Trial-by-Trial Variability of TMS-EEG in Healthy Controls and Patients With Depressive Disorder

Zikang Niu, Lina Jia, Yang Li, Lijuan Yang, Yi Liu, Siyuan Lian, Dan Wang, Wen Wang, Liu Yang, Weigang Pan, and Xiaoli Li

***Abstract*— Depressive disorder has been known to be associated with high variability in resting-state electroencephalography (EEG) signals. However, this phenomenon is often ignored in stimulus-related brain activities. This study proposed a new method to explore the EEG variability evoked by transcranial magnetic stimulation (TMS, TMS-EEG) in depressive disorder (DE) patients. The TMS-EEG data were collected from 34 DE patients and 36 healthy controls (HC). The maximum eigenvalue of the real binary correlation matrix, calculated between different trials using cross-correlation and surrogate methods, was extracted to assess trial-by-trial variability (TTV) of TMS-EEG. The new method was found to more sensitive and reliable than the standard deviation method. DE patients exhibited significantly smaller TTV in Gamma band and greater TTV in Delta band than HC. Furthermore, the HAMD-17 scores were negatively correlated with TTV values in Gamma band. This study represented the first investigation into the TTV in TMS-EEG data and revealed abnormal values in DE patients. Those findings enhance our understanding of TMS-EEG technology and provide valuable insights for studying the characteristics of DE.**

***Index Terms*— TMS-EEG, trial-by-trial variability, depressive disorder.**

Received 15 May 2024; revised 3 September 2024 and 2 October 2024; accepted 9 October 2024. Date of publication 28 October 2024; date of current version 31 October 2024. This work was supported in part by the STI2030 Major Projects under Grant 2021ZD0204300, in part by the Scientific Research Initiated by Beijing Anding Hospital Researchers under Grant 8-202173FS-2, and in part by Beijing Municipal Hospital Research and Cultivation Project under Grant PX2024069. *(Zikang Niu and Lina Jia contributed equally to this work.) (Corresponding authors: Liu Yang; Weigang Pan; Xiaoli Li.)*

This work involved human subjects or animals in its research. Approval of all ethical and experimental procedures and protocols was granted by Beijing Anding Hospital under Application No. 202173FS-2 and by Beijing Normal University under Application No. CNL_A_0010_005.

Zikang Niu is with the Aviation Psychology Research Office, Air Force Medical Center, Beijing 100142, China, and also with the State Key Laboratory of Cognitive Neuroscience and Learning, Beijing Normal University, Beijing 100875, China (e-mail: zikang_niu@163.com).

Lina Jia is with the Xuanwu Hospital, Capital Medical University, Beijing 100053, China, and also with Beijing Key Laboratory of Mental Disorders, Beijing Anding Hospital, Capital Medical University, Beijing 100088, China (e-mail: lnjia@xwhosp.org).

Yang Li, Lijuan Yang, Yi Liu, Siyuan Lian, Dan Wang, Wen Wang, and Weigang Pan are with Beijing Key Laboratory of Mental Disorders, Beijing Anding Hospital, Capital Medical University, Beijing 100088, China (e-mail: liyang2007428@hotmail.com; 1058669692@qq.com; liuyi0225@163.com; 1405307095@qq.com; wangdandoc@ccmu.edu.cn; ww0230@sina.com; panweigang@mail.ccmu.edu.cn).

Liu Yang is with the Aviation Psychology Research Office, Air Force Medical Center, Beijing 100142, China (e-mail: yangliuhenry@aliyun.com).

Xiaoli Li is with Guangdong Artificial Intelligence and Digital Economy Laboratory (Guangzhou), Guangzhou 510335, China, and also with the School of Automation Science and Engineering, South China University of Technology, Guangzhou 510641, China (e-mail: xiaolili@scut.edu.cn).

