Spindle-related brain activation in patients with insomnia disorder: An EEG-fMRI study

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Abstract

Sleep spindles have been implicated in sleep protection, depression and anxiety. However, spindle-related brain imaging mechanism underpinning the deficient sleep protection and emotional regulation in insomnia disorder (ID) remains elusive. The aim of the current study is to investigate the relationship between spindle-related brain activations and sleep quality, symptoms of depression and anxiety in patients with ID. Participants (n = 46, 29 females, 18–60 years) were recruited through advertisements including 16 with ID, according to the Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition, and 30 matched controls. Group differences in spindle-related brain activations were analyzed using multimodality data acquired with simultaneous electroencephalography and functional magnetic resonance imaging during sleep. Compared with controls, patients with ID showed significantly decreased bilateral spindle-related brain activations in the cingulate gyrus (familywise error corrected p < 0.05, cluster size 3834 mm^3). Activations in the cingulate gyrus were negatively correlated with Self-Rating Depression Scale scores (r = -0.454, p = 0.002), Self-Rating Anxiety Scale scores (r = -0.362, p = 0.014) and subjective sleep onset latency (r = -0.374, p = 0.010), while positively correlated with subjective total sleep time (r = 0.343, p = 0.020), in the pooled sample. Besides, activations in the cingulate gyrus were negatively correlated with scores on the Self-Rating Depression Scale (r = -0.650, p = 0.006), in the ID group. These findings underscore the key role of spindle-related brain activations in the cingulate gyrus in subjective sleep quality and emotional regulation in ID.

Introduction

Insomnia disorder (ID) refers to subjectively reported unsatisfactory sleep quantity or quality, leading to the development of depression and anxiety (American-Psychiatric-Association, 2013; Hertenstein et al., 2019). Approximately 8-20% of adults suffer from ID (Adams et al., 2017; Kerkhof, 2017). There are no objective measures to identify and diagnose ID. Although polysomnography (PSG) has been regarded as “gold standard” for screening sleep disorders, it is not considered necessary for a clinical diagnosis of ID. Sleep macro-architecture derived from PSG exhibited lower night-to-night reliability and temporal resolution than quantitative electroencephalogram (EEG) (Israel et al., 2012).

Sleep spindles are distinctive EEG features for sleep analysis (Figure S1). They are bursts of 11-16 Hz oscillations that last for at least 0.5 sec with a waxing-and-waning shape; spindles constitute one of the hallmarks of stage 2 nonrapid eye movement (NREM) sleep (N2) (Berry et al., 2016). Spindles have been shown to play an important role in the preservation of sleep continuity. In a study by Dang-Vu and colleagues, healthy individuals with greater spindle rates were more resistant to external noise during sleep (Dang-Vu, McKinney, Buxton, Solet, & Ellenbogen, 2010). Besset et al. provided additional support for the sleep continuity hypothesis by demonstrating that comparing with good sleeper controls, sleep maintenance insomniacs display significantly lower spindle activity, as well as a diminished inverse relationship between slow wave activity and spindles across sleep time (Besset, Villemin, Tafti, & Billiard, 1998). In the meanwhile, lower spindle activity, especially at the beginning of the night, prospectively predicted larger increases in stress-related insomnia symptoms (Dang-Vu et al., 2015). A functional

magnetic resonance imaging (fMRI) study did not find significant response to noise in the primary auditory cortex during spindle events (Dang-Vu et al., 2011). Manipulations to enhance spindles in rodents directly indicated the role of spindles in gating sensory transmission (Wimmer et al., 2012).

Differences regarding spindle characteristics between patients with ID and good sleepers were noted, albeit inconsistently. Recently published meta-analysis indicated that patients with ID exhibit significant increases in relative sigma power (a proxy for spindles) during NREM sleep and absolute sigma power during rapid eye movement (REM) sleep (Zhao et al., 2021). Bastien and colleagues visually examined the total spindle number and density during N2 sleep in patients with ID (Bastien et al., 2009). In this study, insomnia patients showed trends of higher spindle number and density. Two recent studies adopted the automatic event-detection method to investigate spindle characteristics in insomnia patients, yielding conflicting conclusions (Andrillon et al., 2020; Normand, St-Hilaire, & Bastien, 2016). Overall, the existing studies about between-group differences must be interpreted with caution due to small sample sizes, methodological factors regarding spindle measurement, and variability in insomnia subtypes.

Simultaneous EEG and fMRI (EEG-fMRI) studies have consistently revealed brain regions time-locked to spindles in normal volunteers (Andrade et al., 2011; Bergmann, Molle, Diedrichs, Born, & Siebner, 2012; Caporro et al., 2012; Fogel et al., 2017; Schabus et al., 2007) and in patients with epilepsy (Caporro et al., 2012; Tyvaert, Levan, Grova, Dubeau, & Gotman, 2008); these regions include the thalamus, striatum, frontal, central, temporal, occipital, hippocampus, and cerebellum. However, brain activations coinciding with spontaneous spindles in ID and their potential role in sleep protection have seldom been addressed.

Therefore, in the present study, we sought to identify brain areas associated with spindles in ID patients and healthy controls (HCs) employing EEG-fMRI recordings. An event-related analytic approach was used to evaluate brain activations based on the blood oxygenation level-dependent (BOLD) signals in both groups. We hypothesized that ID patients would show significantly decreased spindle-related brain activations compared with HCs. We also evaluated whether clinical characteristics were correlated with decreased BOLD signals in the whole sample and in the ID group, respectively.

Materials And Methods

Participants

Subjects were recruited through advertisements at Peking University Sixth Hospital from July 2017 to September 2019. All participants signed informed consent before participation. The research protocol was approved by the Ethical Committee of Peking University Sixth Hospital.

Inclusion criteria were as follows: (1) right-handed Han Chinese, (2) 18-60 years old. ID patients should meet the diagnostic criteria for ID by the Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM-5) (American-Psychiatric-Association, 2013), and the Insomnia Severity Index (ISI) score ≥ 15 (Morin, Belleville, Bélanger, & Ivers, 2011). HCs should meet the criteria for normal sleep (Beattie, Espie, Kyle, & Biello, 2015).

Exclusion criteria were as follows: (1) history of psychiatric (Lecrubier et al., 1997) or neurological illness, (2) psychoactive substances abuse, (3) presence of serious heart problems, liver disease, kidney disease, infectious disease, endocrine problems or cancer, (4) apnea-hypopnea index ≥ 5/hr, (5) periodic limb movements during sleep ≥ 20/hr, (6) employment as a shift worker or had experienced a trans-meridian trip in the last 3 months, (7) women who were pregnant or in a breast feeding period, (8) contraindications to magnetic resonance imaging (MRI), (9) body mass index (BMI) ≥ 35 kg/m^2, and (10) using drugs that affect sleep nearly two weeks prior to the study.

