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rTMS Improves Cognitive Function and Brain Network Connectivity in Patients With Alzheimer's Disease*

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Abstract Objective Repetitive transcranial magnetic stimulation (rTMS) has demonstrated efficacy in enhancing neurocognitive performance in Alzheimer's disease (AD), but the neurobiological mechanisms linking synaptic pathology, neural oscillatory dynamics, and brain network reorganization remain unclear. This investigation seeks to systematically evaluate the therapeutic potential of rTMS as a non-invasive neuromodulatory intervention through a multimodal framework integrating clinical assessments, molecular profiling, and neurophysiological monitoring.Methods In this prospective double-blind trial, 12 AD patients underwent a 14-day protocol of 20 Hz rTMS, with comprehensive multimodal assessments performed pre- and post-intervention. Cognitive functioning was quantified using the mini-mental state examination (MMSE) and Montreal cognitive assessment (MoCA), while daily living capacities and neuropsychiatric profiles were respectively evaluated through the activities of daily living (ADL) scale and combined neuropsychiatric inventory (NPI) -Hamilton depression rating scale (HAMD). Peripheral blood biomarkers, specifically Aβ1-40 and phosphorylated tau (p-tau181), were analyzed to investigate the effects of TMS on molecular metabolism. Spectral power analysis was employed to investigate TMS-induced modulations of neural rhythms in AD patients, while brain network analyses incorporating topological properties were conducted to examine stimulus-driven network reorganization. Furthermore, systematic assessment of correlations between cognitive scale scores, blood biomarkers, and network characteristics was performed to elucidate cross-modal therapeutic associations.Results Clinically, MMSE and MoCA scores improved significantly (P<0.05). Biomarker showed that Aβ1-40 level increased (P<0.05), contrasting with p-tau181 reduction (P<0.05). Moreover, the levels of Aβ1-40 were positively correlated with MMSE and MOCA scores. Significant post-intervention alterations were observed in oscillatory power, with marked reductions in delta (P<0.05) and theta bands (P<0.05), contrasted by gamma band power elevation (P<0.05). No significant changes were observed in alpha and beta band EEG powers (P>0.05). Network analysis revealed frequency-specific reorganization: clustering coefficients were significantly enhanced in delta, theta, and alpha bands (P<0.05), while global efficiency improvement was exclusively detected in the delta band (P<0.05). The alpha band demonstrated concurrent increases in

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average nodal degree (P<0.05) and characteristic path length reduction (P<0.05). Further research findings indicate that the changes in the clinical scale HAMD scores before and after rTMS stimulation are negatively correlated with the changes in the blood biomarkers Aβ1-40 and p-tau181. Additionally, the changes in the clinical scales MMSE and MoCA scores are negatively correlated with the changes in the node degree of the alpha frequency band and negatively correlated with the clustering coefficient of the delta frequency band. However, the changes in MMSE scores are positively correlated with the changes in global efficiency of both the delta and alpha frequency bands. Conclusion 20 Hz rTMS targeting dorsolateral prefrontal cortex (DLPFC) significantly improves cognitive function and enhances the metabolic clearance of β-amyloid and tau proteins in AD patients. This neurotherapeutic effect is mechanistically associated with rTMS-mediated frequency-selective neuromodulation, which enhances the connectivity of oscillatory networks through improved neuronal synchronization and optimized topological organization of functional brain networks. These findings not only support the efficacy of rTMS as an adjunctive therapy for AD but also underscore the importance of employing multiple assessment methods—including clinical scales, blood biomarkers, and EEG-in understanding and monitoring the progression of AD. This research provides a significant theoretical foundation and empirical evidence for further exploration of rTMS applications in AD treatment.

Key words transcranial magnetic stimulation, Alzheimer's disease, power spectral density, electroencephalogram, brain functional network

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Alzheimer's disease (AD) is a neurodegenerative condition marked by cognitive decline, abnormal mental behavior, and impaired daily living abilities, making it the most common form of dementia[1-2]. The substantial incidence and disability rate of this disease impose a significant burden on patients' families and society[3-4]. Existing pharmacotherapies for AD serve only to ameliorate symptoms without arresting the underlying disease progression[5]. Consequently, the pursuit of secure and efficacious alternative or complementary therapeutic strategies is of paramount importance for the management of AD.

Repetitive transcranial magnetic stimulation (rTMS) is a non-invasive, safe, and painless mature brain stimulation technology [6-7]. rTMS uses a pulsed magnetic field to penetrate the skull and affect the cerebral cortex, altering the membrane potential of cortical nerve cells and generating induced currents. This affects brain metabolism and neural activity, leading to various physiological and biochemical reactions [8-9]. Li *et al*. [10] utilized 20 Hz rTMS to assess the motor evoked potentials of synaptic plasticity in AD patient. Their results indicated that rTMS facilitated long-term potentiation (LTP) and was linked to improvements in cognitive behavior in AD patients. Jia *et al*. [11] demonstrated that high frequency rTMS enhances cognitive function, particularly memory, in the left parietal cortex of patients with mild to moderate AD. Consequently, 20 Hz transcranial magnetic stimulation is employed in the treatment of AD patients. Currently, TMS is thought to regulate brain function by influencing neurorhythms through an entrainment effect. However, whether TMS impacts cerebral neurorhythm activity and brain network reorganization to improve cognitive function in patients requires further investigation.

The primary mechanism for information integration and processing in the brain involves the synchronous oscillation of neural networks, which is essential for mediating inter-regional communication. Due to its real-time, easy-to-use, non-invasive, and low-cost nature, electroencephalogram (EEG) has been used in numerous neurological studies to analyze the relationship between brain wave activity and cognitive processes[12-13]. EEG activity may be a more sensitive indicator of the effects of TMS on brain function compared to some behavioral assessments[14].