Digital Object Identifier 10.1109/TNSRE.2024.3486759

I. Introduction

As a common and recurrent disorder, depressive disorder (DE) is typically characterized by a lowered mood, reduced energy, and lowered enjoyment. Even with adequate full-course medication, most adult patients cannot effectively achieve remission [1], [2]. In the clinical diagnosis and evaluation of intervention effects, scales such as HAMD-17 are deemed inefficient and lack objectivity due to their reliance on the doctors' mature experience. Therefore, exploring the neurophysiological mechanism of DE is critical to help doctors and researchers understanding this disease and optimize treatment therapy. Over the past few decades, noninvasive electroencephalogram (EEG) has been favored by doctors and researchers for its high accuracy, safety, simplicity, and high temporal resolution in assessing DE [3], [4], [5].

Previous studies have indicated that the dorsolateral prefrontal cortex (DLPFC) plays a significant role in the pathophysiology of DE [6]. Lots of studies have demonstrated that synchronized EEG data, evoked by Transcranial Magnetic Stimulation (TMS, TMS-EEG) on the DLPFC, can reflect the cortical excitability, inhibition, plasticity, and the changes of connectivity patterns in DE patients [7], [8], [9]. It has been observed that the amplitude of N100 and N45 components of DE patients is lower than normal individuals, and these two components exhibit high accuracy in predicting disease state [10], [11]. In addition to being biomarkers for disease diagnosis, TMS-EEG excitability indicators can also predict the treatment effect of depression and monitor its biological response. For instance, after a Magnetic Seizure Therapy, a decrease of N100 was observed in patients with refractory depression, and this change associated with the Scale for Suicide Ideation. The accuracy, sensitivity, and specificity of this index in predicting suicidal ideation reached 89%, 90%, and 89%, respectively [12]. In addition to being excitatory indicators, TMS-induced brain connection activity can also be utilized as a biomarker for detecting and diagnosing depression. Electroconvulsive Therapy studies have found that

phase locking value induced by single TMS pulse in EEG can characterize depressive states and evaluate the antidepressant effects of neuromodulation [13].

Neural variability is a robust phenomenon which has been observed in intracellular membrane potential, extracellular recordings of spiking activity, and human EEG, electrocorticography, magnetoencephalography, and functional magnetic resonance imaging studies [14], [15], [16]. In resting-state EEG studies, nonlinear correlation analysis showed that depressive symptoms were associated with high variability of resting-state EEG signals [17], [18]. For instance, Jaworska et al. found reduced multiscale entropy at fine temporal scales, especially in frontal-central region, and the increased multiscale entropy value diffusely distributed in coarser temporal scales, which was related to the magnitude of the antidepressant treatment response [19]. Lee et al. found that the severity of major depressive disorder had a positive correlation with the long-range temporal autocorrelation of resting-state EEG [20]. Indexes calculated based on Higuchi's fractal dimension and Lempel-Ziv complexity correlation analysis indicated that the complexity of brain activity in DE patients was greater than that of healthy controls (HC), indicating a greater variability [21]. In recent years, studies have found that Fuzzy Measure Entropy, an improved method based on Fuzzy Entropy, was related to the severity of the disease [22].

In event-related potential studies, researchers typically analyze and discussed the data by averaging over trials. This averaging operation requires the assumption of linear superposition between basic random ongoing background activity and highly stereotyped, repeatable evoked responses [23], [24]. However, numerous studies have demonstrated that the neural response to repeated identical stimulation is highly variable [25], [26], [27]. One possible approach to measure neural variability is to calculate the trial-by-trial variability (TTV), as demonstrated in animal and human studies [16], [28]. Several pieces of evidence suggest that TTV can impact behavioral performance. First, greater TTV quenching in sensory cortices was associated with better perceptual performance [29]. Second, the behavioral performance can be improved by reducing the trial-by-trial response variability of the visual cortex in visual-related attention experiments [30]. In the DE, those activities are usually assessed through visual and auditory pathways. For instance, the inter-trial phase coherence of 40 Hz-auditory steady-state responses may serve as potential neurophysiological markers for early depression detection, and aid in understanding the underlying symptom severity in first-episode major depressive disorder [31]. Combining the steady-state topographical probe and 13Hz steady-state visually evoked potentials, Kemp et al. discovered that during acute serotonergic augmentation in a serotonergic antidepressant research, the response to pleasant valences of DE was potentiated while the response to unpleasant valences was suppressed [32].