Participants were asked to complete the ISI (Yu, 2010), Pittsburgh Sleep Quality Index (PSQI) (Tsai et al., 2005), Self-Rating Depression Scale (SDS) (Huang et al., 2020) and Self-Rating Anxiety Scale (SAS) (Liu et al., 2013), during the recruitment process. Finally, 16 patients with ID and 30 controls were included in the final analyses (Figure 1).

Experimental procedure

Before the experiment, participants were asked to follow a two-week regular sleep-wake schedule, which was assessed through actigraphy and sleep diaries. Eligible participants then underwent two consecutive nights of PSG (Grael, Compumedics, Australia) monitoring at the sleep center, the PSG-derived sleep parameters of the first night were used to exclude sleep disorders other than ID. Within a week after the completion of the second night PSG, an adaptation of an MRI scanning session (3.0T Prisma Scanner, Siemens Healthineers, Erlangen, Germany) was conducted to promote sleep and to reduce the first-night effect at the Center for MRI Research, Peking University. Participants were informed about the requirements and procedures while lying in the scanner with a 64-channel MRI-compatible EEG cap (Brain Products, Munich, Germany) and underwent 6 min of T1-weighted scanning and 30 min of BOLD fMRI scanning. Approximately one week later, the experimental sleep EEG-fMRI recordings were conducted at the participants' usual bedtime, at which time the patients had not experienced sleep deprivation. EEG-fMRI data obtained here were included in the final analysis.

EEG-fMRI data acquisition

EEG-fMRI data during the night were acquired using a 3.0T Prisma Scanner and the 64-channel MR-compatible EEG system. The subjects were asked to lie on their backs during the entire scan. Earplugs and foam cushions were provided to offer noise protection and to reduce movement-related EEG artifacts. The resistance of the reference and ground channels was kept below 10 kΩ, whereas the resistance of the other channels was kept below 20 kΩ. EEG data were digitized at a 5,000 Hz sampling rate, and data were filtered with a low cutoff of 10 sec and a high cutoff of 250 Hz. The resistance of all the channels was verified again before MR scanning was initiated. Wires connecting the cap and the amplifiers were fixed to avoid any potential vibration during MR scanning.

For registration purposes, high-resolution T1-weighted structural images were acquired by using a three-dimensional magnetization-prepared rapid acquisition gradient-echo sequence (repetition time (TR) =

2530 ms, echo time (TE) = 2.98 ms, inversion time = 1100 ms, flip angle (FA) = 7°, number of slices = 192, and voxel size = 0.5 × 0.5 × 1 mm^3).

Subjects were scanned during the first half of the night, starting at their habitual bedtime. The sleep BOLD fMRI images were obtained with a gradient echo-planar sequence (TR = 2000 ms, TE = 30 ms, FA = 90°, number of slices = 33, slice thickness = 3.5 mm, gap = 0.7 mm, matrix = 64 × 64, and in-plane resolution = 3.5 × 3.5 mm^2). The “sleep” session was terminated when the participants wanted to get out, or a maximum of 4096 volumes was reached.

EEG data preprocessing and sleep stage scoring

EEG data were preprocessed using Brain Vision Analyzer 2.1 (Brain Products). MR gradient and ballistocardiograph artifacts were removed by the average artifact subtraction method (Allen, Josephs, & Turner, 2000). The lowest residual gradient artifacts (16.5 Hz) did not compromise spindle frequency (11-16 Hz). Data were downsampled to 500 Hz while MR gradient removal was performed. Then, data were rereferenced to averaged mastoids and temporally filtered (10-100 Hz for electromyogram channels and 0.3-35 Hz for the other channels). Sleep stages were visually scored in accordance with the guidelines of the American Academy of Sleep Medicine (AASM) (Berry et al., 2016) by a registered polysomnographic technologist.

Spindle detection

We detected spindles using offline algorithms adopted from references (http://fieldtrip.fcdonders.nl/) (Mölle, Marshall, Gais, & Born, 2002). For each subject, individual peak frequencies were identified in the NREM sleep power spectra for fast and slow spindles. The root mean square (RMS) of the filtered signal in a specific time window (0.2 sec) was determined for each sample, resulting in a moving RMS signal. The moving RMS signal was further smoothed by a moving average of another window length (again 0.2 sec). After smoothing, algorithms started to detect spindles by setting a specific time frame threshold in the moving RMS (0.5-3 sec). The spindles were identified during all NREM epochs at Cz, and the analytical amplitude was extracted after applying a Hilbert transform. The highest 25% of RMS amplitudes of the filtered EEG signal segments were classified as spindles. Spindle detection was visually verified by an expert scorer after automated detection. The total number of spindle events in NREM sleep was compiled. For each detected spindle event, three variables of interest were considered: density (spindles per min), duration (definite onset-to-offset time, in sec), and amplitude (maximal peak-to-peak voltage, in μV).

fMRI data processing

After identifying spindles on the EEG data, fMRI volumes corresponding to the onset of spindles were located according to the EEG-MRI synchronization markers (Figure S2). Then, for spindles with long temporal intervals (> 50 sec) between two consecutive occurrences, fMRI data starting from 20 sec before the onset of a spindle until 30 sec after the same onset were extracted as an fMRI episode. For

continuous spindles with relatively short temporal intervals (< 50 sec), fMRI data starting from 20 sec before the onset of the first spindle until 30 sec after the onset of the last spindle were extracted as an episode. Each fMRI episode was treated as a session in the following general linear model (GLM) analysis (Figure S3).

Functional volumes were analyzed by using Statistical Parametric Mapping 12 (SPM12, Welcome Department of Imaging Neuroscience, University College, London, UK; http://www.fil.ion.ucl.ac.uk/spm/software/spm12/) implemented in MATLAB (Ver. 8.5 R2013b) for Windows (Microsoft, Inc.). The preprocessing steps included slice timing, realignment, coregistration, segmentation, spatial normalization (voxel size, 3 × 3 × 3 mm^3) to the International Consortium for Brain Mapping standard template, and smoothing (full width at half-maximum, 6 mm). Episodes of fMRI data with excessive head motion (displacement greater than 2 mm or rotation larger than 2 degrees of each volume relative to the average of all volumes in an episode (Zou et al., 2017) were excluded. Then, the onset time of the spindles was convolved with the hemodynamic response function and modeled as regressors for fMRI analysis; pre- and post-spindle sleep data in the same episode served as the baseline. Head motion parameters estimated during realignment (translations in the *x*, *y*, and *z* directions and rotations around the *x*, *y*, and *z* axes) were included in the matrix as variables of no interest. High-pass filtering was implemented in the design using a cutoff at 128 sec to remove low-frequency drifts from the time series. These analyses generated spindle-related contrast maps for all spindle events. Then, contrast maps were entered into second-level GLMs for group-level analysis.

One-sample *t*-tests were conducted for the brain activities time-locked to spindles within each group, and two-sample *t*-tests adjusting for age, gender, and education level were used between patients with ID and HCs for between-group comparisons. Statistical inferences for one sample *t*-tests were performed at a threshold of $p < 0.001$ (uncorrected) and $p < 0.005$ (uncorrected) for two-sample *t*-test at the voxel level. One- and two-sample *t*-tests were performed at a threshold of $p < 0.05$, familywise error (FWE) corrected at the cluster level.

