For each seed region, the blood oxygen level dependent (BOLD) time series of the voxels within the seed region were averaged to generate the region's reference time series.

For each subject and each seed region, a correlation map was produced by computing the correlation coefficients between the reference time series and the time series from all other brain voxels. Correlation coefficients were converted to *z*-values using Fisher's *r*-to-*z* transformation to improve the normality [14]. Then, the individual *z*-values were entered into a random effect one-sample *t*-test in a voxel-wise manner to determine brain regions showing significant connectivity to the left and right amygdala within each group. The survived clusters were under a combined threshold of $p < 0.01$ and cluster size > 405 mm^3. This yield a corrected threshold of $p < 0.001$, determined by Monte Carlo simulation using the AlphaSim program (parameters were: FWHM = 4 mm, within a mask of the whole brain gray matter tissues) (http://afni.nimh.nih.gov/pub/dist/doc/manual/AlphaSim.pdf). Two mask images were then made by combining the one sample *t*-test results of healthy controls and PI patients for both side of amygdala independently using Image Calculator in SPM5.

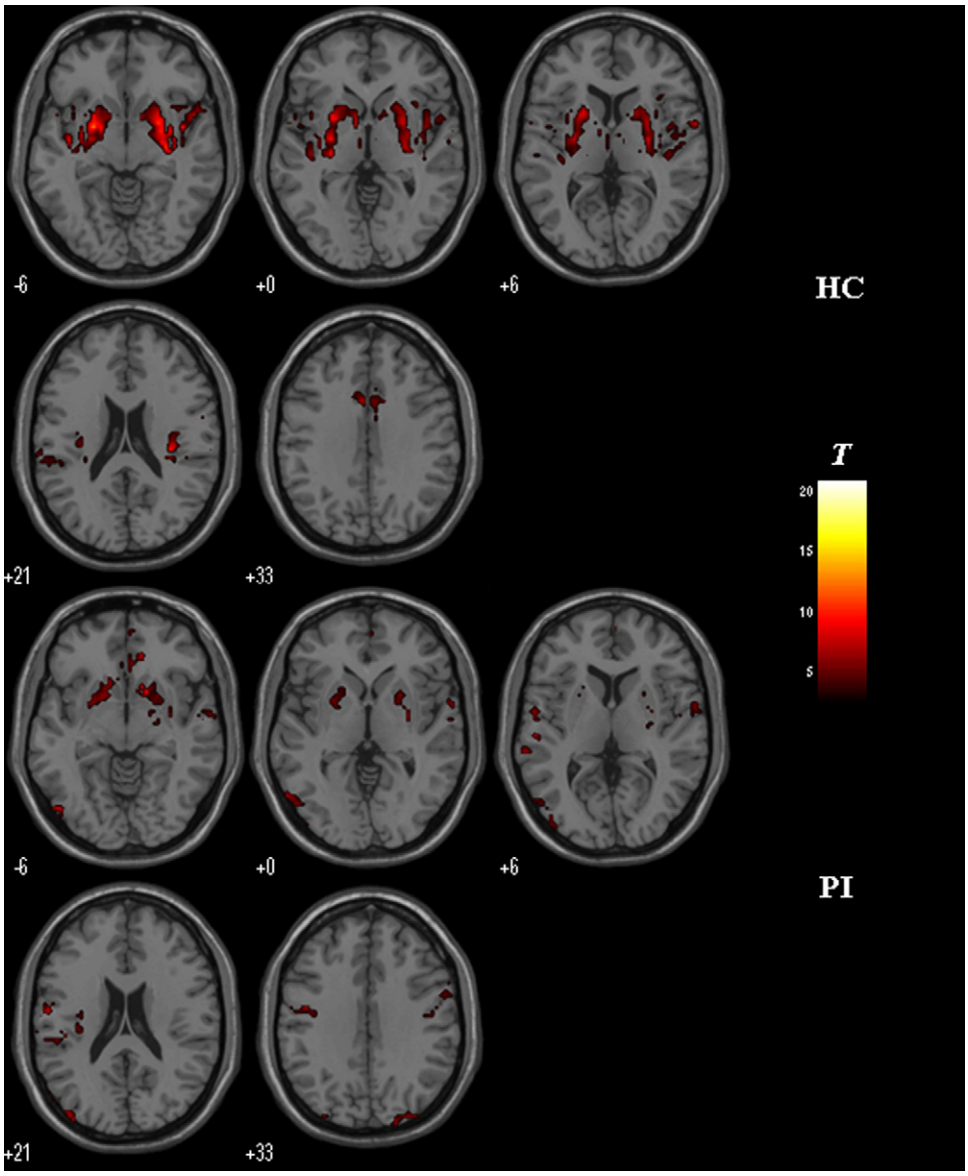

Fig. 2. Resting-state connectivity maps of the right amygdala in healthy controls and PI patients. A combined threshold was set at $p < 0.01$ and cluster size > 405 mm^3; this yields a corrected threshold of $p < 0.001$. HC: healthy controls; PI: primary insomnia.

The *z*-values were also entered into a random effect two-sample *t*-test to identify the regions showing significant differences in connectivity to the bilateral amygdala between PI patients and healthy controls. The activations reported for group differences survived a combined threshold of $p < 0.01$ and cluster size of 162 mm^3, which yielded a corrected threshold of $p < 0.05$ (using the AlphaSim program with parameters: FWHM = 4 mm and within each bilateral masks).

2.5. Statistical analysis