This technique captures neural oscillatory signals across the brain, which can be decomposed into different frequency bands of activity (delta, theta, alpha, beta, and gamma bands)[15], Empirical research has established correlations between specific EEG frequency bands and discrete cognitive functions. Reduced rapid (alpha, beta, and gamma) and increased slower rhythms (delta and theta) are general resting-state EEG metrics in patients with AD[16]. Bai *et al*.[17] showed that the energy in delta and gamma bands of rats in the rTMS group was enhanced, reflecting the regulatory effect of high frequency rTMS on the cerebral cortex, thus affecting neural information activities related to cognitive function and enhancing working memory (WM) ability. Similarly, Guan *et al.*[18] observed that gamma oscillations helped to improve cognitive and spatial memory deficits in AD model mice. Building on these findings, our subsequent investigation will focus on assessing the potential of TMS to ameliorate the neurorhythmic abnormalities in AD patients and on exploring the nexus between rhythmic alterations and advancements in cognitive function.

The human brain comprises multiple intricately connected regions, with each network supporting different cognitive functions. The control network includes multiple medial prefrontal cortex, inferior frontal, and inferior parietal areas, with the dorsolateral prefrontal cortex (DLPFC) serving as its nucleus[19]. These regions are primarily associated with cognition and emotion. The control network is involved in high-level cognitive tasks and plays a crucial role in adaptive cognitive control. Consequently, rTMS usually targets the DLPFC in AD patients. Jones *et al*.[20] found reduced connectivity in the default mode network of AD patients through the analysis of resting-state functional magnetic resonance imaging (fMRI) data. Zhou *et al*.[21] noted an increase in functional connectivity (FC) within the frontoparietal network and a decrease in FC in the hippocampus and several other brain regions in individuals with AD. Lv *et al*.[22] categorized 31 cases of preclinical AD patients into a low-connectivity group (LCG) and a high-connectivity group (HCG). The LCG exhibited increased DMN connectivity and significantly positive memory improvement, while the HCG demonstrated a contrasting decrease in connectivity and maintained or slightly improved their cognitive function following neuro-navigation rTMS treatment.

Given that the incidence of AD is directly related to age, it is imperative to improve the detection rate of AD and monitor the progression of AD. Several studies have confirmed the existence of biomarkers in the blood that can reflect the pathological process of AD. Further study on the changes of blood markers and disease course can provide a basis for early diagnosis and treatment of AD patients.

In this investigation, we collected clinical scales, blood markers and resting-state EEG data from 12 individuals with AD, both prior to and following rTMS. Initially, we calculated the EEG power spectral density before and after stimulation and analyzed the delta, theta, alpha, beta, and gamma power bands. We constructed the brain functional network (BFN) of different frequency bands based on phase-locked values and analyzed network parameters, including degree, clustering coefficient, characteristic path length, and global efficiency. This study partially substantiates the therapeutic efficacy of TMS in AD and offers novel perspectives on the dynamics of cognitive function and brain network connectivity in this patient population.

1 Materials and methods

1.1 Subjects

This study enrolled twelve patients diagnosed with AD at Tianjin Huanhu Hospital, who underwent high-frequency TMS (20 Hz). All participants have signed written consent, allowing their medical information and biological samples to be used for research purposes. This study has been approved by the Biomedical Ethics Committee of Hebei University of Technology (Approval No: HEBUThMEC2024009) and adheres to the guidelines of the Declaration of Helsinki of 1975. Inclusion criteria were as follows: (1) fulfillment of the hospital's diagnostic criteria for AD; (2) patients with mini-mental state examination (MMSE) score≤26, Montreal cognitive assessment (MOCA) score<26, activities of daily living (ADL) score>26, neuropsychiatric inventory (NPI) score>0, and Hamilton depression rating scale (HAMD)≥8; (3) all participants signed informed consent. Exclusion criteria included: (1) patients with stroke, schizophrenia, affective disorders, emergency-related conditions, drug or alcohol-induced mental disorders,

epilepsy, head trauma, or serious physical illnesses; (2) patients with contraindications to transcranial magnetic stimulation, such as metal foreign bodies, cardiac pacemakers, cochlear implants, or elevated intracranial pressure; (3) patients who are inability to cooperate with rTMS therapy. The high-frequency stimulation group comprised five males and seven females, aged 55 – 82 years (mean age: (69.17±7.25) years), disease duration: 1 – 10 years, (mean duration: (4.83±1.75) years). Education levels varied: with 6 patients having primary school education or less, 2 with middle or high school education, and 4 with junior college education or higher.

1.2 Equipment and parameters

Subjects sat relaxed in a chair and wore a positioning cap. The left DLPFC was selected as the stimulation site and marked on the cap to ensure the stimulator coil remained correctly positioned throughout the experiment. The transcranial magnetic stimulator (Magstim, UK, model: Rapid^2) coil was placed tangent to the marked position on the patient's skull, used 5 times a week for 20 min per session over 3-week course, totaling 14 treatments. Head movements and patient state were visually monitored during treatment. In the observation group, each patient's motor evoked potential threshold was set at 90%, with a stimulation frequency of 20 Hz, a pulse duration of 2 s, and intervals of 28 s. Each session comprised 40 stimulation trains, culminating in a daily treatment duration of 20 min.

1.3 Observation indicators

In this study, cognitive function was comprehensively assessed in subjects before and after TMS utilizing the AD Assessment Scale, which includes the MMSE and the MOCA. The MOCA, compared to the MMSE, has more complex questions and is more suitable for early-stage patients. The NPI was employed to assess alterations in mental status, while the ADL questionnaire assessed daily living skills. Additionally, HAMD was applied to measure the intensity of depressive and anxiety symptoms. For the collection of blood samples, participants were advised to abstain from food intake for at least two hours prior to the procedure. Venipuncture was performed to obtain blood samples, which were then placed into heparinized tubes. The samples were centrifuged at 2 000*g* for 10 min at a temperature of 4° C within 2 h of collection. The plasma supernatant should be divided into aliquots and stored at – 80° C for subsequent analysis. Sioma technology is used to quantify β -amyloid protein 1-42 (Aβ1-42) and phosphorylated tau 181 (p-tau181) in the plasma samples.

1.4 EEG Data Acquisition and Preprocessing

Resting EEG signals were collected before and after rTMS. The EEG data were captured and archived utilizing a 64-channel wireless EEG acquisition system (NeuSen W, Brikon Technology Inc., China). The system was set to a default sampling rate of 1 000 Hz, with Cz as the reference electrode. Prior to the experiment, scalp impedance was required to be below 500 Ω.