However, the traditional TMS-EEG analysis, which was based on averaging all EEG trials, have ignored the neural variability between repeatable stimulations. In order to investigate the TTV of TMS-EEG data and the abnormal variability in DE patients, we collected TMS-EEG data of DE patients and HC. Based on the cross correlation and surrogate method, we extracted the maximum eigenvalue of the real binary correlation matrix among different trial data to assess the TTV of TMS-EEG.

TABLE I
DEMOGRAPHIC AND CLINICAL DATA

Group	HC	DE
Sample size	36	34
Age(mean±std)	33.72z±12.21	33.44±11.67
Sex(male/female)	5/31	3/31
HAMD-17		25.09±5.70

HC: healthy control; DE: depressive disorder, HAMD-17: Hamilton Depression Scale -17 items.

II. MATERIAL AND METHODS

A. Participants

We recruited 34 DE patients and 36 age-matched HC in Beijing Anding Hospital and Beijing Normal University. All participants were right hand, aged between 18 and 60 years old, and Wilcoxon's signed test results showed no significant age differences between groups (detailed information in Table I). The DE patients were diagnosed according to ICD-10 criteria and didn't have any other mental illnesses. In addition, individuals with a history of neurological disorder, seizures, head injury and other conditions that made them unsuitable for receiving TMS stimulation were excluded. The Hamilton Depression Scale-17 item (HAMD-17) of DE patients were used to assess the severity of depression in DE patients by two trained doctors who underwent consistency training for assessments. Finally, patients with a HAMD-17 score greater than 17 were enrolled in the study and TMS-EEG data were collected within 24 hours after the HAMD-17 evaluation. Due to the actual situation of patients and hospital, only medicated patients were included in the study without controlling the type of medicine. To minimize the potential influence of medicine on the results, only patients who had taken the medicine steadily for more than half a month were recruited and their TMS-EEG data were collected at least 8 hours after taking their medicine.

B. Experimental Equipment and Procedure

Based on a large number of previous studies [33], [34], [35], we utilized the classical experimental equipment and single pulse stimulation paradigm, which have been previously employed in the HC and DE patients. In this experiment, we used a Rapid2 system with D70 coil (Magstim, UK) to stimulate the DLPFC of participants' brain. Additionally, 64-channel synchronization EEG data (EASYCAP GmbH and BrainAmp MR amplifier, Brian Products, Germany) were collected in an electrically and acoustically shielded room at a sampling rate of 2500 Hz.

