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Original Article

# A study for the mechanism of sensory disorder in restless legs syndrome based on magnetoencephalography

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ABSTRACT

In spite of the relatively high incidence rate, the etiology and pathogenesis of restless legs syndrome (RLS) are still unclear. Long-term drug treatments fail to achieve satisfying curative effects, which is reflected by rebound and augmentation of related symptoms. An electrophysiological endophenotype experiment was done to investigate the mechanism of somatosensory disorder among RLS patients. Together with 15 normal subjects as the control group, with comparable ages and genders to the RLS patients, 15 primitive RLS patients were scanned by Magnetoencephalography (MEG) under natural conditions; furthermore, the somatosensory evoked magnetic field (SEF) with single and paired stimuli, was also measured. Compared to the control group, the SEF intensities of RLS patients' lower limbs were higher, and the paired-pulse depression (PPD) for SEF in RLS patients was attenuated. It was also revealed by time-frequency analysis of somatosensory induced oscillation (SIO) in RLS patients, that 93.3% of somatosensory induced Alpha (8–12 Hz) oscillations were successfully elicited, while 0% somatosensory induced Gamma (30–55 Hz) oscillations were elicited; which was significantly different from the control group. Additionally, in RLS patients exhibit increased excitability of the sensorimotor cortex, a remarkable abnormality existing in early somatosensory gating control (GC) and an attenuated inhibitory interneuron network, which consequently results in a compensatory mechanism through which RLS patients increase their attention-driven lower limb sensory gating control via somatosensory-induced Alpha (8–12 Hz) oscillation. This hyperexcitability, partially due to an electrocortical disinhibition, may have an important therapeutical implication, and become an important target of neuromodulatory interventions.

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## 1. Introduction

Restless legs syndrome also known as Willis-Ekbom's disease, has a relatively high incidence rate, but is often misdiagnosed [1]. RLS patients often have objectively painful sensations. However, apart from some mild demyelination-like lesions in the brain white matter [2–4], there have been no reported validated structural lesions in brain gray matter, indicating a brain functional disorder is the main factor in RLS. Functional brain imaging shows that some brain network deregulatory features exist in RLS patients [5–7].

From the electrophysiological perspective, RLS should be regarded as a complex sensorimotor disorder. Cortical, subcortical, spinal cord, and peripheral nerve generators are all involved in a network disorder, which results in enhanced excitability and/or decreased inhibition [8]. Studies indicate that the brain network functional hub spots are mainly located in substantia nigra Area A8 and A9, hypothalamus Area A11 and thalamus [9–11]. These areas' abnormal functions are probably associated with iron deficiency in the central nervous system and deregulation of neural transmitter systems.

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The iron deficiency in the central nervous system is the main symptom of RLS, it causes a demyelination in brain white matter, and interferes with neural transmitter systems as well as monoamine metabolism. Glutamate and GABA [12] homeostasis are all influenced by the brain's iron status [13]. Possible reasons causing the iron deficiency are the genetic susceptibility of some genes, inadequate uptake, inflammation regulation, and epigenetic mutations [14–16]. Detailed pathogenesis of RLS is complicated, and researchers could only speculate that it closely related to abnormal dopamine uptake and/or delivery systems' decline. However, there is still no model for a deeper understanding of RLS's mechanism [17–19].

Currently it is believed that a spinal cord-involved neural network disorder is the main pathogenesis of RLS. Electrophysiological research shows that spinal hyperexcitability causes the periodic leg movements (PLMs) in RLS; RLS patients always have a low pain threshold with associated lower limb pain, which suggests the presence of sensitization in the central nervous system (spinal cord or superspinal [8]); diencephalic-spinal dysfunction may result in the disinhibition of sensory inputs to the dorsal horn. Levodopa is secreted from the A11 region and participates in sensory-motor integration in the spinal cord. Supplementation with exogenous levodopa can improve sensorimotor symptoms [20]. Conversely, active or passive movement of the limbs can increase the sensory gain control of the spinal cord and filter too much redundant information to the center, in order to reduce the feeling of conflict and relieve the sensory symptoms [21].

RLS's sensory symptoms might be the core of its pathogenesis, since abnormal activity in basic sensorimotor and other related brain systems have been found [22]. Some high-level brain regions related to attention and alertness are also involved. Abnormal electrical activity in the brain probably could be used as the electrophysiological endophenotype to describe RLS's pathophysiological features [23]. To further elucidate the specific mechanism for sensorimotor disorder, high tempo-spatial resolution brain imaging tools are still required.

Current common methods to examine brain function include functional magnetic resonance imaging (fMRI), positron emission tomography (PET) and electroencephalography (EEG). However, the temporal resolution of fMRI and PET cannot reach the required level of precision. Although the time resolution of EEG is high enough, the spatial resolution of EEG is not high enough, which means it is difficult to locate the deep source, especially in the deep part of the cortex, such as the area supporting the lower extremities.

Magnetoencephalography (MEG) is currently one of the most powerful tools for brain functional disorders research due to its millisecond temporal resolution and millimeter scale spatial resolution. MEG maps the brain activity by recording the brain's magnetic fields. MEG measures the field from the ionic currents flowing in the dendrites of neurons during synaptic transmission. Notably magnetic fields are less distorted than electric fields by the skull and scalp, which results in better spatial resolution of the MEG. MEG is non-invasive, without ionizing radiation, as opposed to PET and has high temporal resolution as opposed to fMRI [24].