The following statistical analyses were performed using SPSS 17.0 software (SPSS Inc., Chicago, IL, USA). Statistical comparisons of the demographic data and clinical characteristics between the two groups were performed using an independent-samples *t*-test. In order to investigate the relationship between the abnormal functional connectivity and insomnia severity, Pearson's correlation test was performed to examine the correlation between the abnormal functional connectivity and the total PSQI score in PI patients. The level of statistical significance was set at $p < 0.05$.

3. Results

3.1. Demographic and clinical characteristics

The PSQI ($t = -12.88$, $p < 0.01$) and STAI-State ($t = -3.18$, $p < 0.05$) scores of PI patients were significantly higher than those of healthy controls. No significant differences were observed in the STAI-Trait ($t = 0$, $p = 1.00$) and BDI-II ($t = -0.33$, $p = 0.74$) scores between the two groups. The demographic and clinical characteristics of the subjects are summarized in Table 1.

3.2. Functional connectivity of the amygdala: within group results

In healthy controls, multiple brain regions were identified to be functionally connected to the amygdala, including the medial frontal gyrus (Brodmann area [BA] 10), anterior cingulate cortex (BA 24/32), inferior frontal gyrus (BA 47), middle frontal gyrus (BA 11), postcentral gyrus (BA1/40), superior temporal gyrus (BA 38), middle temporal gyrus (BA 21), insula (BA 13), and several

Table 1
Demographic and clinical characteristics of the subjects.

Variable	PI patients Mean (SD)	Controls Mean (SD)	*p*-value
Cases	10	10	
Male/female	5/5	5/5	1.00[#]
Age	37.50 (12.38)	35.50 (8.67)	0.68[*]
PSQI	13.70 (2.63)	2.10 (1.10)	<0.01[*]
STAI-S	27.50 (3.47)	23.30 (2.31)	<0.05[*]
STAI-T	31.00 (4.94)	31.00 (5.31)	1.00[*]
BDI-II	7.20 (2.66)	6.80 (2.74)	0.74[*]

Notes: PSQI: Pittsburgh Sleep Quality Index; STAI-S: State-Trait Anxiety Inventory-State; STAI-T: State-Trait Anxiety Inventory-Trait; BDI-II: Beck Depression Inventory-II.

[*] The *p* value was obtained by a two-sample two-tailed *t*-test.

[#] The *p* value was obtained using a Pearson χ^2 two-tailed test, with continuity correction for $n < 5$.

subcortical regions (hippocampus, putamen, caudate, lateral globus pallidus and thalamus). By visual inspection, the overall pattern of the amygdala functional connectivity in PI patients was similar to that of the healthy controls (Figs. 1 and 2).