Statistical analysis

Data in the table are presented as the mean ± standard deviation (SD) or quartile. The normal distribution of the data and the homogeneity of variance were tested using Shapiro-Wilk and Levene's tests, respectively. Two-sample *t*-tests, Mann-Whitney *U* tests, and c^2 tests were used to detect group differences between patients with ID and HCs in demographic, clinical and spindle measures using SPSS 26.0 software. The level of significance was set at $p < 0.05$.

Exploratory correlation analysis

Brain regions with significant between-group difference were identified as the regions of interest and were saved as masks, then spindle-related contrast maps of subjects from the two groups were entered to extract the BOLD signals. After that, Spearman's correlation analysis in all subjects and in ID patients was

adopted to investigate the relationship between spindle-related BOLD values and SDS scores, SAS scores, PSQI scores, sleep onset latency (SOL), and total sleep time (TST) (two quantitative indices of the PSQI).

Results

Demographic and clinical characteristics

Sixteen ID patients with moderate to severe disease (ISI score no less than 15) and thirty HCs participated in this study. The demographic and clinical data of the participants are presented in Table 1. No significant difference was detected between ID patients and HCs in terms of age, gender, BMI, or education level (all *ps* > 0.05). The mean duration of insomnia was 9.38 years. The ID group had significantly higher scores on the SDS, SAS, and PSQI, as well as longer SOL and shorter TST than the HC group (all *ps* < 0.001).

Spindle-related electrophysiological data

A total of 4914 spindles were included in the final analysis (853 for the ID group and 4061 for the HC group). ID patients showed significantly lower spindle numbers and density than HCs (*ps* < 0.05). There was no significant difference in measures of spindle duration or amplitude between the two groups (*ps* > 0.05) (Table 2).

Spindle-related brain activations

Spindle-related brain activations are shown in Figure 2. Significantly positive responses related to spindles were identified in the frontal, temporal, parietal, and fusiform gyrus, insula, cingulate gyrus, amygdala, striatum, hippocampus, and parahippocampus in both the HC (Figure 2A) and the ID (Figure 2B) groups. In addition, HCs showed activations in the lingual gyrus, calcarine, occipital gyrus, thalamus, and brainstem. No significant brain deactivation was observed in either group.

Comparison between the two groups (HCs minus ID patients) revealed a significantly decreased response in the anterior and middle cingulate gyrus in the ID group (cluster size 3834 mm^3, peak coordinates [-3, 9, 24]) (Figure 3).

Relationship between spindle-related brain activations and clinical characteristics

When putting the two groups together, as shown in Figure 4**,** spindle-related BOLD values in the cingulate gyrus negatively correlated with SDS scores (r = -0.454, p = 0.002, Figure 4A), SAS scores (r = -0.362, p = 0.014, Figure 4B), and SOL (r = -0.374, p = 0.010, Figure 4C), while positively correlated with TST (r = 0.343, p = 0.020, Figure 4D). No significant correlation was observed between BOLD values and total PSQI scores. In the ID group, spindle-related BOLD values in the cingulate gyrus negatively correlated with SDS scores (r = -0.650, p = 0.006, Figure 5) even when the significance for correlation analysis was set as p < 0.01 (0.05/5 for Bonferroni correction considering five clinical parameters).

In addition, partial correlation analysis was applied to estimate the relationship between spindle-related BOLD values and clinical characteristics when controlling age, gender, and education level. The results showed that spindle-related BOLD values in the cingulate gyrus positively correlated with TST (r = 0.313, p = 0.041), while negatively correlated with SDS scores (r = -0.308, p = 0.045), in the pooled sample.

Discussion

The primary finding of this study was the significantly decreased spindle-related BOLD signals in the cingulate gyrus in ID patients. Exploratory analysis revealed that decreased BOLD values in the cingulate gyrus had significant correlations with higher SDS and SAS scores, longer SOL and shorter TST, suggesting that spindle-related brain activations in the cingulate gyrus are relevant for emotional regulation and subjective sleep quality.

Decreased spindle-related brain activations in the cingulate gyrus correlated with clinical indices

Earlier EEG-fMRI studies conducted with healthy young subjects (Andrade et al., 2011; Bergmann et al., 2012; Caporro et al., 2012; Schabus et al., 2007) and patients with epilepsy (Caporro et al., 2012) reported spindles to be associated with increased signals in the anterior cingulate cortex (ACC) and middle cingulate cortex (MCC). Our results confirmed the findings of previous studies and extended them to insomnia sufferers. Furthermore, we found significant association between spindle-related BOLD responses in the cingulate gyrus and emotional regulation, subjective sleep quality based on exploratory analysis.

The negative correlation between spindle-related brain activations in the cingulate gyrus and SOL, as well as the positive correlation between spindle-related brain activations in the cingulate gyrus and TST, were consistent with previous structural and functional imaging findings (O'Byrne, Berman Rosa, Gouin, & Dang-Vu, 2014). A morphometric analysis revealed that the ACC volumes in insomnia patients were significantly larger than those in HCs; the increased ACC volumes correlated with prolonged SOL and wake time after sleep onset (WASO) (Winkelman et al., 2013). In a positron emission tomography study, patients with insomnia were found to have a relatively smaller reduction in metabolism from wakefulness to NREM sleep in the ACC than controls (Nofzinger et al., 2004); the increased relative cerebral metabolic rate in the ACC during NREM sleep correlated with increased subjective and objective WASO (Nofzinger et al., 2006). In addition, insomnia patients in proton magnetic resonance spectroscopy studies exhibited regional reductions in gamma-aminobutyric acid, one of the primary inhibitory neurotransmitters in human brain, compared with controls (Plante, Jensen, Schoerning, & Winkelman, 2012). Further exploratory analysis suggested a significant positive association between gamma-aminobutyric acid levels in the ACC and TST in the second night of sleep in the laboratory (Spiegelhalder et al., 2016). Recently, fMRI studies have explored the regional homogeneity (ReHo) and functional connectivity (FC) characteristics of brain regions associated with insomnia and their relationship with sleep indices. In Dai's study, insomnia patients showed lower ReHo in bilateral cingulate gyrus (Dai et al., 2014). However, Wang et al. found increased ReHo in the right ACC while decreased ReHo in the right MCC in insomnia

patients compared with controls (Wang et al., 2016). fMRI-based voxel-mirrored homotopic connectivity (VMHC) provides a feasible way to observe insomnia-related interhemisphere coordination alterations. Both healthy participants with insomnia symptoms (Li et al., 2017) and insomnia patients (Yan et al., 2018) showed increased VMHC in the ACC. Furthermore, VMHC values in the ACC positively correlated with SOL (Yan et al., 2018).