Subjects were instructed to maintain a state of relaxation and immobility throughout the experimental procedure. The experimental environment was low light, quiet, and free of electromagnetic interference. Pre-processing mainly included filtering (passband frequency 0.5 – 45 Hz), re-referencing, and independent component analysis (ICA). Behavioral scale assessment, biomarker blood collection, EEG data collection process. See Figure 1a, behavioral scales and biomarkers for blood statistical analysis, EEG power, and brain functional connectivity analysis, as shown in Figure 1b.

1.5 Methods

(1) Power spectral density: the welch method based on fast Fourier transform algorithm was used to calculate the power spectral density of each channel. The window length is 2 s and the overlap rate is 50%. Finally, the power of five bands is calculated: delta (0.5 – 4 Hz), theta (4 – 8 Hz), alpha (8 – 13 Hz), beta (13 – 30 Hz), and gamma (30 – 45 Hz).

(2) Phase-locked value: the phase-locked value (PLV) indicates the instantaneous phase difference between two signals. When constructing a brain network, it can well reflect the phase relationship between EEG signals collected from different channels. Typically, when different brain regions cooperate to accomplish a task, a higher phase-locked value between them indicates greater participation and efficiency of the corresponding brain regions in processing the task[23]. which is defined as follows:

$$PLV = \frac{1}{N}\left|\sum_{n=1}^{N}\exp\left(j\theta(t,n)\right)\right| \quad (1)$$

N represents the number of trials, and $\theta\ (t,\ n)$ represents the instantaneous phase difference between

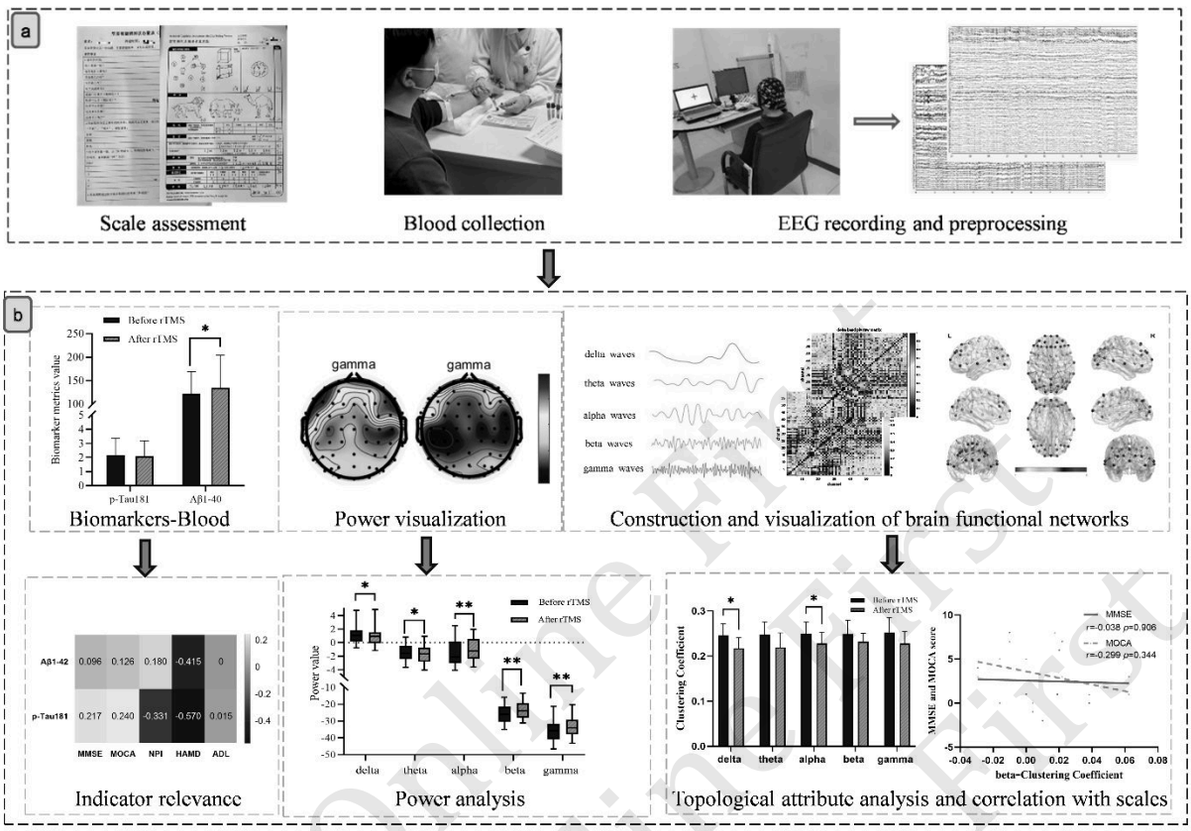

Fig. 1　Schematic diagram of data analysis framework

（a） Data acquisition process： behavioral scale assessment， biomarker blood collection， EEG data collection. （b） Data results analysis. behavioral scale and biomarker blood statistical analysis， EEG power and brain functional connectivity analysis.

the same trail with different leads.

(3) Degree is the most fundamental topological attribute of a network, indicating the importance of a node. A higher degree value signifies more connected edges, and thus, a more important node[24]. The degree of a node i is expressed as:

$$k_i = \sum_{j=1}^{N} a_{ij} \tag{2}$$

The average degree is the mean value of the degrees of all nodes in a complex network, used to analyze the global characteristics of the complex network. The definition of the average degree can be expressed as:

$$K = \frac{1}{N}\sum_{i=j}^{N} k_i \tag{3}$$

a_{ij} represents the connection between nodes i and j; N represents the number of nodes.

(4) Clustering coefficient: The clustering coefficient measures the extent of clustering among nodes in the network. The greater the clustering coefficient, the closer the local connectivity of the nodes[25]. It is expressed as:

$$C_i = \frac{2E_i}{K_i(K_i - 1)} \tag{4}$$

E_i represents the number of edges between all neigh-boring nodes of node i; K_i represents the number of neighbor nodes of node i.

(5) Characteristic path length: The characteristic path length is the average of the shortest path lengths between all pairs of nodes in the network. The smaller the characteristic path length value, the faster the signal exchange rate[26]. It is expressed as:

$$P = \frac{1}{N(N-1)} \sum_{i,j \in N, i \neq j} l_{ij} \tag{5}$$

N represents the number of nodes; l_{ij} represents the shortest path connecting nodes i and j.