Based on previous research results, we hypothesized that [1] sensorimotor integration is abnormal at the cerebral cortex level in RLS, probably due to an impairment of the inhibitory intermediate neuron network, thus resulting in increased excitability of the primary sensory and motor cortices. Cortical excitability in RLS patients can be detected by examining somatosensory evoked magnetic field intensity [2]. Increased cortical excitability can be caused by an increased excitability of neural ensembles or by a decline of inhibition between neural ensembles. There may be different levels of gating mechanisms in some important nodes during the transmission of somatosensory information in the brain network. A paired-pulse depression test was conducted to test whether there was decreased inhibition in RLS [3]. The early somatosensory induced magnetic field, to some extent reflects the response state (excitability) of the partial somatosensory stimulation in the primary somatosensory region; the subsequent somatosensory oscillation activity reflects the process of transmitting, integrating and forming output of somatosensory information in the wider range of the brain. Somatosensory oscillations in different frequency bands reflect the synchronization status of neural ensembles in different parts and ranges. Using a time-frequency analysis of somatosensory oscillation, it can be demonstrated whether RLS patients exhibit hyperexcitability in somatosensory cortex, and it can also be demonstrated whether the brain has a corresponding compensatory strategy or a secondary change in the course of further somatosensory information transmission.

In order to gain better insight into the neural mechanism of RLS's sensory symptoms, we designed the following experiment using MEG, an experiment which could also provide guidance for RLS diagnosis and therapy; for example it could be a criteria for neuromodulation by Transcranial Magnetic Stimulation (TMS).

## 2. Material and methods

For this study, 15 RLS patients and 15 control subjects were recruited. Clinical manifestations of all subjects were evaluated. MEG and matching MRI 3D high resolution T1 scans were performed, capturing MEG, single-pulse SEF and paired-pulse SEF data, while subjects were in a neutral (non-task) state. Statistical analysis for somatosensory cortical excitation was made, and somatosensory gating rate and time-frequency oscillation was evaluated.

### *2.1. Inclusion criteria for experimental group and control group*

The RLS group: 15 cases who were outpatients at Xuanwu Hospital, Capital Medical University, Beijing, China.

Criteria for inclusion:

The subject meets the diagnostic criteria for RLS International Restless legs Syndrome Study Group (IRLSSG) in 2014.

[1] Severity score of IRLSSG grade > 11;
[2] Aged between 18~80 (fully understand the content of the informed consent);
[3] right-handed (Edinburgh Handedness Inventory > 40);
[4] Taking no medication of dopaminergic agents for four weeks before being re-cruited;
[5] Having signed the informed consent.

Criteria for exclusion:

I. Combined with severe anxiety: Hamilton Anxiety Rating Scale (HAM-A) > 21;
II. Combined with severe depression: Hamilton Depression Rating Scale (HAM-D) > 20;
III. Positive history of drug or alcohol abuse;
IV. Positive history of uremia, blood dialysis, hypoferric anemia, pregnancy, Sjogren syndrome, Parkinson's disease, peripheral neuropathy of diabetes, rheumatic arthritis, hypothyroidism, porphyria, and folate deficiency;
V. Diagnosed with other organic diseases of brain or other severe somatic diseases;
VI. Being a carrier of foreign bodies that could interfere with collection of magnetic field (ie intrauterine device) artificial tooth and so on.

The control group: 15 cases of healthy volunteers from the general population.

Criteria for inclusion:

I. Aged between 18 and 80 (fully understand the content of the informed consent), right-handed (Edinburgh Handedness Inventory > 40);
II. No severe somatic diseases or any mental or neurological diseases with con-firmed diagnosis, Mini-Mental State Examination (MMSE) score >28, Montreal Cognitive Assessment (MoCA) score >27, Neuropsychiatric Inventory (NPI) score of 0;
III. No family history of RLS;
IV. Comparable gender ratio, age and dominant handedness with RLS group;
V. Having signed the informed consent.

### 2.2. Data acquisition

#### 2.2.1. General information collection form

The information collection form covered general information about each subject: name, age, nationality, educational level, career, dominant handedness, family history of mental disorders, course of disease, past medication, mental therapeutics, and the time of the experiment between 10:00~18:00.

#### 2.2.2. Collection of MEG data at spontaneous state

A 306 channels full head MEG from Elekta Neuromag was used for data collection. Ag/AgCl recording electrodes were used for electro-oculography (EOG) and electrocardiography (ECG). Frequency for signal capturing was set at 1000 Hz, with a filter section of 0.1–330 Hz. Head position was monitored by scanning three basal coils on the head. MEG data were discarded if head movement was more than 0.5 cm. Data collection was performed after 12:00 in a magnetically shielded room. MEG signals from a neutral (non-task) state were collected after establishing a local coordinate system. The subject was in neutral state, without any stimulus or any active movements during the scan (the subject was not permitted to fall asleep). Sequential recording consisted of 20-minute periods with eyes opened and 20-minute periods with eyes closed. The status of the subject was under surveillance via camera and microphone.

#### 2.2.3. Collection of SEF MEG data

After the neutral state measurements were taken, the subject was given a SEF test: the subject lay down horizontally, and persistent square-wave pulses at 0.2 ms intervals were applied to the left posterior tibial nerve percutaneously via regular felt-tip bipolar electrodes. Electrodes were fixed to the subject's ankle. We employed the twitching of the great toe as a parameter to indicate that the electrodes were in their proper position. The stimulus current (about 12~16 mA) was applied over 1000 ms (900~1100 ms) intervals, in a pseudorandom manner). A stimulus sequence contained 150 stimuli. Frequency for signal capturing was set at 1000 Hz, with low-pass filtering at 0.1 Hz, high-pass filtering at 60 Hz. DC-offset is removed based on the whole trial after filtering. For the SEF under single-stimulus, the average window is from −100 ms to 300 ms, relative to the trigger S (0 ms). At least 100 trials without artifact defects were used for averaging. Paired-stimuli were applied to the subject after a 3 min-long rest. The interval within paired-stimuli S1~S2 was 240 ms, and the interval between paired-stimulus was 1000 ms. The stimulus current was set somewhere between 12~16 mA, and the stimulant sequence consisted of 150 paired-stimuli. Frequency for signal capturing was set at 1000 Hz, with low-pass filtering at 0.1 Hz, high-pass filtering at 60 Hz, DC-offset is removed based on the whole trial after filtering, and the average window is from −300 ms to 1000 ms, relative to stimulus S1 (0 ms). At least 100 trails without artifact defects were used for averaging. All subjects underwent head MRI examination after collecting MEG data.