Notably, we found significant negative correlations between BOLD values during spontaneous spindle events in the cingulate gyrus and scores on the SDS and SAS. This finding is consistent with previous fMRI studies showing significant correlations between lower ReHo in bilateral cingulate gyrus and higher anxiety (Dai et al., 2014), between decreased ReHo in the right MCC and increased SDS, SAS scores (Wang et al., 2016), and between increased VMHC values in the left ACC and increased SDS scores (Yan et al., 2018). Generally, insomnia patients have more depression and anxiety symptoms than good sleepers; insomnia is a common comorbidity symptom in patients with depressive and anxiety disorders (Soehner & Harvey, 2012; Sunderajan et al., 2010). Meanwhile, insomnia sufferers are more susceptible to developing depression and anxiety (Hertenstein et al., 2019). The ACC, a part of the emotional circuitry, has been thought to be an important area in the processing of depression and anxiety (Bush, Luu, & Posner, 2000). The increased FC in the ACC associated with both subjective sleep quality and depressive symptoms; mediation analysis further showed this FC underlie the association of the depression with poor sleep quality (Cheng, Rolls, Ruan, & Feng, 2018). Patients with major depressive disorder (MDD) and people who are at high risk for MDD (Sesso et al., 2017) had significantly lower spindle activity compared with HCs (Lopez, Hoffmann, & Armitage, 2010). Less sleep spindle activity in early adolescence was associated with more depressive symptoms and worse subjectively reported sleep quality (Hamann, Rusterholz, Studer, Kaess, & Tarokh, 2019). Children and adolescents suffering from social anxiety disorder showed a widespread reduction in fast spindle activity with negative correlation with negative stimuli arousal rate (Wilhelm, Groch, Preiss, Walitza, & Huber, 2017). Spindle density of youths with general anxiety disorder, though did not differ from that of HCs, positively correlated with worry (Meers, Ferri, Bruni, & Alfano, 2020). These findings reflect that spindle-related BOLD signals in the cingulate gyrus may be the common neurophysiological basis of insomnia, depression, and anxiety.

Spindle characteristics between the two groups

Our results showed significantly lower spindle numbers and density in ID patients than in HCs. This is inconsistent with Bastien's finding, in which no significant group difference was found (Bastien et al., 2009). This may be related to methodological issues, the study population, and the experimental environment. First, spindles were scored visually by two independent technicians at C3 in N2 sleep throughout the night in Bastien's study, whereas we automatically detected spindles followed by manually checks at Cz in NREM sleep during 2h16min scanning period. Second, the definition of spindles differs in frequency and amplitude range, of which 12-14 Hz and 20-40 μV, respectively, were used in Bastien's study. Our study defined the frequency range of spindles as 11-16 Hz, with no amplitude requirement. Third, the insomnia criteria adopted in Bastien's study vary from ours. In their study, patients with insomnia were required to have a duration of more than 6 months and meet the diagnostic criteria

for chronic psychophysiological insomnia. In our study, patients with ID should have a duration of at least 3 months. Fourth, Bastien's study was conducted at the sleep laboratory, while participants' sleep might be disturbed by the noise of the MRI in this study. We did not find group difference in spindle duration or amplitude. However, Andrillon et al. found that insomnia patients exhibited increased spindle duration and reduced spindle amplitude compared with HCs (Andrillon et al., 2020). Potential drug-induced effects could have contaminated their findings on spindle characteristics, as patients in their study were allowed to take medicine, including antidepressants (Dotan, Suraiya, & Pillar, 2008), benzodiazepines or benzodiazepine-like drugs (Hirshkowitz, Thornby, & Karacan, 1982; Kaestner, Wixted, & Mednick, 2013; Plante et al., 2015; Suetsugi, Mizuki, Ushijima, Kobayashi, & Watanabe, 2001; Wamsley et al., 2013), which led to increased spindles. Normand and colleagues found that paradoxical insomniacs had significantly shorter spindles than HCs, but patients with psychophysiological insomnia and HCs did not differ in spindle length (Normand et al., 2016). Variations in spindle parameters may be only seen in a particular subtype of insomnia. Insomnia patients in our study were recruited from the general public, not through physician referral to a sleep clinic, and were diagnosed according to subjective reports without excluding "pseudo insomnia" based on PSG data (i.e., sleep efficiency less than 85%; WASO more than 30 min, etc. (Baglioni et al., 2014)). The ID group exhibited poor sleep continuity with significantly shorter TST and lower SE, which is in line with the results shown in a meta-analysis of polysomnographic studies (Baglioni et al., 2014). Our patients had no significantly increased SOL, increased WASO, reduced N3% or reduced REM% (Table S1). Thus, patients with moderate to severe insomnia assessed by ISI score greater than 15 were included in this study, in order to minimize the impact of ID heterogeneity on the results.

There are several limitations in the present study. First, we detected spindles in only one bandwidth rather than dividing them into slow spindles (11-13 Hz) and fast spindles (13-16 Hz). These two types of spindles share a common activation pattern, involving both the thalamus, ACC, left anterior insula, and bilateral superior temporal gyrus. The activity associated with slow spindles largely corresponded to the common activation pattern, with additional recruitment of the right superior frontal gyrus. Fast spindles were associated with a number of significant activations beyond the common pattern in the supplementary motor area, sensorimotor area, and MCC (Schabus et al., 2007). Second, spindles in N2 and N3 were analyzed together. Evidence suggests that spindle characteristics during N2 and N3 differ in the following aspects. Spindles in N2 are more frequent, larger in amplitude and in activity integrated over time (Cox, Schapiro, Manoach, & Stickgold, 2017). Besides, slow wave activity in N3 synchronized with spindles are very likely to influence spindle-related brain responses. However, only a small number of patients with ID entered N3, and limited time was spent in this stage (5 volunteers for a total duration of 32.5 min; Table S2). Finally, the small sample size limited the explanation of the role of spindles in sleep protection in ID. Future investigations with larger sample sizes and a homogenous group of insomnia patients are needed.

Conclusions

In conclusion, this pilot study identified significantly decreased spindle-related brain activation patterns in patients with ID. The hypoactivation associated with spindles in the cingulate gyrus negatively correlated with SDS scores, SAS scores, subjective SOL, and positively correlated with subjective TST. The current study provides further evidence for spindle deficits in patients with ID and supports that decreased BOLD signals in the cingulate gyrus are involved in the pathological mechanisms of poor emotional regulation and subjective sleep quality in ID.

Declarations

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Declarations

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References

Adams, R. J., Appleton, S. L., Taylor, A. W., Gill, T. K., Lang, C., McEvoy, R. D., & Antic, N. A. (2017). Sleep health of Australian adults in 2016: Results of the 2016 Sleep Health Foundation national survey. Sleep Health, 3(1), 35-42. doi:10.1016/j.sleh.2016.11.005.

Allen, P. J., Josephs, O., & Turner, R. (2000). A method for removing imaging artifact from continuous EEG recorded during functional MRI. Neuroimage, 12(2), 230-239. doi:10.1006/nimg.2000.0599.