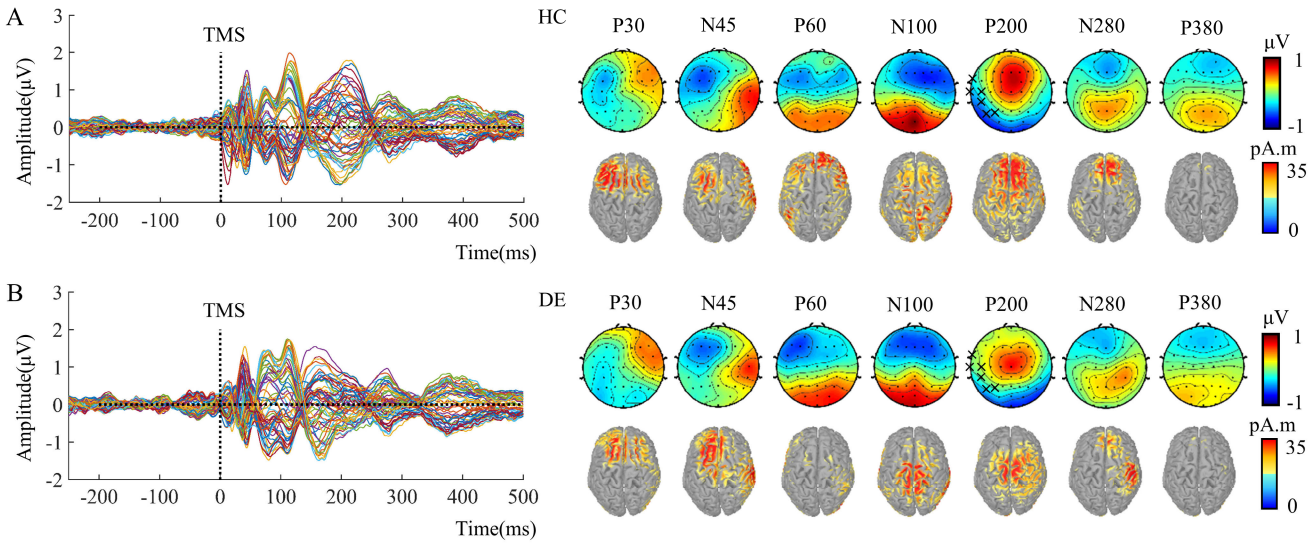

Fig. 1. TMS evoked potentials (TEPs) of healthy controls (A) and depression patients (B). Left: Butterfly plot of all electrode waveforms; right: voltage distribution in 2D and current density map at the cortex in each peak. "×" indicates p<0.05 between HC and DE in one channel.

During the experiment, all electrode impedances were maintained below 5kΩ and participants were instructed to sit comfortably in a chair with their arms at rest. First, located C3 position by 10-20 system, and determined the M1, which can be induced the max motor-evoked potentials in the first dorsal interosseous muscle, with a 45° angle between the coil handle and Anterior posterior sagittal line, by using the conventional nonnavigated strategies in Roland's paper [36]. Then stimulated the M1 to determine the resting motor threshold, which was defined as the minimum stimulation intensity to evoke 50μV peak-to-peak motor evoked potential amplitude at least 5 of 10 trials in the relaxed first dorsal interosseous muscle [37]. After that, stimulate the DLPFC in a similar way to M1, which was located by the BeamF3 method [38], for a total 120 times with a 2∼2.5s by using the 110% RMT. In order to avoid possible auditory responses and the bone-conducted sound caused by the TMS click, the participants wore earphone with a white noise masking sound while a thin sponge was placed under the coil [39].

C. TMS-EEG Data Preprocessing

All EEG data were preprocessed by using the TESA toolbox [40], [41], [42] with the following procedures: (1) Found bad channels and replaced them with the superfast spherical interpolation method; (2) Extracted epochs from −900 to 900ms and performed baseline correction by using data from −500ms to 0ms (2) Removed -2 to 10ms data, which contains TMS pulse artifacts, and interpolated them by using the cubic interpolation method; (4) Rejected bad trials and down sampled data to 512 Hz; (5) Performed the first ICA decomposition and removed muscle, electrical and movement noise components (number=9.765±2.254) from the first 15 components; (6) Filtered the data by the fourth-order Butterworth band-stop filter (48-52 Hz) and fourth-order Butterworth band-pass filter (!-100 Hz); (7) Performed the first ICA decomposition and removed muscle, blink, eye movement and electrode noise artifact (number=17.838±4.730) from all components(number=43.221±3.689); (10) Converted the data into average reference. and extracted the data from -250 to 500ms, for the further analysis.

D. TMS-Evoked Potential Analysis

An average TEP signal was obtained from each electrode. The butterfly outputs of HC and DE are shown in Fig. 1. To further analysis the differences of each component between the two groups, we also performed 2D topographic scalp mapping and source estimation analysis.