### 2.3. Data processing

#### 2.3.1. Preparation of data

Pre-processing of MEG data was done primarily via the software Elekta Max-filter. Outside interferences (ie, metal items and environmental noises) were excluded via the spatial–temporal MaxFilter. Then the data were processed using an independent component analysis (ICA) and threshold detection in the MEG Processor software, to exclude interference signals from eye movements and heartbeats. Each segmental record was ultimately checked manually, and those with significant artifact defects were rejected.

#### 2.3.2. Filtering

The modified infinite impulse response (IIR) digital filter was used for filtering at a frequency band of 0.1–60 Hz, and the power-line noise of 50 Hz was removed at the same time. The DC offset was removed based on the whole trail after filtering.

#### 2.3.3. Average

Evoked potentials and MEG data from every electrode and every state were put through an averaging superposition. For the single-stimulus trials the total time for averaging was set to start at 100 ms ahead of the stimulus S (0 ms), until 300 ms afterwards. The averaging time for paired-stimuli was set at 300 ms ahead of the stimulus S1 (0 ms), until 1000 ms afterwards. For each subject the average of each state was performed more than 100 times. Fig. 1 (upper part) shows the SEF intensity after a single-stimulus measured by a magnetometer; the lower one is measured by a gradiometer.

#### 2.3.4. Time-frequency analysis

The data underwent a time-frequency analysis via wavelet transformation, whose targeting frequency band was 0.8–60 Hz, as shown in Fig. 2. The color shows the intensity of SEF. Compared to the control group, the RLS patient induced a different rhythm to the same stimulus.

#### 2.3.5. Analysis for source location

A common Beamformer traceable algorithm was used. Specifically, a synthetic aperture magnetometer (SAM) beamformer was used to perform source location analysis for the concerned time point [25–27]. It mainly involved the analysis of source distribution at 40 ms, 60 ms and 100 ms, after stimulus artifact. Next, co-registration with MRI images to generate the magnetic source image was done.

Spatial positioning was performed on 3 mm grids. The source distribution at 40 ms after a single-stimulus is shown in Fig. 3.

### 2.4. Statistical analysis

The software SPSS 19.0 was used for statistical analysis; an independent sample t-test was made to compare the potential differences between RLS patients and control subjects in brain activation.

Peak values within the time points of interest were chosen as the corresponding somatosensory activating intensity values, and the average intensity obtained from 15 subjects was used as the average activating intensity value of the group; the group average value was used for the inter-group comparison. Peak values refer to the maximum magnetic field value measured by sensors at the