American-Psychiatric-Association. (2013). Diagnostic and statistical manual of mental disorders (5th ed.). Washington, DC: American Psychiatric Publishing.

Andrade, K. C., Spoormaker, V. I., Dresler, M., Wehrle, R., Holsboer, F., Samann, P. G., & Czisch, M. (2011). Sleep spindles and hippocampal functional connectivity in human NREM sleep. The Journal of neuroscience : the official journal of the Society for Neuroscience, 31(28), 10331-10339. doi:10.1523/JNEUROSCI.5660-10.2011.

Andrillon, T., Solelhac, G., Bouchequet, P., Romano, F., Le Brun, M. P., Brigham, M., Chennaoui, M., & Léger, D. (2020). Revisiting the value of polysomnographic data in insomnia: More than meets the eye. Sleep medicine, 66, 184-200. doi:10.1016/j.sleep.2019.12.002.

Baglioni, C., Regen, W., Teghen, A., Spiegelhalder, K., Feige, B., Nissen, C., & Riemann, D. (2014). Sleep changes in the disorder of insomnia: A meta-analysis of polysomnographic studies. Sleep Medicine Reviews, 18(3), 195-213. doi:10.1016/j.smrv.2013.04.001.

Bastien, C. H., St-Jean, G., Turcotte, I., Morin, C. M., Lavallee, M., & Carrier, J. (2009). Sleep spindles in chronic psychophysiological insomnia. Journal of psychosomatic research, 66(1), 59-65. doi:10.1016/j.jpsychores.2008.05.013.

Beattie, L., Espie, C. A., Kyle, S. D., & Biello, S. M. (2015). How are normal sleeping controls selected? A systematic review of cross-sectional insomnia studies and a standardized method to select healthy controls for sleep research. Sleep medicine, 16(6), 669-677. doi:10.1016/j.sleep.2015.01.010.

Bergmann, T. O., Molle, M., Diedrichs, J., Born, J., & Siebner, H. R. (2012). Sleep spindle-related reactivation of category-specific cortical regions after learning face-scene associations. Neuroimage, 59(3), 2733-2742. doi:10.1016/j.neuroimage.2011.10.036.

Berry, R. B., Brooks, R., Gamaldo, C. E., Harding, S. M., Lloyd, R. M., Marcus, C. L., & Vaughn, B. V. (2016). The AASM manual for the scoring of sleep and associated events: Rules, terminology and technical specifications, version 2.3. Darien, Illinois: American Academy of Sleep Medicine.

Besset, A., Villemin, E., Tafti, M., & Billiard, M. (1998). Homeostatic process and sleep spindles in patients with sleep-maintenance insomnia: Effect of partial (21 h) sleep deprivation. Electroencephalography and clinical neurophysiology, 107(2), 122-132. doi:10.1016/s0013-4694(98)00048-0.

Bush, G., Luu, P., & Posner, M. I. (2000). Cognitive and emotional influences in anterior cingulate cortex Trends in cognitive sciences, 4 (6), 215–222. doi:10.1016/s1364-6613(00)01483-2.

Caporro, M., Haneef, Z., Yeh, H. J., Lenartowicz, A., Buttinelli, C., Parvizi, J., & Stern, J. M. (2012). Functional MRI of sleep spindles and K-complexes. Clinical neurophysiology : official journal of the International Federation of Clinical Neurophysiology, 123(2), 303-309. doi:10.1016/j.clinph.2011.06.018.

Cheng, W., Rolls, E. T., Ruan, H., & Feng, J. (2018). Functional connectivities in the brain that mediate the association between depressive problems and sleep quality. JAMA Psychiatry, 75(10), 1052-1061. doi:10.1001/jamapsychiatry.2018.1941.

Cox, R., Schapiro, A. C., Manoach, D. S., & Stickgold, R. (2017). Individual differences in frequency and topography of slow and fast sleep spindles. Frontiers in human neuroscience, 11, 433. doi:10.3389/fnhum.2017.00433.

Dai, X. J., Peng, D. C., Gong, H. H., Wan, A. L., Nie, X., Li, H. J., & Wang, Y. X. (2014). Altered intrinsic regional brain spontaneous activity and subjective sleep quality in patients with chronic primary insomnia: A resting-state fMRI study. Neuropsychiatric disease and treatment, 10, 2163-2175. doi:10.2147/NDT.S69681.

Dang-Vu, T. T., Bonjean, M., Schabus, M., Boly, M., Darsaud, A., Desseilles, M., Degueldre, C., Balteau, E., Phillips, C., Luxen, A., Sejnowski, T. J., & Maquet, P. (2011). Interplay between spontaneous and induced brain activity during human non-rapid eye movement sleep. Proceedings of the National Academy of Sciences of the United States of America, 108(37), 15438-15443. doi:10.1073/pnas.1112503108.

Dang-Vu, T. T., McKinney, S. M., Buxton, O. M., Solet, J. M., & Ellenbogen, J. M. (2010). Spontaneous brain rhythms predict sleep stability in the face of noise. Current biology : CB, 20(15), R626-R627. doi:10.1016/j.cub.2010.06.032.

Dang-Vu, T. T., Salimi, A., Boucetta, S., Wenzel, K., O'Byrne, J., Brandewinder, M., Berthomier, C., & Gouin, J. P. (2015). Sleep spindles predict stress-related increases in sleep disturbances. Frontiers in human neuroscience, 9, 68. doi:10.3389/fnhum.2015.00068.

Dotan, Y., Suraiya, S., & Pillar, G. (2008). Sleep spindles in post traumatic stress disorder: Significant importance of selective serotonin reuptake inhibitors. Harefuah, 147(10), 763-840.

Fogel, S., Albouy, G., King, B. R., Lungu, O., Vien, C., Bore, A., Pinsard, B., Benali, H., Carrier, J., & Doyon, J. (2017). Reactivation or transformation? Motor memory consolidation associated with cerebral activation time-locked to sleep spindles. PLoS One, 12(4), e0174755. doi:10.1371/journal.pone.0174755.

Hamann, C., Rusterholz, T., Studer, M., Kaess, M., & Tarokh, L. (2019). Association between depressive symptoms and sleep neurophysiology in early adolescence. Journal of child psychology and psychiatry, and allied disciplines, 60(12), 1334-1342. doi:10.1111/jcpp.13088.

Hertenstein, E., Feige, B., Gmeiner, T., Kienzler, C., Spiegelhalder, K., Johann, A., Jansson-Frojmark, M., Palagini, L., Rucker, G., Riemann, D., & Baglioni, C. (2019). Insomnia as a predictor of mental disorders: A systematic review and meta-analysis. Sleep Medicine Reviews, 43, 96-105. doi:10.1016/j.smrv.2018.10.006.

Hirshkowitz, M., Thornby, J. I., & Karacan, I. (1982). Sleep spindles: Pharmacological effects in humans. Sleep, 5 (1), 85-94. doi:10.1093/sleep/5.1.85.