3.3. Functional connectivity of the amygdala: between-group results

Compared to the healthy controls, the left amygdala of PI patients exhibited decreased functional connectivity with the right inferior frontal gyrus (BA 47), bilateral superior temporal gyrus (BA 22/38), left insula (BA 13), left thalamus, bilateral caudate and bilateral lentiform nucleus, and increased functional connectivity with the bilateral precentral gyrus (BA 4/6) and left inferior frontal gyrus (BA 9).

The right amygdala of PI patients exhibited decreased functional connectivity with the bilateral superior temporal gyrus (BA 13/22), right insula and left lateral globus pallidus, and increased functional connectivity with the bilateral precentral gyrus (BA 6), left postcentral gyrus (BA 43), left middle temporal gyrus (BA 39), left middle occipital gyrus (BA 37), and right cuneus (BA 19) (Tables 2 and 3; Figs. 3 and 4).

3.4. Correlation between abnormal functional connectivity and total PSQI score

The connectivity of the right amygdala showed significant (right premotor cortex, $r = 0.65$, $p = 0.03$) or near significant (left premotor cortex, $r = 0.55$, $p = 0.06$) correlation with the total PSQI score. The correlation coefficient between the connectivity of the left amygdala with a cluster in the right premotor cortex and the total PSQI score was 0.45, not significant ($p = 0.11$). Other correlations were all below 0.4.

4. Discussion

In the present study, resting-state fMRI was used to examine alterations in functional connectivity of the amygdala in PI patients to test the hypothesis that dysfunction of emotional circuit might contribute to the neurobiological mechanisms underlying insomnia. Using an amygdala-based connectivity analysis, we found some regions showed decreased functional connectivity with the amygdala while other regions showed increased functional connectivity with the amygdala.

We found decreased functional connectivity between the amygdala and insula, striatum, and thalamus in PI patients. Emotional processing is linked to anatomically distinct and well-defined brain regions, including the amygdala, orbital and medial prefrontal cortex, cingulate cortex, insula, hippocampus, striatum, thalamus and hypothalamus [5–7,10]. Reciprocal connections between these structures form the emotional circuit, which has been implicated in various aspects of emotion [6,7]. Findings from animal, human lesion, and functional neuroimaging studies have confirmed that the amygdala plays a key role in the emotional processing by interacting with other regions [5–7]. As the amygdala is the central part of the emotional circuit, decreased functional connectivity between the amygdala and insula, thalamus and striatum suggest dysfunction of emotional circuit in PI patients.

Although we found abnormalities in functional connectivity of the amygdala in PI patients, it was not sure whether these abnormalities are the consequence of sleep loss, or involved in the pathogenesis of PI. In a recent study, Fernández-Mendoza et al. [15] investigated whether cognitive-emotional hyperarousal is a premorbid characteristic of good sleepers vulnerable to stress-related insomnia, as measured by the Ford Insomnia Response to Stress Test (FIRST) scores. They concluded that cognitive-emotional hyperarousal is not a consequence of insomnia, but rather a predisposing factor to insomnia. However, findings from studies on sleep deprivation showed that alterations in the amygdala reactivity could be induced by sleep deficits [16]. Future studies should investigate this relationship in more details.

We also found increased functional connectivity between the amygdala and premotor cortex, sensorimotor cortex. Moreover, the connectivity between the amygdala and premotor cortex has positive correlation with the total PSQI score. Being exposed to fear signals makes us feel threatened and prompts us to prepare an adaptive response. In contrast to external threat, the PI patients are under "internal" threat, i.e., fear of being unable to sleep, and the associated consequences on other issues like work and social relations [2]. The amygdala plays a critical role in initiating adaptive behavioural response to threats via its connections with subcortical regions such as the hypothalamus and brainstem, and higher cortical regions, such as prefrontal cortex and premotor cortex [17]. The premotor cortex is known to be implicated in the preparation of an adapted motor action in response to the perception of fear signals [18]. Thus, the increased functional connectivity between the amygdala and premotor cortex, sensorimotor cortex might reflect an adaptive response to the "internal" threat in PI patients.