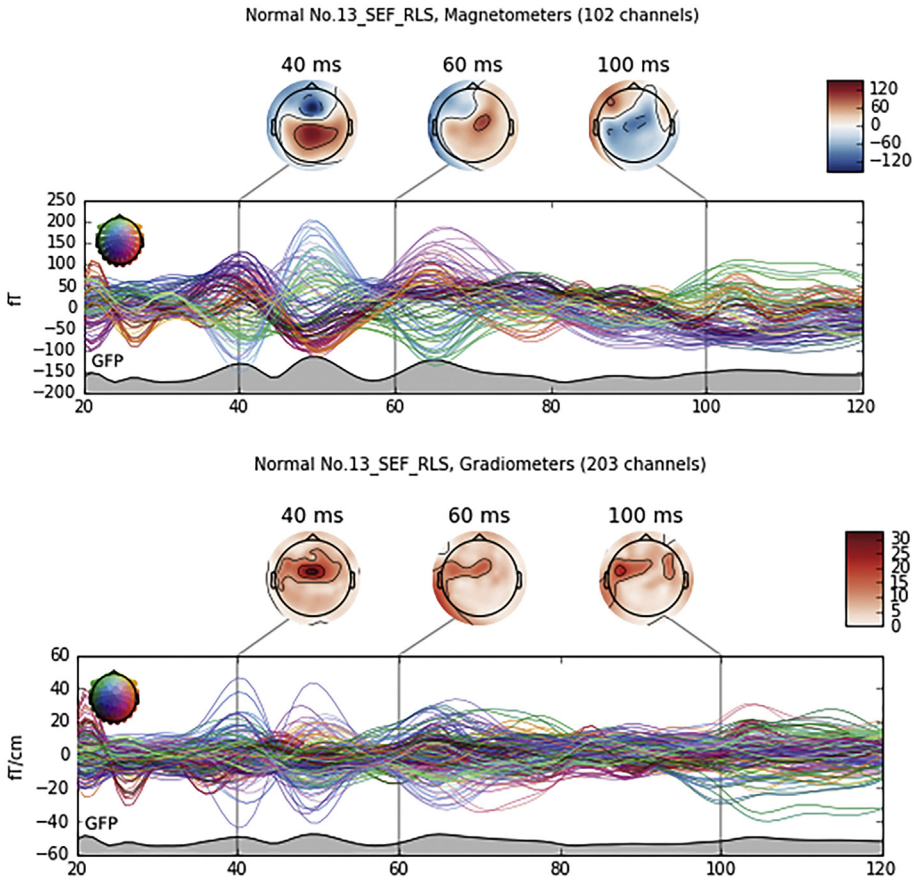


**Fig. 1.** SEF intensity after single-stimulus (No. 13 of control group).

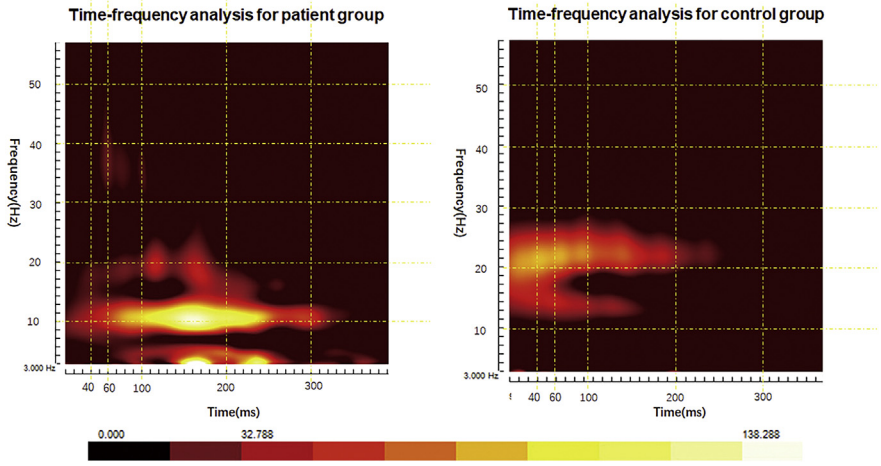


**Fig. 2.** Result of time-frequency analysis after single-stimulus (No. 3 of patient group and No. 10 of control group).

same time point; as stated above, the time points of interest are 40 ms, 60 ms, and 100 ms after S1, and 40 ms, 60 ms, and 100 ms after S2. The interval time between S1 and S2 was 240 ms. Evoked time variation (relative to the time point of interest) <10 ms.

Fig. 4 shows the comparisons among responding magnetic field intensities 40 ms (P40), 60 ms (P60), 100 ms (P100) after stimulus S1, and 280 ms (P40), 320 ms (P60), and 340 ms (P100) after stimulus S2. Data from the experimental group and the control group went through a hypothetical test, with the initial hypothesis H0 being: “there is no significant difference between these two groups”, and the alternative hypothesis H1, “there is a significant difference”, to obtain the values of t-test-related statistics and p-value. Interim calculations and analysis for gating control rates were performed based on the average somatosensory responding intensities of each group; the related p value were also obtained.

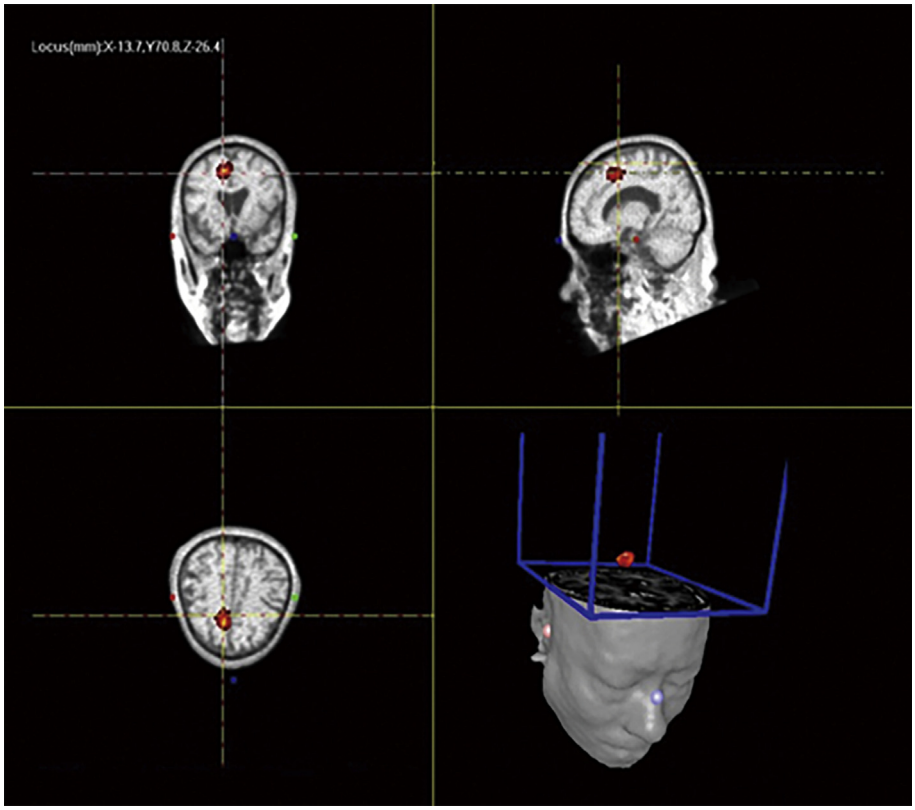


**Fig. 3.** Source location at 40 ms after stimulus (No. 1 of patient group, Blue marker: nasion of nose, Red marker: right ear, Green marker: left ear).

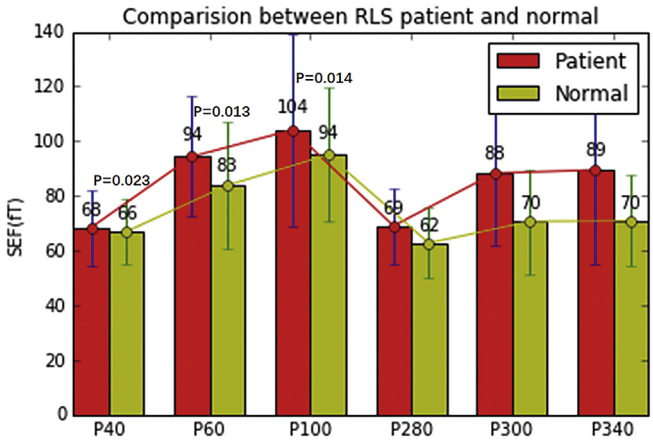


**Fig. 4.** Comparison of average responding magnetic field intensities under paired-stimuli, and p values representing statistical difference after a Student *t*-test.