(6) Global efficiency: global efficiency quantifies a network's overall capability to transmit and process information. The efficiency of information exchange is directly proportional to the global efficiency value[27]. It is expressed as:

$$L = \frac{1}{N(N-1)} \sum_{i,j \in N, i \neq j} \frac{1}{l_{ij}} \quad (6)$$

N represents the number of nodes; l_{ij}represents the shortest path connecting nodes i and j.

(7) Statistical analysis: the 59-channel average power value of the BFN of 12 subjects, the 59-channel degree value of the average BFN, the clustering coefficient value, the feature path length value, the global efficiency value and the small-world attribute value were taken as continuous variables. The difference of power values and BFN characteristic parameters before and after rTMS was analyzed by paired sample t test. The statistical significance was set at P<0.05. Partial correlation analysis was used to assess the relationship between network topology attributes and MMSE and MOCA scores in AD patients.

2 Results

2.1 Scale evaluation

The MMSE and MOCA scores of patients with AD patients demonstrated a significant increase following stimulation compared to baseline, with this enhancement being statistically significant. These results indicate that TMS may enhance the cognitive abilities of individuals with AD. Additionally, the scores related to activities of ADL, HAMD, and NPI in AD patients exhibited a decrease after stimulation, but without statistically significant. This implies that TMS might modestly ameliorate mental and behavioral symptoms of AD patients to some extent (Figure 2a). In addition, in AD patients, the level of the blood biomarker p-tau181 decreased after stimulation, and a statistically significant increase was observed in the level of Aβ1-40. This suggests that rTMS induces more pronounced alterations in p-tau181 levels (Figure 2b). Moreover, the levels of Aβ 1-40 were positively correlated with MMSE and MOCA scores, and negatively correlated with NPI and HMAD scores (Figure 2c).

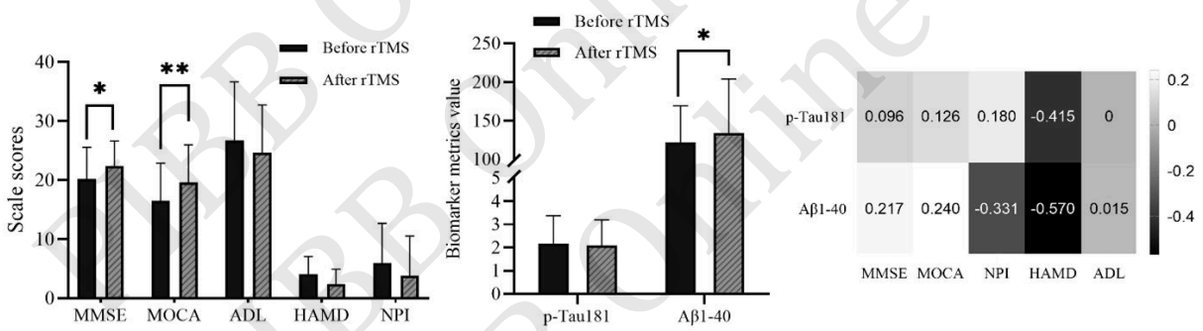

Fig. 2 A comprehensive analysis of the effects of rTMS on cognitive function and biomarkers in patients with AD

（a） （b） （c）（a） Comparison of cognitive function and psych behavioral symptom correlation scale scores before and after stimulation. （b） Comparison of biomarker markers before and after stimulation. （c） Correlation analysis between changes in cognitive scales and changes in biomarker indicators before and after stimulation. ∗P<0.05， ∗∗P<0.01.

2.2 Power spectral density

The average values of delta, theta, alpha, beta, and gamma power for each channel were calculated and after rTMS, and the brain topographic map was depicted (Figure 3). Then, the changes in the power spectrum of whole brain regions before and after rTMS were analyzed. Specifically, Figure 4a shows the average power calculations for all frequency bands in the whole brain region. The power changes of alpha, beta and gamma bands before and after rTMS intervention were further investigated in detail, as shown in Figure 4b. The study found that after receiving rTMS treatment, the average power of whole brain regions in alpha, beta, and gamma bands increased significantly (P<0.05). On the contrary, compared with the pre-treatment period, the average power of the delta and theta bands showed a significant downward trend (P<0.05).

2.3 Brain functional network

The BFN connectivity before and after rTMS

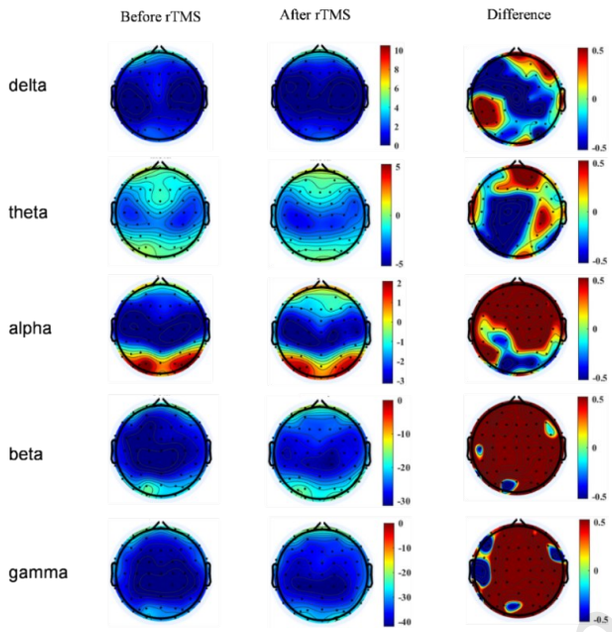

Fig. 3　Analyze the average power imaging of different frequency bands before and after rTMS stimulation，as well as the changes in average power

stimulation is depicted in Figure 5. BFN connectivity in alpha, beta, and gamma bands predominantly clusters in the frontal, temporal, and occipital lobes, whereas in theta and alpha bands, it primarily centers on the frontal and occipital lobes. Following rTMS, an increase in delta band connectivity was noted in the frontal, temporal, and occipital lobes, and theta, alpha, beta, and gamma band connectivity between frontal and left occipital lobes increased. In contrast, the connectivity between the temporal and parietal lobes remained stable across all frequency bands. Meanwhile, most of the functional connectivity centers in each frequency band were concentrated within the frontal and occipital lobes. Subsequently, a comprehensive analysis of characteristic network parameters was conducted, including degree, clustering coefficient, characteristic path length, global efficiency, and small-world attributes.