Studies in normal sleepers after sleep deprivation consistently show impaired psychomotor performance, which include slow reaction time, impaired vigilance, impaired memory and reasoning [19]. On the contrary, neuropsychological studies investigating daytime performance in PI patients have failed to detect significant daytime deficits [2]. These findings suggest that increased functional connectivity between the amygdala and premotor cortex, sensorimotor cortex demonstrate a compensatory mechanism to overcome the negative effects of sleep deficits and maintain the psychomotor performances. In addition, when partially deprived of sleep, normal individuals with shorter daily sleep duration (e.g., 3 h) exhibits a greater impairment of psychomotor performances than longer daily sleep duration (e.g., 5 or 7 h) [20]. The PSQI assesses sleep quality during the previous month, and higher score of the PSQI indicates worse sleep quality. Thus, PI patients with worse sleep quality are assumed to have invested more compensatory mechanism to overcome the negative effects of sleep deficits, which explains the positive correlation between the connectivity of the amygdala with premotor cortex and the total PSQI score.

The present study still has several limitations. First, it should be noted that, like most resting-state functional connectivity studies, it is difficult to completely eliminate the effects of physiological noise because we used a relatively low sampling rate (TR = 2 s) for multi-slice acquisitions, and thus cardiac effects would be aliased into the low-frequency fluctuations. In future studies, these physiological effects may be estimated and removed by simultaneously recording the respiratory and cardiac cycles during data

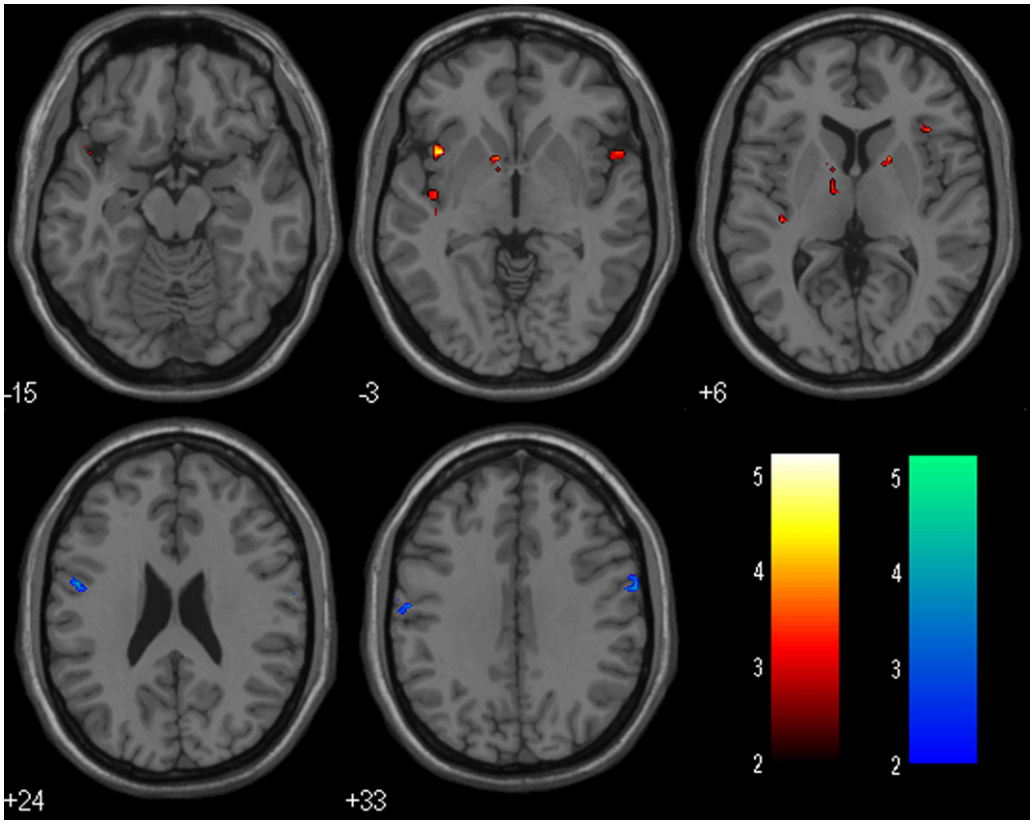

Fig. 3. Regions showing significant differences in functional connectivity to the left amygdala between PI patients and healthy controls. Yellow areas indicate brain regions with significantly decreased functional connectivity with the left amygdala in PI patients compared to healthy controls. Blue areas indicate brain regions with significantly increased functional connectivity with the left amygdala in PI patients compared to healthy controls. The threshold was set at $p < 0.01$ with a minimum cluster size of 6 contiguous voxels, which yield a corrected threshold of $p < 0.05$.