Huang, P., Wei, D., Xiao, W., Yuan, M., Chen, N., Wei, X., Xie, J., Lu, J., Xia, X., Lu, M., Shen, S., He, J., & Qiu, X. (2020). Maternal dietary patterns and depressive symptoms during pregnancy: The Born in Guangzhou Cohort Study. Clinical nutrition (Edinburgh, Scotland), S0261-5614(0220)30659-30652. doi:10.1016/j.clnu.2020.11.038.

Israel, B., Buysse, D. J., Krafty, R. T., Begley, A., Miewald, J., & Hall, M. (2012). Short-term stability of sleep and heart rate variability in good sleepers and patients with insomnia: For some measures, one night is enough. Sleep, 35(9), 1285-1291. doi:10.5665/sleep.2088.

Kaestner, E. J., Wixted, J. T., & Mednick, S. C. (2013). Pharmacologically increasing sleep spindles enhances recognition for negative and high-arousal memories. Journal of cognitive neuroscience, 25(10), 1597-1610. doi:10.1162/jocn_a_00433.

Kerkhof, G. A. (2017). Epidemiology of sleep and sleep disorders in The Netherlands. Sleep medicine, 30, 229-239. doi:10.1016/j.sleep.2016.09.015.

Lecrubier, Y., Sheehan, D. V., Weiller, E., Amorim, P., Bonora, I., Harnett Sheehan, K., Janavs, J., & Dunbar, G. C. (1997). The Mini International Neuropsychiatric Interview (MINI). A short diagnostic structured interview: Reliability and validity according to the CIDI. European psychiatry : the journal of the Association of European Psychiatrists, 12(5), 224-231. doi:10.1016/S0924-9338(97)83296-8.

Li, X., Guo, S., Wang, C., Wang, B., Sun, H., & Zhang, X. (2017). Increased interhemispheric resting-state functional connectivity in healthy participants with insomnia symptoms: A randomized clinical consort study. Medicine (Baltimore), 96(27), e7037. doi:10.1097/MD.0000000000007037.

Liu, L., Pang, R., Sun, W., Wu, M., Qu, P., Lu, C., & Wang, L. (2013). Functional social support, psychological capital, and depressive and anxiety symptoms among people living with HIV/AIDS employed full-time. BMC Psychiatry, 13, 324. doi:10.1186/1471-244X-13-324.

Lopez, J., Hoffmann, R., & Armitage, R. (2010). Reduced sleep spindle activity in early-onset and elevated risk for depression. Journal of the American Academy of Child and Adolescent Psychiatry, 49(9), 934-943. doi:10.1016/j.jaac.2010.05.014.

Meers, J. M., Ferri, R., Bruni, O., & Alfano, C. A. (2020). Sleep spindle density is associated with worry in children with generalized anxiety disorder and healthy controls. Journal of affective disorders, 260, 418-

425. doi:10.1016/j.jad.2019.09.065.

Mölle, M., Marshall, L., Gais, S., & Born, J. (2002). Grouping of spindle activity during slow oscillations in human non-rapid eye movement sleep. The Journal of neuroscience : the official journal of the Society for Neuroscience, 22(24), 10941-10947. doi:10.1523/JNEUROSCI.22-24-10941.2002.

Morin, C. M., Belleville, G., Bélanger, L., & Ivers, H. (2011). The Insomnia Severity Index: Psychometric indicators to detect insomnia cases and evaluate treatment response. Sleep, 34(5), 601-608. doi:10.1093/sleep/34.5.601.

Nofzinger, E. A., Buysse, D. J., Germain, A., Price, J. C., Miewald, J. M., & Kupfer, D. J. (2004). Functional neuroimaging evidence for hyperarousal in insomnia. The American journal of psychiatry, 161(11), 2126-2128. doi:10.1176/appi.ajp.161.11.2126.

Nofzinger, E. A., Nissen, C., Germain, A., Moul, D., Hall, M., Price, J. C., Miewald, J. M., & Buysse, D. J. (2006). Regional cerebral metabolic correlates of WASO during NREM sleep in insomnia. J Clin Sleep Med, 2(3), 316-322. doi:10.1016/j.seizure.2005.11.001.

Normand, M. P., St-Hilaire, P., & Bastien, C. H. (2016). Sleep spindles characteristics in insomnia sufferers and their relationship with sleep misperception. Neural Plast, 2016, 6413473. doi:10.1155/2016/6413473.

O'Byrne, J. N., Berman Rosa, M., Gouin, J. P., & Dang-Vu, T. T. (2014). Neuroimaging findings in primary insomnia. Pathologie-biologie, 62(5), 262-269. doi:10.1016/j.patbio.2014.05.013.

Plante, D. T., Goldstein, M. R., Cook, J. D., Smith, R., Riedner, B. A., Rumble, M. E., Jelenchick, L., Roth, A., Tononi, G., Benca, R. M., & Peterson, M. J. (2015). Effects of oral temazepam on sleep spindles during non-rapid eye movement sleep: A high-density EEG investigation. European neuropsychopharmacology : the journal of the European College of Neuropsychopharmacology, 25(10), 1600-1610. doi:10.1016/j.euroneuro.2015.06.005.

Plante, D. T., Jensen, J. E., Schoerning, L., & Winkelman, J. W. (2012). Reduced gamma-aminobutyric acid in occipital and anterior cingulate cortices in primary insomnia: A link to major depressive disorder? Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology, 37(6), 1548-1557. doi:10.1038/npp.2012.4.

Schabus, M., Dang-Vu, T. T., Albouy, G., Balteau, E., Boly, M., Carrier, J., Darsaud, A., Degueldre, C., Desseilles, M., Gais, S., Phillips, C., Rauchs, G., Schnakers, C., Sterpenich, V., Vandewalle, G., Luxen, A., & Maquet, P. (2007). Hemodynamic cerebral correlates of sleep spindles during human non-rapid eye movement sleep. Proceedings of the National Academy of Sciences of the United States of America, 104(32), 13164–13169. doi:10.1073/pnas.0703084104.

Sesso, G., Bat-Pitault, F., Guyon, A., Plancoulaine, S., Banfi, T., Milioli, G., Parrino, L., Faraguna, U., & Franco, P. (2017). Electrophysiological and microstructural features of sleep in children at high risk for depression: A preliminary study. Sleep medicine, 36, 95-103. doi:10.1016/j.sleep.2017.05.013.

Soehner, A. M., & Harvey, A. G. (2012). Prevalence and functional consequences of severe insomnia symptoms in mood and anxiety disorders: Results from a nationally representative sample. Sleep, 35(10), 1367-1375. doi:10.5665/sleep.2116.

Spiegelhalder, K., Regen, W., Nissen, C., Feige, B., Baglioni, C., Riemann, D., Hennig, J., & Lange, T. (2016). Magnetic resonance spectroscopy in patients with insomnia: A repeated measurement study. PLoS One, 11(6), e0156771. doi:10.1371/journal.pone.0156771.