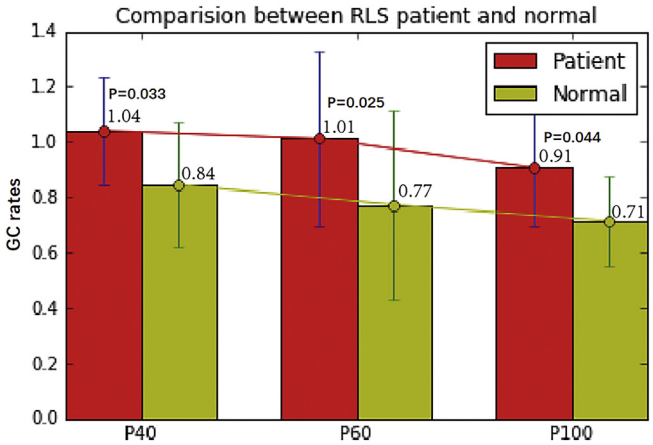


**Fig. 5.** Comparison of average GC rates and p values representing statistical difference after a Student's *t*-test.

Fig. 5 shows the comparison of the average gating control (GC) rate of paired somatosensory stimuli, where the calculation for GC rates is based on the ratio of the magnetic field intensity of the second stimulus (S2) to the magnetic field intensity of the first stimulus (S1). This means that the smaller the value GC rate is, the stronger the GC effects are (and vice versa) (see Figs. 6 and 7).

## 3. Results

As shown in Fig. 3, precise source tracing for a somatosensory stimulus in the lower limbs is achieved via the Beamformer method, and the projecting sites match up with the expected classical anatomic areas. Determination of the single somatosensory stimulus intensities and the GC ratio of paired-stimuli were performed for the experimental group and the control group, respectively; there were significant inter-group differences observed:

1. The evoked magnetic field intensities of the experimental group are all significantly higher than that of the control group, in three sets of comparisons ($P < 0.05$). There are three peaks located at the contour line of SEF (non-polar, distances of the contour line above and below the base line are roughly equal), residing 40 ms, 60 ms and 100 ms from the artifactual defect of the stimulus, respectively, after measuring the intensities of P40,