acquisition. Second, we only examined functional connectivity during resting state, which may not reflect connectivity during task performance. Previous studies suggest that the amplitude of resting-state network fluctuations is modulated by the transition between task performance and rest. Although recent studies suggest that networks identified in the resting-state mimic those identifiable across a wide array of task paradigms, future studies should also examine task-related changes in the functional connectivity of the amygdala in PI patients.

Table 2
Abnormal functional connectivity of the left amygdala in PI patients compared with healthy controls.

Brain regions	MNI coordinates			BA	Cluster size	*T*-score
	x	*y*	*z*			
Decreased functional connectivity						
Rt. inferior frontal gyrus	39	24	3	47	6	3.87
Lt. superior temporal gyrus	−45	12	−15	38	6	3.20
Rt. superior temporal gyrus	54	12	−3	22	6	3.55
Lt. insula	−39	12	−3	13	17	4.92
	−36	21	3	13		4.38
Lt. insula	−42	−21	0	13	37	3.83
	−36	−21	15	13		3.55
	−42	−12	0	13		3.52
Lt. thalamus	−9	−9	0		19	3.65
	−9	−9	12			3.23
Lt. caudate	−9	6	0		18	5.17
Lt. lentiform nucleus	−18	9	9			2.73
Rt. caudate	12	9	0			2.97
Rt. lentiform nucleus	18	9	6		8	3.97
Increased functional connectivity						
Lt. precentral gyrus	−39	−15	54	4	18	4.10
Lt. postcentral gyrus	−48	−18	51	3		3.58
Lt. precentral gyrus	−57	−12	33	4	18	3.81
Lt. postcentral gyrus	−57	−21	36	4		2.97
Rt. precentral gyrus	63	−3	33	6	10	3.96
	66	−3	21	6	6	4.23
Lt. inferior frontal gyrus	−51	0	24	9	9	4.08

Notes: The threshold was set at $p < 0.01$ with a minimum cluster size of 6 contiguous voxels, which yield a corrected threshold of $p < 0.05$. Rt: right; Lt: left; BA: Brodmann's area; MNI: Montreal Neurological Institute. Cluster size is in number of voxels.