Suetsugi, M., Mizuki, Y., Ushijima, I., Kobayashi, T., & Watanabe, Y. (2001). The effects of diazepam on sleep spindles: A qualitative and quantitative analysis. Neuropsychobiology, 43 (1), 49-53. doi:10.1159/000054865.

Sunderajan, P., Gaynes, B. N., Wisniewski, S. R., Miyahara, S., Fava, M., Akingbala, F., DeVeaugh-Geiss, J., Rush, A. J., & Trivedi, M. H. (2010). Insomnia in patients with depression: A STAR*D report. CNS spectrums, 15(6), 394-404. doi:10.1017/s1092852900029266.

Tsai, P. S., Wang, S. Y., Wang, M. Y., Su, C. T., Yang, T. T., Huang, C. J., & Fang, S. C. (2005). Psychometric evaluation of the Chinese version of the Pittsburgh Sleep Quality Index (CPSQI) in primary insomnia and control subjects. Quality of life research : an international journal of quality of life aspects of treatment, care and rehabilitation, 14(8), 1943-1952. doi:10.1007/s11136-005-4346-x.

Tyvaert, L., Levan, P., Grova, C., Dubeau, F., & Gotman, J. (2008). Effects of fluctuating physiological rhythms during prolonged EEG-fMRI studies. Clinical neurophysiology : official journal of the International Federation of Clinical Neurophysiology, 119(12), 2762-2774. doi:10.1016/j.clinph.2008.07.284.

Wamsley, E. J., Shinn, A. K., Tucker, M. A., Ono, K. E., McKinley, S. K., Ely, A. V., Goff, D. C., Stickgold, R., & Manoach, D. S. (2013). The effects of eszopiclone on sleep spindles and memory consolidation in schizophrenia: A randomized placebo-controlled trial. Sleep, 36(9), 1369-1376. doi:10.5665/sleep.2968.

Wang, T., Li, S., Jiang, G., Lin, C., Li, M., Ma, X., Zhan, W., Fang, J., Li, L., Li, C., & Tian, J. (2016). Regional homogeneity changes in patients with primary insomnia. European radiology, 26(5), 1292-1300. doi:10.1007/s00330-015-3960-4.

Wilhelm, I., Groch, S., Preiss, A., Walitza, S., & Huber, R. (2017). Widespread reduction in sleep spindle activity in socially anxious children and adolescents. Journal of psychiatric research, 88, 47-55. doi:10.1016/j.jpsychires.2016.12.018.

Wimmer, R. D., Astori, S., Bond, C. T., Rovo, Z., Chatton, J. Y., Adelman, J. P., Franken, P., & Luthi, A. (2012). Sustaining sleep spindles through enhanced SK2-channel activity consolidates sleep and elevates

arousal threshold. The Journal of neuroscience : the official journal of the Society for Neuroscience, 32(40), 13917-13928. doi:10.1523/JNEUROSCI.2313-12.2012.

Winkelman, J. W., Plante, D. T., Schoerning, L., Benson, K., Buxton, O. M., O'Connor, S. P., Jensen, J. E., Renshaw, P. F., & Gonenc, A. (2013). Increased rostral anterior cingulate cortex volume in chronic primary insomnia. Sleep, 36(7), 991-998. doi:10.5665/sleep.2794.

Yan, C. Q., Liu, C. Z., Wang, X., Huo, J. W., Zhou, P., Zhang, S., Fu, Q. N., Zhang, J., Wang, Z. Y., & Liu, Q. Q. (2018). Abnormal functional connectivity of anterior cingulate cortex in patients with primary insomnia: A resting-state functional magnetic resonance imaging study. Frontiers in aging neuroscience, 10, 167. doi:10.3389/fnagi.2018.00167.

Yu, D. S. (2010). Insomnia Severity Index: Psychometric properties with Chinese community-dwelling older people. Journal of advanced nursing, 66(10), 2350-2359. doi:10.1111/j.1365-2648.2010.05394.x.

Zhao, W., Van Someren, E. J. W., Li, C., Chen, X., Gui, W., Tian, Y., Liu, Y., & Lei, X. (2021). EEG spectral analysis in insomnia disorder: A systematic review and meta-analysis. Sleep Medicine Reviews, 59, 101457. doi:10.1016/j.smrv.2021.101457.

Zou, Q., Wu, X., Hu, J., Tang, W., Mao, Y., Zhu, J., Lu, L., Zhang, Y., & Gao, J. H. (2017). Longitudinal recovery of local neuronal activity and consciousness level in acquired brain injury. Human brain mapping, 38(7), 3579-3591. doi:10.1002/hbm.23611.