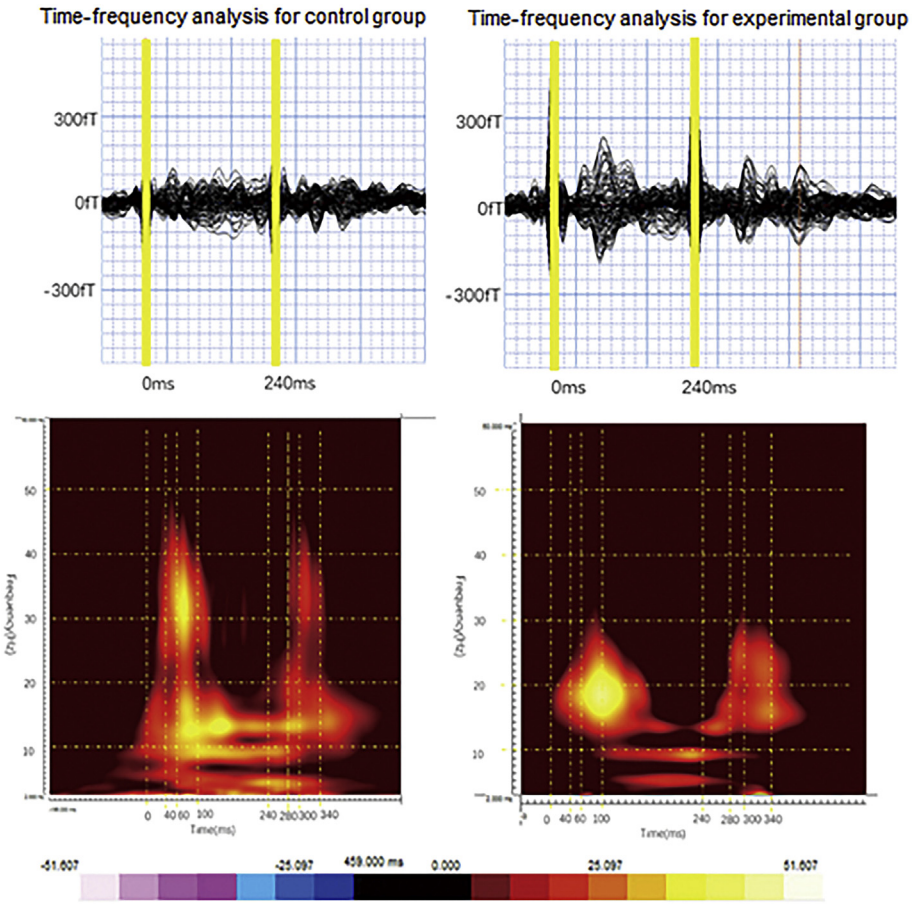


**Fig. 6.** Contrast of response intensity and time frequency of paired-stimuli.

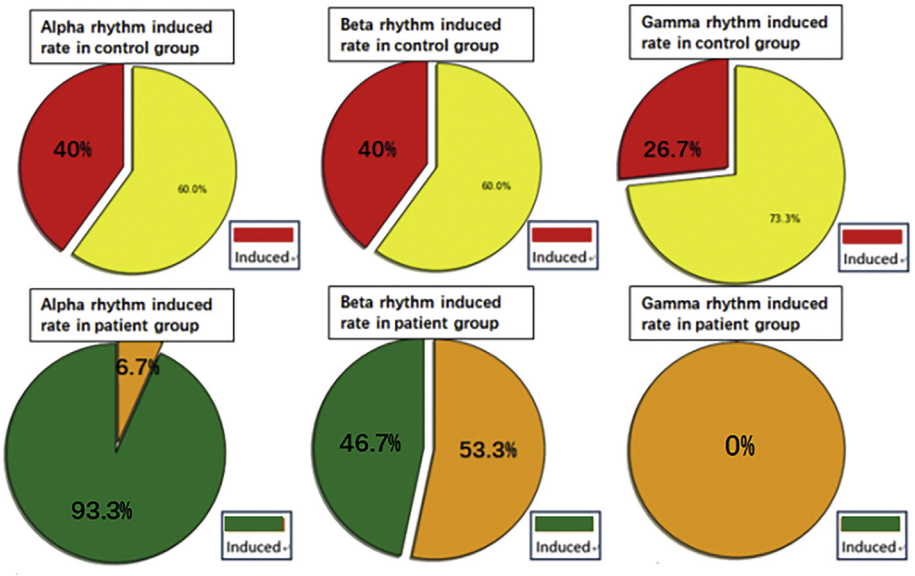


**Fig. 7.** Activation percentages for individual frequency bands.

P60 and P100 (Evoked time variation < 10 ms, P represents the T-test result) and analyzing each of them individually.

2. Gating controls exist in both the experimental and control group after paired-stimuli, but there are also significant differences between the two groups. There are three peaks located at the contour line of SEF residing 40 ms, 60 ms and 100 ms from the artifactual defect of stimulus S1, respectively. Similar to S1, there are also three peaks located at the contour line of SEF residing 280 ms, 300 ms and 340 ms from the artifactual defect of stimulus S1 (the time interval between S1 and S2 is

240 ms). After calculation and statistical analysis of the magnetic field intensity ratio of SEF2/SEF1, (SEF2 is the evoked field intensity of S2, SEF1 is the evoked field intensity of S1) it was found that GCs exist at 40 ms, 60 ms and 100 ms after the stimulus in the control group (SEF2/SEF1 <1 shows the GC exists), but GC only exists at 100 ms in the experimental group. The gating effect is weaker in the experimental group compared to the control group.

3. The evoked magnetic field intensities for each rhythm are shown in Tables 1 and 2. Significant differences between the induced frequency bands of both groups was revealed: the SEF intensity > 60 fT (based on the spontaneous state measurement) is taken as an efficient induced strength; there are somatosensory induced oscillations at three frequency bands ($\alpha$,$\beta$,$\gamma$) in control group, while for the experimental group there are only two frequency bands ($\alpha$ and $\beta$). For $\alpha$ oscillations (8~12 Hz), there are six cases (40% of total cases) in the control group and 14 cases (93.3% of total cases) in experimental group, showing significant differences. For $\beta$ oscillations (13~29 Hz), there are six cases (40% of total cases) in control group and seven cases (46.7% of total cases) in experimental group, showing no significant differences between two groups. For $\gamma$ oscillations (30~60 Hz), there are four cases (26.7% of total cases) in control group and no case with low $\gamma$ oscillation in experimental group, showing significant differences between the groups.

**Table 1**
Rhythm induced intensity (fT) of patient.

| Patient No. | Alpha | Beta | Gamma |
|---|---|---|---|
| 1 | 80 | 13 | 13 |
| 2 | 80 | 10 | 11 |
| 3 | 62 | 10 | 10 |
| 4 | 77 | 20 | 12 |
| 5 | 78 | 80 | 10 |
| 6 | 30 | 5 | 5 |
| 7 | 77 | 77 | 10 |
| 8 | 65 | 80 | 10 |
| 9 | 70 | 77 | 12 |
| 10 | 70 | 60 | 11 |
| 11 | 77 | 70 | 10 |
| 12 | 65 | 20 | 11 |
| 13 | 77 | 30 | 11 |
| 14 | 65 | 70 | 10 |
| 15 | 70 | 70 | 13 |
| Mean | 69 | 46 | 11 |

**Table 2**
Rhythm induced intensity (fT) of controls.

| Controls No. | Alpha | Beta | Gamma |
|---|---|---|---|
| 1 | 75 | 65 | 10 |
| 2 | 35 | 58 | 10 |
| 3 | 30 | 25 | 77 |
| 4 | 20 | 55 | 12 |
| 5 | 40 | 65 | 11 |
| 6 | 10 | 55 | 70 |
| 7 | 70 | 57 | 55 |
| 8 | 40 | 50 | 55 |
| 9 | 30 | 55 | 65 |
| 10 | 70 | 65 | 10 |
| 11 | 50 | 55 | 30 |
| 12 | 75 | 70 | 10 |
| 13 | 77 | 65 | 60 |
| 14 | 70 | 70 | 10 |
| 15 | 55 | 20 | 70 |
| Mean | 50 | 55 | 37 |

## 4. Discussion

### *4.1. SEF intensity in RLS patients' lower limb is significantly higher than that of the control group*

It takes about 40 ms for a somatic sensory input to the lower limbs to be delivered to primitive somatosensory cerebral cortex. With the aid of the Beamformer algorithm-based MEG Processor for temporo-spatial tracing of sources, the source could be precisely projected onto the somatosensory region of the lower limbs. The SEF intensity at 40 ms likely reflects the excitability of primitive somatosensory cerebral cortex. Past studies have shown that the nerve conduction velocity, the amplitude of the H reflex wave, the ratio of H/M and the amplitude of the F wave are all normal in RLS; indicating that excitable input is normal at the level of the peripheral nerves and spinal cord [28–30]. Our experiment shows that SEF intensities in the lower limbs of RLS patients are enhanced compared to that of the control group, which might be evidence that the excitability of primitive somatosensory cerebral cortex is increased.