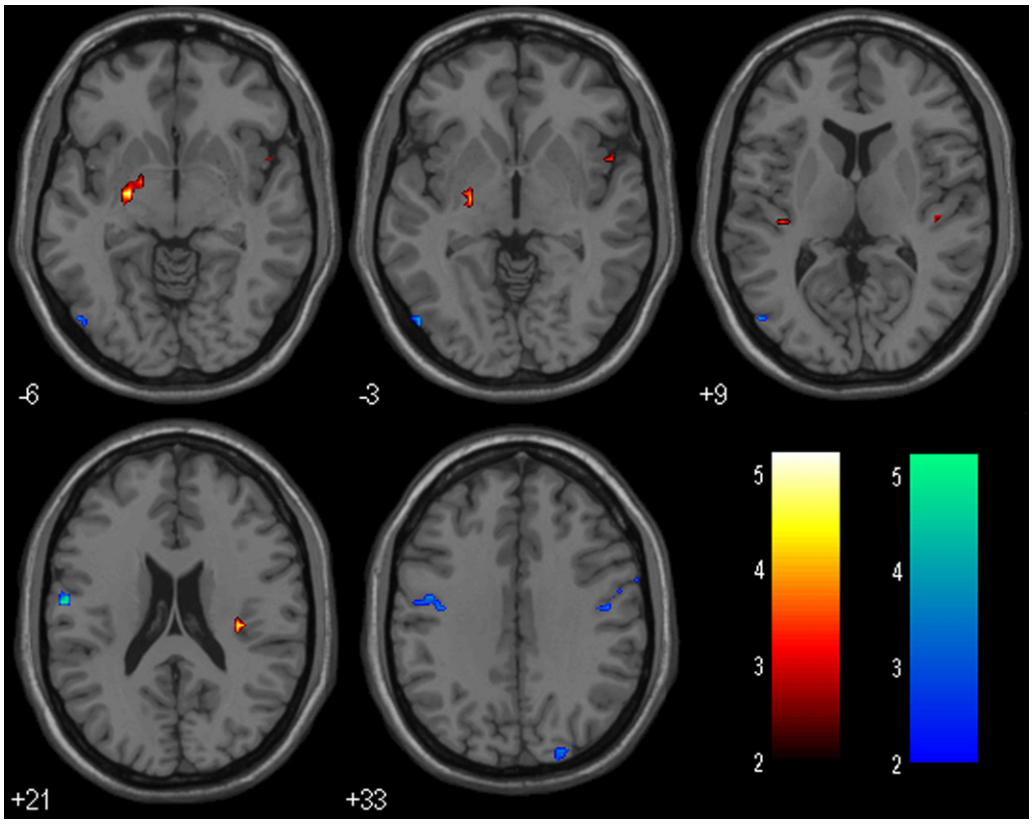

Fig. 4. Regions showing significant differences in functional connectivity to the right amygdala between PI patients and healthy controls. Yellow areas indicate brain regions with significantly decreased functional connectivity with the right amygdala in PI patients compared to healthy controls. Blue areas indicate brain regions with significantly increased functional connectivity with the right amygdala in PI patients compared to healthy controls. The threshold was set at $p < 0.01$ with a minimum cluster size of 6 contiguous voxels, which yield a corrected threshold of $p < 0.05$.

Table 3
Abnormal functional connectivity of the right amygdala in PI patients compared with healthy controls.

Brain regions	MNI coordinates			BA	Cluster size	*T*-score
	x	*y*	*z*			
Decreased functional connectivity						
Lt. superior temporal gyrus	−39	−24	9	13	7	3.54
Rt. superior temporal gyrus	51	9	−3	22	6	3.39
	45	−21	6	13	6	3.61
Rt. insula	33	−21	21	13	12	4.70
	33	−33	18	13		3.16
Lt. lateral globus	−24	−9	−6		23	5.18
pallidus	−18	−3	−6			3.68
Increased functional connectivity						
Lt. precentral gyrus	−51	−3	39	6	17	4.33
	−45	−6	33	6		3.79
	−39	−12	33	6		3.67
Lt. postcentral gyrus	−60	−9	21	43	15	5.08
Lt. precentral gyrus	−54	−3	24	6		4.52
Rt. precentral gyrus	63	3	30	6	7	4.33
Rt. precentral gyrus	48	−12	33	6	14	3.49
	54	−3	33	6		3.28
Lt. middle occipital gyrus	−54	−72	0	37	20	4.56
Lt. middle temporal gyrus	−51	−75	9	39		3.99
Rt. cuneus	24	−87	33	19	14	3.50

Notes: The threshold was set at $p < 0.01$ with a minimum cluster size of 6 contiguous voxels, which yield a corrected threshold of $p < 0.05$. Rt: right; Lt: left; BA: Brodmann's area; MNI: Montreal Neurological Institute. Cluster size is in number of voxels.

5. Conclusions

In conclusion, the current study is the first report of abnormalities in resting-state functional connectivity of the amygdala in PI patients. The decreased functional connectivity between the amygdala and insula, striatum, and thalamus suggests dysfunction of the emotional circuit in PI patients, which supports the hypothesis that dysfunction of the emotional circuit might contribute to the neurobiological mechanisms underlying insomnia. The increased functional connectivity of the amygdala with premotor cortex and sensorimotor cortex in PI patients demonstrates a compensatory mechanism to overcome the negative

effects of sleep deficits and maintain the psychomotor performances.