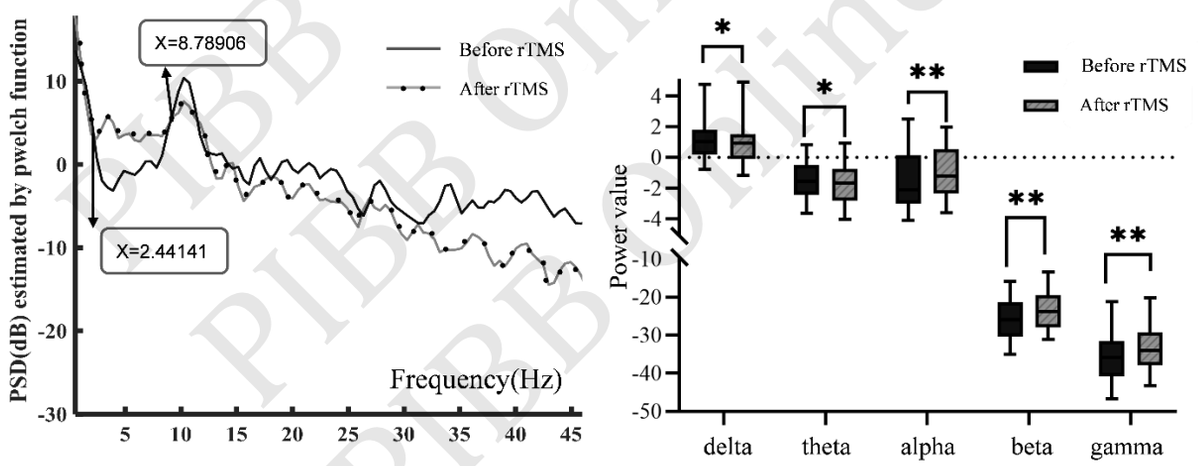

Fig. 4　Research on the effects of rTMS on the power of AD patients

（a） Changes in power spectrum before and after rTMS stimulation. （b） Average power of whole brain area before and after rTMS stimulation. ∗P <0.05， ∗∗P<0.01.

2.4　Average degree

Figure 6a depicts the average degree of BFN across various frequency bands before and after rTMS. The findings reveal distinct alterations in BFN connectivity following rTMS. Post-stimulation, alpha band BFN activity showed a significant increase in average degree (P<0.05), with non-significant elevations observed in delta, theta, beta, and gamma bands, suggesting potential global connectivity enhancements in alpha bands. At the same time, it was found that the change in the average degrees of the alpha was positively correlated with the changes in MMSE and MOCA (t= － 1.39, t= － 1.95; Figure 6b). Notably, average degree change value of alpha band

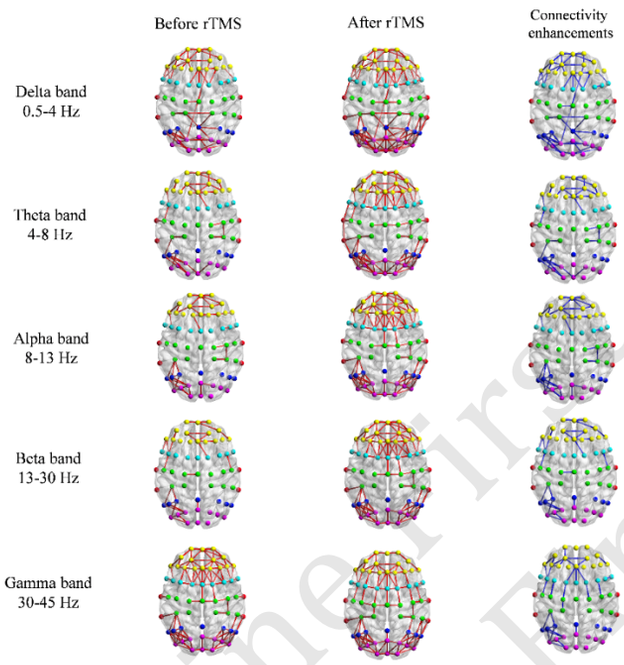

Fig. 5　Comparison of BFN connectivity before and after rTMS. The blue line represents enhanced connectivity after stimulation

was correlated with MMSE (r= − 0.737, P=0.006) and MoCA scores (r= − 0.681, P=0.015).

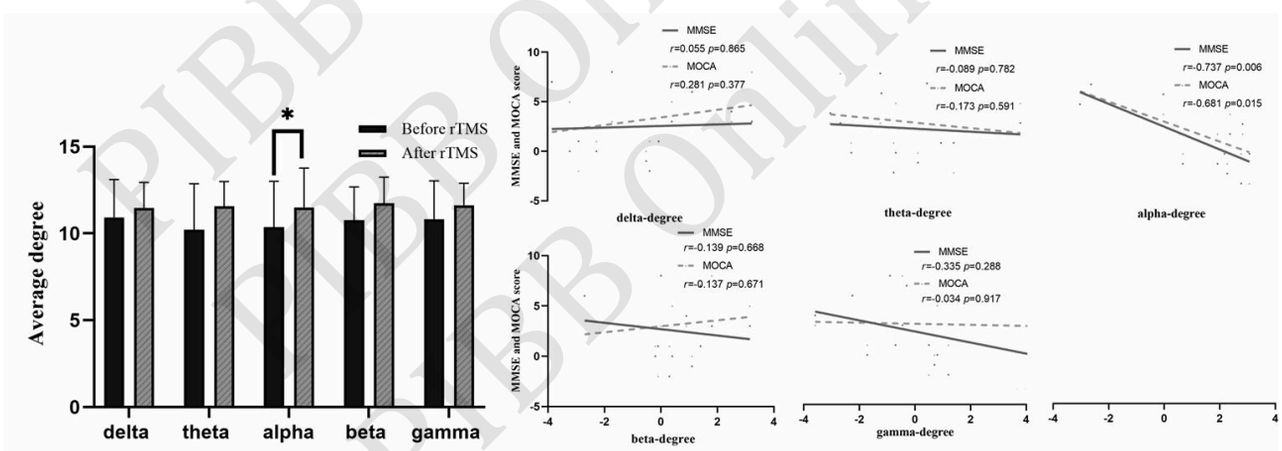

Fig. 6　A study on the effects of rTMS stimulation on brain functional networks in patients with AD

（a） Average degree of BFNs. ∗represents a significant difference. *P< 0.05. （b） The correlation between the change in the average degree and the change in MMSE and MOCA.