Furthermore, for the following records at 60 ms and 100 ms, SEF intensities of the RLS group are also higher than those of the control group. The continuous source tracing for somatosensory signals shows that the somatosensory signals at corresponding time points are conducted from the primitive somatosensory area towards the frontal motor area, supplementary motor area, somatosensory region of contralateral parietal lobe and somatosensory area of ipsilateral parietal lobe. The evidence also indicates that the contralateral supplementary motor area, and the ipsilateral parietal area also maintained high excitability during the processing of somatosensory information.

Results of previous studies show that the motor cortical silent period (C-SP) is reported to be shortened and the peripheral silent period (P-SP) is normal in RLS patients, which indicates that the intracortical inhibition is attenuated and excitability of motor cortex is increased in RLS [31]. PSG studies reveal that from the beginning of sleep, the energy levels of each frequency band ($\alpha$,$\beta$,$\sigma$,$\theta$) in apical areas (C3 and C4) are higher than the control group [32], which is in accordance with our results.

RLS is often associated with depression, and depression [33] could cause changes in cortical plasticity. However, depression affects cerebral cortex excitability in a complex pattern [33]. In contrast, RLS patients demonstrate excitability primarily of the somatosensory cerebral cortex. This more specific local excitability is in good agreement with recent research results from Transcranial Magnetic Stimulation (TMS) studies [34,35]. Therefore, it appears that the increase of the excitability for RLS somatosensory cortex should be considered as a trait of the disease itself rather than reflect a general consequence of depression, sleep instability or fragmentation [34,35].

Magnetic resonance spectroscopy (MRS) spectrum analysis revealed increases in the excitable transmitter glutamic acid in the thalamus and as well, abnormal activation of the thalamus [36,37]. Experimental results show sensorimotor cortex is in a state of increased excitability, so it might be related to an increased excitable transmitter in RLS patients.

### *4.2. PPD exists in both groups, but the PPD of the RLS group is significantly weaker compared to that of the control group*

The event-related potential/event-related magnetic field of paired-stimuli is a common measurement for GC. When there is a pair of suitable short stimuli manifested asynchronously, the attenuated nervous processing of the second identical stimulus is called the sensory gating effect, which reflects auto-repression of cortical

function [38]. Furthermore if the wave amplitude of the second stimulus is conserved (not decreased), it indicates a loss of GC.

GC consists of stimulus-driven gating control (bottom up) and mission of interest-driven gating control (top down) [39]. Paired-pulse inhibitory gating control (PPIGC) is stimulus-driven, which is only related to the nature of the stimulus and irrelevant to the mission of interest. Moreover, PPIGC is pre-attention inhibitory processing by the central nervous system. The first stimulus S1 of the paired-stimuli is the guiding stimulus, while the second stimulus S2 is the testing stimulus. Under normal conditions, paired-stimuli code and collect information with bandpass-filtering, and have more intensive influence on stimuli with high prominence, which could increase the signal-noise ratio (SNR), filter redundant sensory influx and avoid information overloading of the central nervous system [40].

In our experiment, a continuous source location is performed for three peaks (ie, peaks 40 ms, 60 ms and 100 ms apart from the artifactual defect), combined with detection by virtual electrodes. The analysis shows that the signal of the first peak at 40 ms is projected to the contralateral first somatosensory area responsible for lower limbs after source location, and the supplementary motor area and central parietal area are activated sequentially. The second peak signal in 60 ms is traced to the contralateral first sensory area close to the cortical area. Finally, the third peak signal in the cortex at 100 ms may correspond to the signal of a virtual electrode in 90 ms, indicating the activation of thalamus, anterior insula and anterior temporal area sequentially (1 cm of conduction distance brought about delay of 8~10 ms).

After determination of the GC of SEF by paired-pulse stimuli in lower limbs, it was found that, in the control group, a relatively strong gating control process had been manifested during the first peak (40 ms) and the second peak (60 ms) (corresponding to primitive somatosensory area after source location). In addition, and GC is further enhanced during the third peak (100 ms) (corresponding to thalamus, anterior insula and anterior temporal lobe). For the RLS group, no GC was shown during the first and the second peaks. Although GC occurred during the third peak at 100 ms, it was weaker compared to that of controls. Collectively, somatosensory GC processed at an early stage (before the occurrence of attention) was lost and the somatosensory GC was attenuated in later stages.

Somatosensory GC exists in normal people and attenuates with aging [41]. It is under the control of multiple neural transmitters, and, especially concerning the forebrain area, charged by dopaminergic nerve nuclei and neurons expressing the D1 receptor in dorsal striatum to take part in GC [42]. Dopaminergic projection reaches the striatum via a direct and an indirect pathway, via an inhibitory effect on the thalamus to decrease glutamatergic projection onto the cortex, and exerting GC [43].

Some studies point out that RLS might belong to the same disease spectrum as attention deficit-hyperactivity disorder (ADHD) and transient tic disorder (TTD). It might also be related to an abnormality in the catecholaminergic transmitter system, since ADHD and TTD also share similar types of sensorimotor gating control disorders [44].

As stated above, evidence indicates that a functional disorder exists in the dopaminergic transmitter system in RLS patients. Since the result of our experiment shows that there are somatosensory gating control defects in RLS patient, one might speculate that it is connected to functional deregulation in the dopaminergic transmitter system. In recent studies, the inhibition of rTMS induced an LTD-like role of RLS patients is weakened, suggesting that RLS has GABA loop damage, and that the fade-away of gate control in RLS may be associated with this mechanism [45,46], Furthermore, our experiment reveals that there are GC defects during the early (before attention) processing for somatosensory information in RLS patients. This also indicates that there might be an abnormal excitation in primitive sensory cortex at first in RLS patients (abnormal sensation at first), and then abnormal consciousness is further developed on this pathological base; which might be of great significance in defining the property of RLS sensory characteristics.