In addition, the source location was calculated using Brainstorm MATLAB toolbox with the follows [43]: (1) Due to the absence of MRI data, co-registered the EEG cap to the Colin27 head model; (2) Second, used the OpenMEEG to computed the geometric head model [44]; (3) Third, calculated the noise covariance matrix based on the -200∼-20ms data; (4) Finally, constrained the dipoles o the cortex surface and computed the inverse solution by using the current density analysis.

E. Discrete Stationary Wavelet Transform Analysis

First, since the length of input data must be divided by 2^Q in discrete stationary wavelet transform, we extended the TMS-EEG data to a length of 512 by using the periodic extension method:

$$S(t) = [X(64), \cdots, X(1), X(1), \cdots, X(H), \\ X(H), \cdots, X(H-64+1)], H = 384 \quad (1)$$

where X is the original signal of TMS-EEG, H is the length of X and t is the time variable of TMS-EEG's trial signal X with the unit is $\frac{1}{512}ms \approx 0.00195ms$;

After that, computed the discrete stationary wavelet transform using the method employed by Dutt and Saadeh [45].

Then, we extracted the 1∼8 levels' detail coefficients (CD, CD1∼8) in -200∼400ms for further analysis.

Since the binary based filtering process of discrete stationary wavelet transform, Nyquist Sampling Theorem and 512Hz sampling rate of data, the CD1∼8 can reflect the signal X's information in the 128∼256Hz (CD1), 64∼128Hz (CD1), 32∼64Hz (CD2), 16∼32Hz (CD3), 8∼ 16Hz (CD4), 4∼8Hz (CD5), 2∼4Hz(CD6) and 1∼2Hz(CD7) frequency bands, respectively.

F. Trial-by-Trial Variability Analysis

A. Based on the maximum eigenvalue of correlation matrix, we calculated TTV values of the CD1∼8. Taking the CD3 series (d) as an example, the algorithm is as follows:

(1) Extracted the TMS-EEG data in T_{pre} and T_{post} form N TME-EEG trials and normalized them using z score method;

where the T_{pre} is -200∼0ms, T_{post} is 0∼400ms and N is the number of TMS-EEG trial.

(2) Calculated the cross correlation $C_{i,j}$ between two trials in T_{pre} period:

$$C_{i,j} = \frac{1}{M}\sum_{t=t_1}^{t_2} d_{i,t} \times d_{j,t} \tag{2}$$

where $d_{i,t}$ and $d_{j,t}$ is CD3 series of trial i or j ($1 \le i, j \le N$) in time point t ($1 \le t \le M$), M is the data length of $d_{i,t}$ from t_1 to t_2, $t_1 = -200ms, t_2 = 0ms$.

(3) In order to test the reliability of cross correlation between trials, generated 50 fake TMS-EEG data by randomly shuffling each single trial TMS-EEG data in the time ranging from -200 to 0ms. And, calculated the cross correlation $CF_{i,j}$ from those fake data. Then, transformed the cross correlation matrix into a binary matrix ($CB_{i,j}$):

$$CB_{i,j} = \begin{cases} 1 \; if \;\; C_{i,j} \le A1_{i,j} \\ 0 \; if \;\; A1_{i,j} < C_{i,j} < A2_{i,j} \\ 1 \; if \;\; C_{i,j} \ge A2_{i,j} \end{cases} \tag{3}$$

where $A1_{i,j}$ and $A2_{i,j}$ is the 5% and 95% largest value of the 50 fake TMS-EEG data's $CF_{i,j}$ between trial i and j;

(4) Decomposed the binary correlation matrix CB using the eigenvalue decomposition method:

$$CBv_i = \lambda_i v_i \tag{4}$$

where λ_i is the eigenvalue and v_i is the corresponding eigenvector;

(5) Normalized the max eigenvalue $ME_{T_{pre}}$ by the number of trials.

$$ME_{T_{pre}} = \frac{\max(\lambda_i)}{N} \tag{5}$$

Since $CB_{i,j}$ is a real symmetric matrix with all elements are 0 or 1, the $ME_{T_{pre}}$ value are in the range of $[1/60, 1]$ which can provide information about the synchronization of neural activity over trials. If the TMS-EEG data series are completely uncorrelated over trials, then $CB_{i,j}$ is an identity matrix. So the $\max(\lambda_i)$ is 1 and $ME_{T_{pre}}$ is equal to 1/60. If the TMS-EEG data series are completely correlated over trials, then all the elements of $CB_{i,j}$ are equal to 1. So the $\max(\lambda_i)$ is N and $ME_{T_{pre}}$ is equal to 1.