Figures

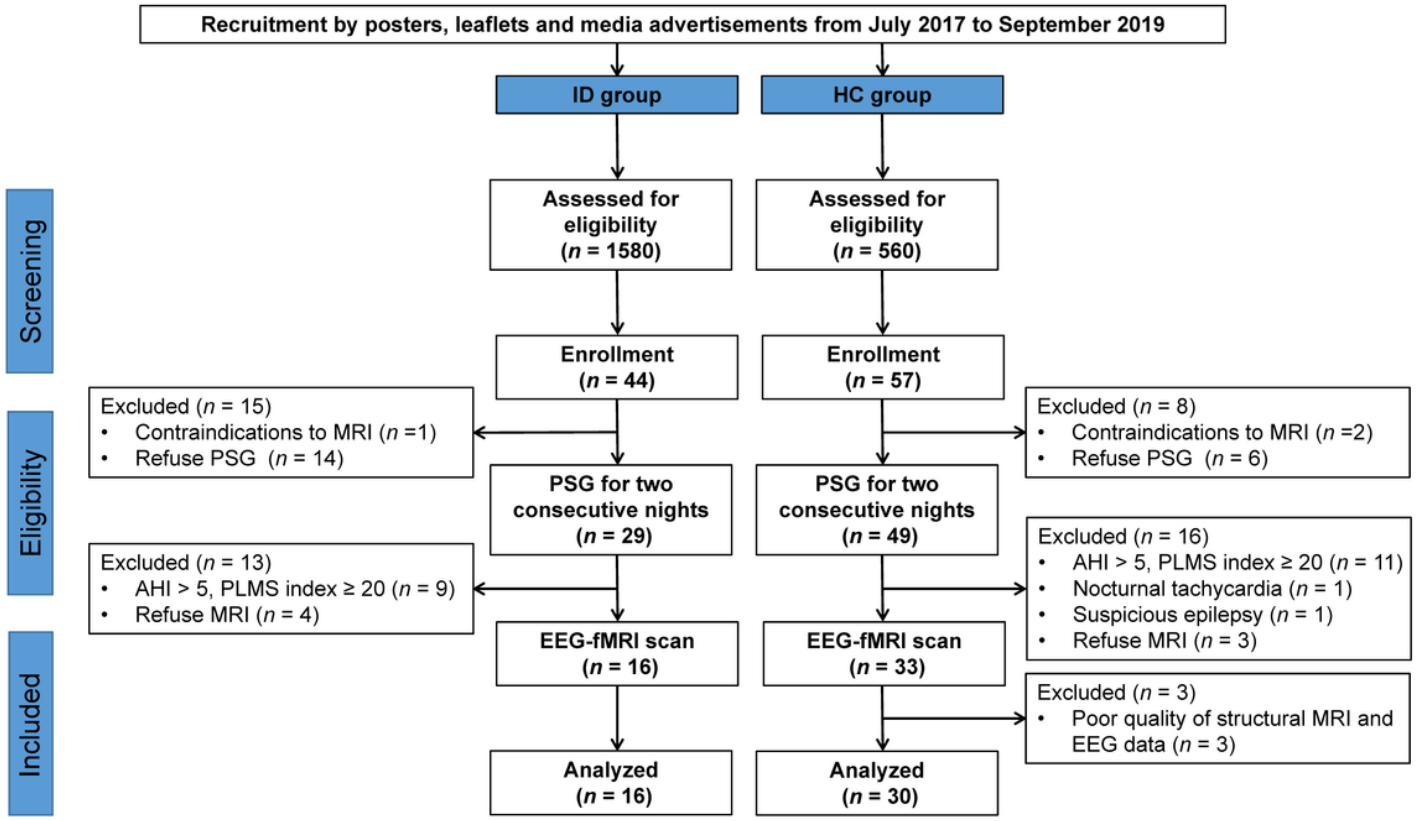

Figure 1

Flowchart of participants. AHI = apnea-hypopnea index. EEG = electroencephalogram. EEG-fMRI = electroencephalography and functional magnetic resonance imaging. HC = healthy control. ID = insomnia disorder. MRI = magnetic resonance imaging. PLMS = periodic leg movements during sleep. PSG = polysomnography.

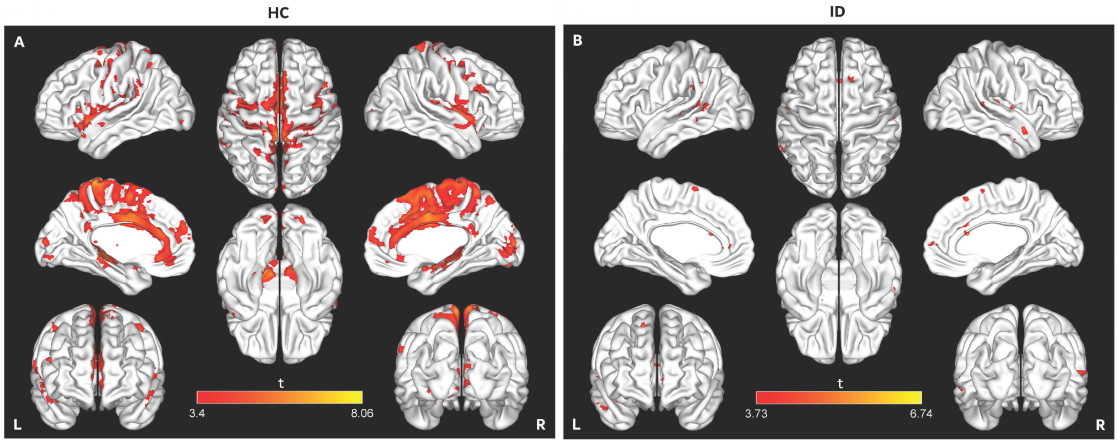

Figure 2

Brain activations associated with spindles. (A) Brain regions related to spindles in the HC group ($p < 0.001$ and cluster-level, FWE-corrected $p < 0.05$; L: left, R: right). (B) Brain regions related to spindles in the ID group ($p < 0.001$ and cluster-level, FWE-corrected $p < 0.05$; L: left, R: right).

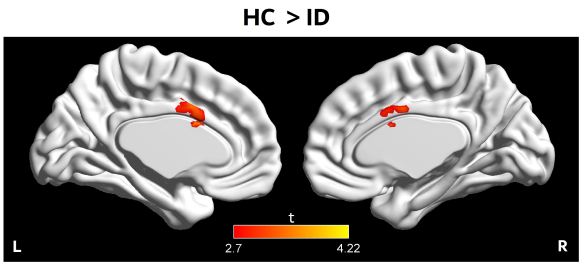

Figure 3

Brain activations differences between the HC and ID groups (p < 0.005 and cluster-level, FWE-corrected p < 0.05; L: left, R: right).

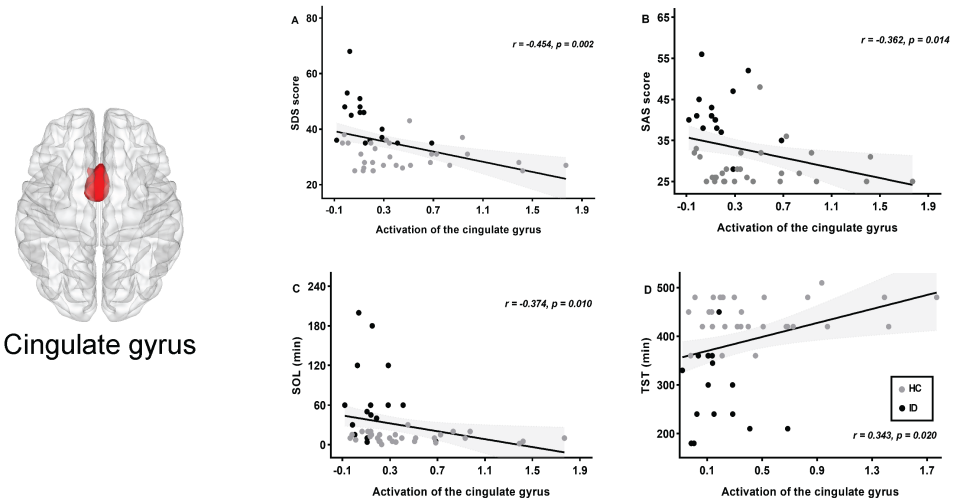

Figure 4

Correlations between activations of the cingulate gyrus and clinical characteristics in all subjects. (A) SDS scores (r = -0.454, p = 0.002). (B) SAS scores (r = -0.362, p = 0.014). (C) SOL (r = -0.374, p = 0.010). (D) TST (r = 0.343, p = 0.020).

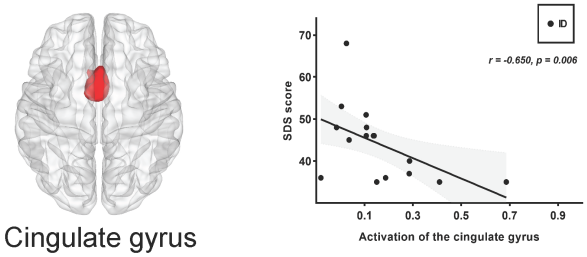

Figure 5

Correlation between activations of the cingulate gyrus and SDS scores (r = -0.650, p = 0.006) in the ID group.

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