2.5　Clustering coefficient

The clustering coefficients of delta, theta, alpha, beta, and gamma band BFNs before and after rTMS were shown in Figure 7a. A significant enhancement in the clustering coefficients of delta and alpha bands BFN was observed after rTMS (P<0.05), while a decrease was observed in the beta, and gamma bands without significant difference (P>0.05). These results indicate enhanced local connectivity in the delta and alpha bands BFN following rTMS. At the same time, it was found that the change in the clustering coefficients of the alpha and beta band were

negatively correlated with the changes in MMSE (t= − 2.77, t= − 2.84). the theta and alpha band were negatively correlated with the changes in MOCA (t= − 3.01, t= − 3.54). As shown in Figure 7b, the clustering coefficients in the delta and alpha bands were positively significantly correlated with the MMSE scores (r= − 0.721, r= − 0.679, P<0.05). The clustering coefficients in delta, theta and alpha bands were negatively significantly correlated with the MOCA scores (r= − 0.612, r= − 0.255, r= − 0.592, P <0.05).

2.6 Characteristic path length

The characteristic path lengths of the delta, theta, alpha, beta, and gamma band BFNs were calculated for all subjects before and after rTMS as shown in Figure 8a. The characteristic path lengths of the alpha band BFN significantly reduced (P<0.05), while a decrease was observed in the delta, theta, beta, and gamma bands without significant difference (P>0.05). At the same time, it was found that the change in the characteristic path lengths of the alpha band before and after stimulation was significantly correlated with the changes in MMSE (t= − 2.58). As shown in Figure 8b. The characteristic path lengths in the alpha band was negatively significantly correlated with the MMSE scores (r= − 0.624, P<0.05).

2.7 Global efficiency

The global efficiency of BFNs delta, theta, alpha, beta, and gamma bands was calculated for all subjects before and after rTMS. As shown in Figure 9a. The global efficiency values of the delta band BFN significantly increased after rTMS (P<0.05), whereas those of theta, alpha, beta, and gamma band BFNs showed no significant change (P>0.05). These

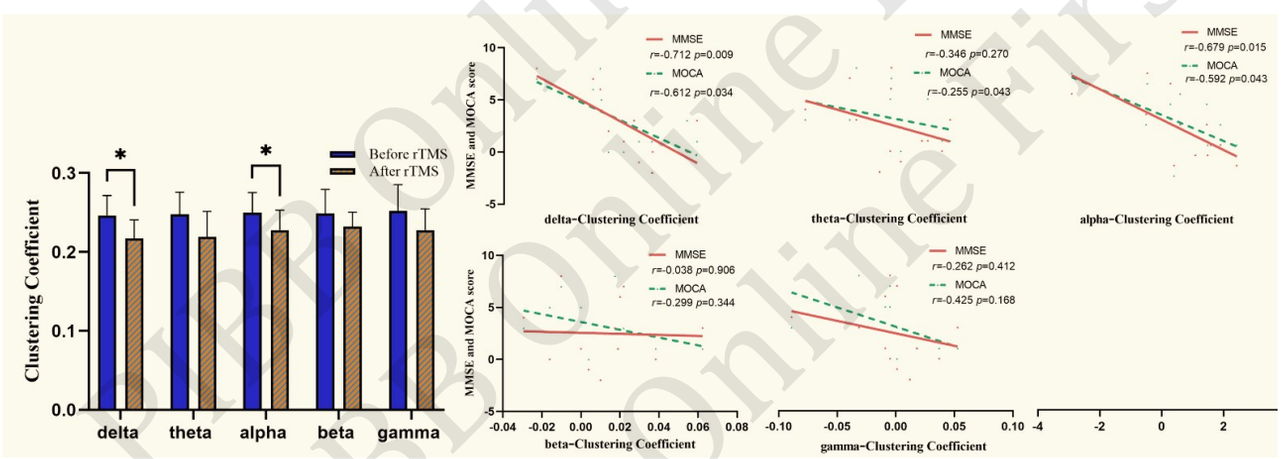

Fig. 7　A study on the effects of rTMS stimulation on brain functional networks in patients with AD

（a） Average clustering coefficient of BFNs. ∗P<0.05. （b） The correlation between the change in clustering coefficient and the change in MMSE and MOCA.

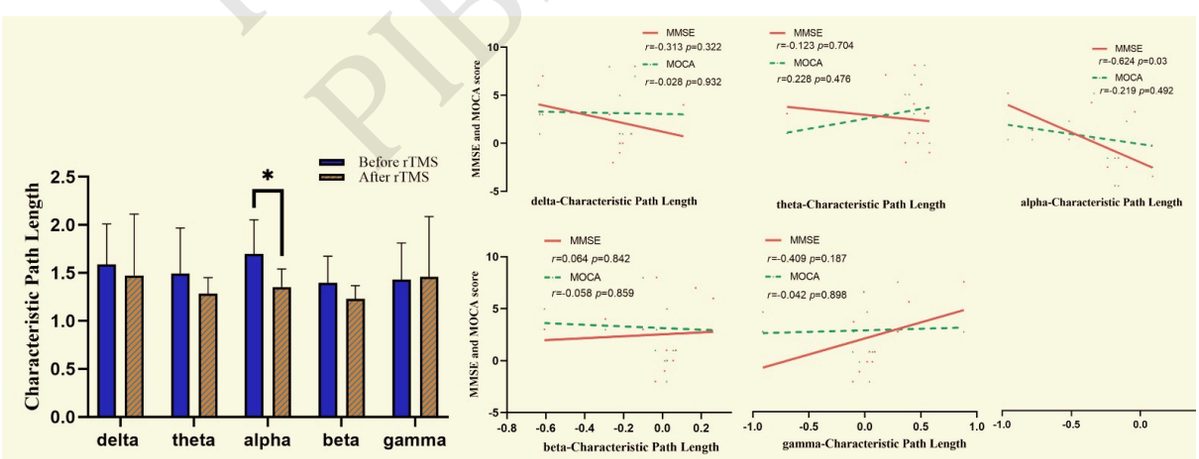

Fig. 8　A study on the effects of rTMS stimulation on brain functional networks in patients with AD