### 4.3. Relation between GC processing and SIO

Continuous repeated somatosensory stimulus may induce somatosensory-related potentials (SRP) with short latent periods (somatosensory related magnetic field, SRF) and also induce somatosensory oscillation having a long latency period (ie, somatosensory-related de-synchronization and synchronization, ERD/ERS). The cortical neuron reactions shown by SRPs originate from altered influx activity, while the alteration of SIO reactions originate from the regional interaction between major neurons and intermediate neurons. Unlike event-related potentials (ERPs), event-induced oscillation (EIO) is a persistent change of signal with locked time, but not locked phase, which reflects the synchronous oscillation of regional neuron clusters and is of high frequency specificity.

Heterotopic neuron networks could show different synchronization patterns via oscillation within different frequency bands. Oscillation and the intrinsic cell membrane properties of neurons are related to the dynamics of synaptic processing, different types of feedback loops and the regulation of collective or regional neural transmitter systems [47].

Research reveals that different rhythm oscillations show a certain specificity as well as overlap. Central somatosensory Beta rhythm is found in areas anterior to the Rolandic fold (at least, its partial components). Somatosensroy Alpha rhythm is mainly generated in areas posterior to the Rolandic fold [48]. While somatosensory Gamma oscillation mainly originates from post-central gyrus, and, less intensively, involves the pre-central gyrus and the anterior parietal lobule [49].

Somatosensory Beta oscillation reflects independently manifested sensory in-flux at an early stage, and regulates reactions in the first somatosensory area and adjacent cortex; while Alpha oscillation joins in the subsequent processing of information via related cortical areas [50,51].

Somatosensory Alpha oscillation might be induced by cortical–cortical interactions and message passing, and it mainly processes GC at later stages. Activity of Alpha oscillation takes part in the inhibitory handling of internal missions of interest via the generation of a suppressive action potential, keeping the cortex idling. Alpha activities are also involved in the handling of noxious pain and $\mu$ $\alpha$ could down-regulate motor cortex via frontal mirror neurons [52,53]. Somatosensory $\alpha$ oscillation also has great significance in integrating and converting “vision” and “hearing” into “action” [54,55]. Moreover, $\alpha$ rhythm is under the regulation of the glutamatergic receptor channel, cerebral development, the 5-HT projection system and circadian rhythm-related genes.

Gamma-oscillation is connected to high-level information processing, which also reflects the integration of repeated afferent cortical excitation in the greater cortical network. Higher frequency of $\gamma$ oscillation is correlated with better somatosensory GC [56,57]. The existence of an inhibitory intermediate neuron network is the product of $\gamma$ oscillation [58].

The result of time-frequency analysis shows significant difference between RLS patients and control subjects in induced rhythms.

First, the occurrence rate of somatosensory $\beta$ oscillation is not significantly different between RLS patients and controls, which implies that the processing of early regional basic somatosensory information is not different. But in RLS patients, somatosensory $\gamma$

oscillation is lost, suggesting lesions in cortical inhibitory neuron network function; the frequency of somatosensory $\alpha$ oscillation activity is 93.3%, suggesting that the attention-related inhibitory gating control mechanism is activated during late stage processing of somatosensory information.

## 5. Conclusion

It is currently believed that the core mechanism of RLS might be a sensorimotor integration disorder. Sensorimotor integration is defined as "the capability of the central nervous system to integrate different sources of stimuli and, at the same time, to transform such inputs into motor actions" [59]. Does sensorimotor integration disorder originate from abnormal peripheral input or defective central nervous system processing? This question is addressed by this experiment. There are sensorimotor integration abnormalities at the level of the cerebral cortex in RLS patients, implying that there is damage to the inhibitory intermediate neuron network as well as increased excitability in primitive sensorimotor cortex. During the process of somatosensory stimulation, early somatosensory GC is attenuated, and attention-driven somatosensory GC is enhanced via $\alpha$ oscillation in later stages, thus motor behaviors are predicted to occur. Therefore, from an electrophysiological point of view, the increased excitability in sensorimotor cortex is an important cause of RLS sensory disturbances. At present, repetitive transcranial magnetic stimulation (rTMS) for RLS has achieved a certain effect [60]. However, to achieve better therapeutic effects, the key is to select the appropriate stimulus target, as well as the appropriate stimulus frequency and pattern. The authors believe that sensory cortex may be one of the potential targets for electrical and magnetic stimulation therapy. Therefore, we can simulate the pathophysiological pattern of RLS patients' processing of somatosensory information to alleviate the clinical symptoms and explore different treatments to induce Alpha oscillations in the cerebrum, thus helping to determine the appropriate frequency of stimulation and the mode of stimulation.

The following limitations are found in this study: (1) First, this study is limited to the MEG electrophysiological perspective to explore the mechanism of RLS sensory disturbance rather than a multidimensional, comprehensive and in-depth study. It could be argued that the electrophysiological phenotype of the central response mechanism of the external electrical stimulation of the lower extremities of the RLS patients is only suggested. (2) The sensory disturbance of RLS patients is endogenous in someone's natural state; as this study uses an exogenous disturbance, it may be difficult to fully reflect the true condition of patients under natural conditions. (3) The experiment is done in a relatively broad range of time (10:00 a.m.–18:00 p.m.); a "temporal bias" might affect the results [61]. Finally, (4) The sensory disorders in RLS patients are state dependent and circadian rhythms dependent; however, this is a cross-sectional study which makes it difficult to fully display central processing in different states dynamically.

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## Conflicts of interest

The ICMJE Uniform Disclosure Form for Potential Conflicts of Interest associated with this article can be viewed by clicking on the following link: https://doi.org/10.1016/j.sleep.2018.07.026.

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