Conflict of interest

All authors have no conflict of interest.

Acknowledgments

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References

[1] Baglioni C, Spiegelhalder K, Lombardo C, Riemann D. Sleep and emotions: a focus on insomnia. Sleep Med Rev 2010;14(4):227–38.
[2] Riemann D, Spiegelhalder K, Feige B, et al. The hyperarousal of insomnia: a review of the concept and its evidence. Sleep Med Rev 2010;14(1):19–31.
[3] Nofzinger EA, Buysse DJ, Germain A, Price JC, Miewald JM, Kupfer DJ. Functional neuroimaging evidence for hyperarousal in insomnia. Am J Psychiatry 2004;161(11):2126–8.
[4] Cano G, Mochizuki T, Saper CB. Neural circuitry of stress-induced insomnia in rats. J Neurosci 2008;28(40):10167–84.
[5] Amaral DG, Price JL. Amygdalo-cortical projections in the monkey (*Macaca fascicularis*). J Comp Neurol 1984;230(4):465–96.
[6] Phillips ML, Drevets WC, Rauch SL, Lane R. Neurobiology of emotion perception I: the neural basis of normal emotion perception. Biol Psychiatry 2003;54(5):504–14.
[7] Price JL, Drevets WC. Neurocircuitry of mood disorders. Neuropsychopharmacology 2010;35(1):192–216.
[8] Jha SK, Ross RJ, Morrison AR. Sleep-related neurons in the central nucleus of the amygdala of rats and their modulation by the dorsal raphe nucleus. Physiol Behav 2005;86(4):415–26.
[9] Germain A, Buysse DJ, Nofzinger E. Sleep-specific mechanisms underlying posttraumatic stress disorder: integrative review and neurobiological hypotheses. Sleep Med Rev 2008;12(3):185–95.
[10] Roy AK, Shehzad Z, Margulies DS, et al. Functional connectivity of the human amygdala using resting state fMRI. Neuroimage 2009;45(2):614–26.
[11] Etkin A, Prater KE, Schatzberg AF, Menon V, Greicius MD. Disrupted amygdalar subregion functional connectivity and evidence of a compensatory network in generalized anxiety disorder. Arch Gen Psychiatry 2009;66(12):1361–72.
[12] Biswal B, Yetkin FZ, Haughton VM, Hyde JS. Functional connectivity in the motor cortex of resting human brain using echo-planar MRI. Magn Reson Med 1995;34:537–41.
[13] Maldjian JA, Laurienti PJ, Kraft RA, Burdette JH. An automated method for neuroanatomic and cytoarchitectonic atlas-based interrogation of fMRI data sets. Neuroimage 2003;19:1233–9.
[14] Lowe MJ, Mock BJ, Sorenson JA. Functional connectivity in single and multislice echoplanar imaging using resting-state fluctuations. Neuroimage 1998;7:119–32.
[15] Fernández-Mendoza J, Vela-Bueno A, Vgontzas AN, et al. Cognitive-emotional hyperarousal as a premorbid characteristic of individuals vulnerable to insomnia. Psychosom Med 2010;72(4):397–403.
[16] Yoo SS, Gujar N, Hu P, Jolesz FA, Walker MP. The human emotional brain without sleep – a prefrontal amygdala disconnect. Curr Biol 2007;17(20):R877–8.
[17] Avendano C, Price JL, Amaral DG. Evidence for an amygdaloid projection to premotor cortex but not to motor cortex in the monkey. Brain Res 1983;264:111–7.
[18] Hoshi E, Tanji J. Functional specialization in dorsal and ventral premotor areas. Prog Brain Res 2004;143:507–11.
[19] Vgontzas AN, Zoumakis E, Bixler EO, et al. Adverse effects of modest sleep restriction on sleepiness, performance, and inflammatory cytokines. J Clin Endocrinol Metab 2004;89:2119–26.
[20] Dinges DF, Pack F, Williams K, et al. Cumulative sleepiness, mood disturbance, and psychomotor vigilance performance decrements during a week of sleep restricted to 4–5 hours per night. Sleep 1997;20:267–77.