（a） Characteristic path length of BFNs. ∗P<0.05. （b） The correlation between the change in characteristic path length and the change in MMSE and MOCA.

findings imply an improvement in the efficiency of information transfer within the delta band BFN after rTMS. At the same time, it was found that the change in the global efficiency of delta and alpha bands before and after stimulation was significantly correlated with the changes in MMSE (t= – 2.65, t= 2.69). Figure 9b illustrates a significant negative correlation between the global efficiency of the delta band and MMSE scores (r= – 0.673, P<0.05), whereas the alpha band's global efficiency exhibited a significant positive correlation with MMSE scores (r= 0.608, P<0.05).

3 Discussion

Patients with AD underwent 20 Hz TMS conducted in this paper. The study focused on the effects of TMS on congnitive function and brain connectivity. Following the intervention, improvements were observed in patients' cognitive scales, biomarker blood indicators, brain electrical power, and brain network connectivity. Bagherzadeh *et al*. [28] stimulated the left DLPFC of healthy individuals using a high-frequency repetitive TMS protocol and assessed their performance on a range of cognitive tasks. It was found that TMS treatment enhanced working memory performance in both language digit span and visual-spatial tasks. One meta-analysis concluded that rTMS treatment improved overall cognitive function immediately, and that more than 20 sessions of high rTMS treatment in the left DLPFC region yielded the best treatment results, and that cognitive improvement could last for 1 month[29]. The above results are consistent with the conclusions of this study, rTMS has a positive effect on cognitive function in AD patients, specifically, MMSE and MOCA total scores are significantly increased, while ADL, HAMD, and NPI scores are reduced. Blood tests assess the impact of treatment on disease pathology and levels, complementing clinical outcomes and imaging data[30]. At the same time, a recent study by Wagemann *et al.* [31] highlighted the potential of blood tests to reflect changes in brain pathology and treatment response in AD, blood biomarkers Aβ1-42 and p-tau181 both decreased following rTMS, this finding suggests that rTMS positively impacted cognitive function in AD patients. In conclusion, rTMS appears to exert a beneficial influence on cognitive function in AD patients, as indicated by improvements across various cognitive assessment scales and biomarker indicators.

Neuronal oscillations, encompassing the delta, theta, alpha, beta, and gamma bands, are integral to effective neural communication during cognitive processing[32]. Ahmed *et al.* [33] reported that EEG signals from AD patients exhibit abnormal patterns, in which there are more low-frequency components than normal people. In a recent study utilizing a mouse model, early AD was also found to be associated with reduced synchronization of hippocampal gamma-theta oscillations but could be largely restored after 14 d of rTMS[34-35]. In this investigation, we evaluated the power within the delta, theta, alpha, beta, and gamma bands to investigate the therapeutic effects of rTMS for AD patients. Our findings indicated a significant reduction in power within the delta and theta bands across the entire brain, after rTMS, coupled with a significant increase in power within the alpha, beta, and gamma bands. Consequently, this study suggests

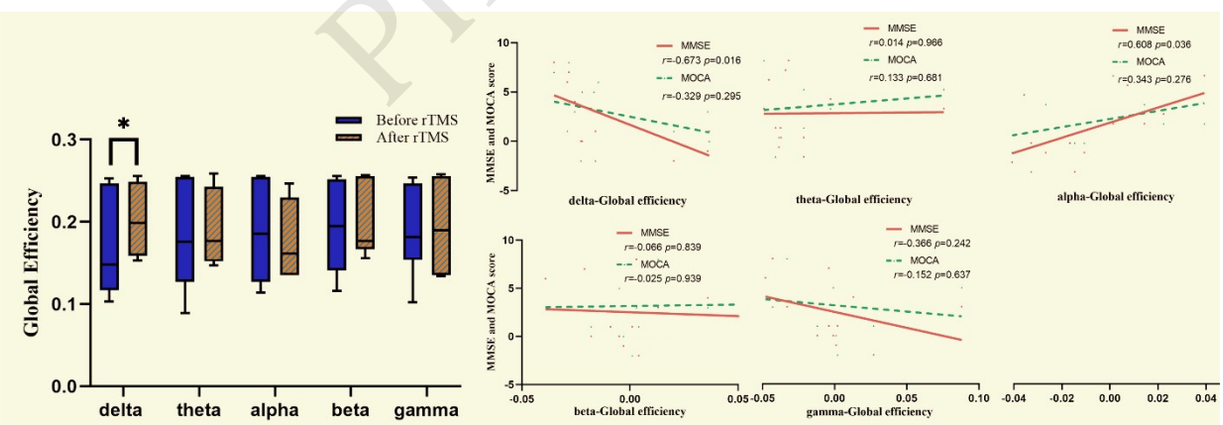

Fig. 9 A study on the effects of rTMS stimulation on brain functional networks in patients with AD

（a） Global efficiency of BFNs. ∗P< 0.05. （b） The correlation between the change in the global efficiency and the change in MMSE and MOCA.

that the frequency abnormalities observed in patients with AD can be ameliorated by modulating the intensity of oscillations across various frequency bands, potentially slowing the progression of the disease. The brain is a complex and highly efficient network of connections, while each brain region has relatively independent functions, connections between regions are engaged during complex tasks[36]. Hampstead *et al.* [37] when analyzing fMRI data from MCI and healthy groups, found disrupted FC in the hippocampus of MCI patients. Our study observed that following rTMS, AD patients demonstrated enhanced FC in the frontal and occipital regions, along with improved long-range connectivity between these areas. This enhancement accelerated network information transfer, thereby improving cognitive function in AD patients. Thus, a potential neurophysiological mechanism of TMS in treating AD may involve regulating oscillation intensity across various frequencies and enhancing connectivity in the frontal and occipital networks.

The application of graph theory analysis to elucidate the pathological features of AD remains in its nascent stages. Zhao *et al*. [38] found that the brain networks of AD patients and neurologically healthy control (NC) group both had small-world characteristics, and the small-world attributes were markedly pronounced in the AD group compared to the NC group, while the clustering coefficient, feature path length and local efficiency of AD group were also enhanced, indicating that the information transmission ability and efficiency of AD patients' brain functional networks were impaired[39-41]. In this study, a functional brain network was constructed using resting-state data and clinical scale scores of AD patients, followed by an analysis of its statistical characteristics and efficiency indicators. This study found that after rTMS, the node degree and clustering coefficient of the alpha band BFN increased, the characteristic path length decreased, and the clustering coefficient and global efficiency of the delta-band BFN increased. These results suggest that the alpha and delta band BFN in AD patients exhibited higher local aggregation and global functional integration. Additionally, the study found that degree and clustering coefficients decreased as MMSE and MOCA scores increased, while global efficiency increased with higher MMSE and MOCA scores. This suggests that as AD severity decreases, the local processing capacity of the brain network diminishes, while overall efficiency improves. The results demonstrate that exploring the topological properties of brain networks offers a robust theoretical foundation for the early diagnosis and clinical recognition of AD. Prior research on the topological properties of AD brain networks has rarely analyzed the correlation between clinical scales and network metrics such as average degree, clustering coefficient, characteristic path length, and global efficiency. Research indicates that rTMS induces significant changes in the connectivity and efficiency of brain functional networks, particularly in the delta and alpha frequency bands. These changes are associated with improvements in cognitive abilities measured by MMSE and MOCA scores, highlighting the potential therapeutic effects of rTMS on brain network dynamics and cognitive function. The correlation between scale changes, blood biomarkers, and network topological attributes suggests a potential link between blood biomarker alterations and topological attributes. This paper's exploration of the interplay between clinical scales, blood biomarkers, and brain functional connectivity aims to provide a more objective basis for the clinical diagnosis of AD.

This study acknowledges several limitations that warrant attention. Firstly, the research is constrained by a small sample size, which may introduce bias into the experimental outcomes. Future investigations should aim to address this by expanding the participant pool. Secondly, the focus of this study was predominantly on functional connectivity, with structural underpinnings being largely unexplored. Future research could benefit from incorporating structural imaging techniques to provide a more comprehensive understanding. Additionally, the study concentrated on resting-state functional connectivity, neglecting the dynamic aspects of connectivity. Future studies should consider examining the temporal dynamics of functional connectivity and their potential correlation with therapeutic outcomes to enhance the depth of understanding in this area.

4 Conclusion

This study analyzes the power of oscillatory bands—delta, theta, alpha, beta, and gamma—as well as the characteristic parameters of different bands of BFN, in AD patients, both prior to and following

rTMS. Specifically, the power of delta oscillations was observed to decrease, concomitant with a reduction in overall network correlation weakened after rTMS in AD patients, whereas alpha oscillation power increased accompanied by an enhancement in both global and local network correlation. The findings from this study propose promising therapeutic strategies for the management of AD.

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rTMS对阿尔茨海默病认知功能与脑网络连接的调控效应*

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摘要　目的　尽管现有研究证实高频重复经颅磁刺激（rTMS）靶向刺激背外侧前额叶皮层（DLPFC）可改善阿尔茨海默病（AD）患者的认知功能，但尚未在生物标志物水平上验证其有效性，且其介导的神经网络重构机制仍不明晰。本研究拟结合临床量表、血液生物标志物及脑电图（EEG）技术多角度探索rTMS对AD的调控效应及其神经网络响应机制。**方法**　本研究采用前瞻性双盲临床试验设计，纳入12例AD患者接受为期14 d的20 Hz的rTMS干预，基于临床量表评分-血液标记物水平-脑电进行基线期与干预后纵向对照研究。采用简易智力状态检查量表（MMSE）和蒙特利尔认知评估量表（MoCA）系统评估rTMS对AD患者总体认知功能的调控效应；通过日常生活能力量表（ADL）分析rTMS对AD患者日常生活能力的影响；结合汉密尔顿抑郁量表（HAMD）与神经精神科问卷评估TMS对AD患者神经精神症状的影响。统计分析刺激前后外周血神经退行性相关生物标志物水平，探究rTMS对AD相关分子代谢的调控效应。基于EEG功率谱动态演化特征探究TMS对神经节律的调控作用；最后基于相位同步（PLV）建立脑网络，并通过图论拓扑参数系统量化AD脑网络连接特性的影响。**结果**　rTMS刺激后，MMSE和MoCA评分显著升高（P<0.05），ADL、HAMD和NPI评分有所降低。血液标记物中Aβ1-40显著升高（P<0.05），p-tau181有所降低。delta和theta功率显着降低（P<0.05），gamma功率显着增加（P<0.05），而alpha和beta功率没有显着变化（P<0.05）。刺激后，delta、theta和alpha的聚类系数显着升高（P<0.05），delta频段的全局效率增加（P<0.05）。在alpha波段，网络平均度显著增加（P<0.05），同时特征路径长度显著降低（P<0.05）。进一步的相关性分析显示，HAMD与Aβ1-40、p-tau181呈负相关；MMSE和MOCA评分的变化与alpha频段节点度的变化呈负相关（P<0.05），与delta频段的聚类系数呈负相关（P<0.05）。然而，MMSE分数的变化与delta和alpha频段的全局效率变化呈正相关（P<0.05）。**结论**　20 Hz的rTMS可通过特异性调节AD患者的神经节律，增强神经元同步化能力及优化功能脑网络的拓扑结构有效强化振荡网络的功能整合，从而显著改善AD患者的认知功能，并提升β-淀粉样蛋白及tau蛋白的代谢清除效率。研究结果不仅证实rTMS作为AD辅助治疗手段的临床有效性，更凸显了多模态评估体系在AD病程监测中的核心价值。本研究为深化rTMS神经调控机制研究及开发精准化AD干预方案提供了关键的理论框架与数据支持。

关键词　经颅磁刺激，阿尔茨海默病，功率谱密度，脑电图，脑功能网络
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