(5) Repeat steps (2)∼(5), extracted the $ME_{T_{\text{post}}}$ of the TMS-EEG signals in T_{post}.

(6) Calculated the TTV_{ME}:

$$TTV_{ME} = \frac{ME_{T_{post}} - ME_{T_{pre}}}{ME_{T_{pre}}} \tag{6}$$

B. To demonstrate the sensitivity of the TTV_{ME}, we also calculated each participant's TMS evoked neural variability (TTV_{STD}) using standard deviation method, which is usually used in EEG signal variability studies but ignores the similarity in time dynamic characteristic between different trial series, The steps were as follows:

(1) Calculated the mean value of variability in a time period T_{pre}:

$$Var_{STD_{T_{pre}}} = \frac{\sum_{t=t_1}^{t_2} \sqrt{\frac{\sum_{i=1}^{N}(d_{i,t} - \bar{d}_t)}{N}}}{M} \tag{7}$$

where $\bar{d}_t$ is the mean value of the sequence $d_{i,t}$ over trials, N is the number of TMS-EEG trials. M is the data length of $d_{i,t}$ from t_1 to t_2, $t_1 = -200ms, t_2 = 0ms$.

(2) Same as step (1), extracted the $Var_{STD_{T_{post}}}$ of the TMS-EEG signals in T_{post}.

(3) Calculated the TTV_{STD}:

$$TTV_{STD} = \frac{Var_{STD_{T_{\text{pos}_t}}} - Var_{STD_{T_{\text{pre}}}}}{Var_{STD_{T_{\text{pre}}}}} \tag{8}$$

It should be noticed that the TTV_{STD} describes desynchronization variability over TMS-EEG trials while the TTV_{ME} represent the synchronization variability. To describe the variability in a uniform manner, we used "variability" to represent desynchronization variability. In other words, when the TTV_{ME} is larger, the TTV of TMS-EEG is small.

G. Statistical Analysis

To study the differences between the TMS-EEG data at HC and DE, we first conducted cluster based permutation test between two groups on all channels' TEPs. Subsequently, to examine the changes in TTV induced by TMS, we performed cluster based permutation test on Var_{STD} of TTV_{STD} before and after TMS pulse, as well as on the real correlated trial number of TTV_{ME}. In the TTV_{ME} method, N are set as 10∼75 to explore the influence of trial number on TTV_{ME}. Thirdly, we executed cluster based permutation test to examine the significant differences of TTV_{ME} or TTV_{STD} value between two groups [46]. In order to reduce the probability of false positive, Bonferroni procedures was performed between two TTV calculating method, that is to say, the alpha levels (p-values) was set to $0.05/2 = 0.025$. Finally, in the correlation analysis between TTV_{ME} or TTV_{STD} and HAMD-17 of DE, we also performed cluster based permutation correlation analysis, which was similar with the cluster based permutation test, between the symptom scores and every electrode.

TABLE II
THE DETAIL STATISTICAL VALUES OF TEP

Channel	HC(μV)	DE(μV)	p Value	T Value
P3	-0.645±1.003	-0.164±0.889	0.020	2.092
P5	-1.086±1.149	-0.581±0.900	0.024	2.014
C5	-0.444±0.902	0.166±0.819	0.002	2.918
CP5	-0.875±0.967	-0.151±0.768	0.001	3.415
T7	-0.916±1.161	-0.162±0.990	0.003	2.883
FT7	-0.569±1.053	0.008±0.869	0.008	2.467

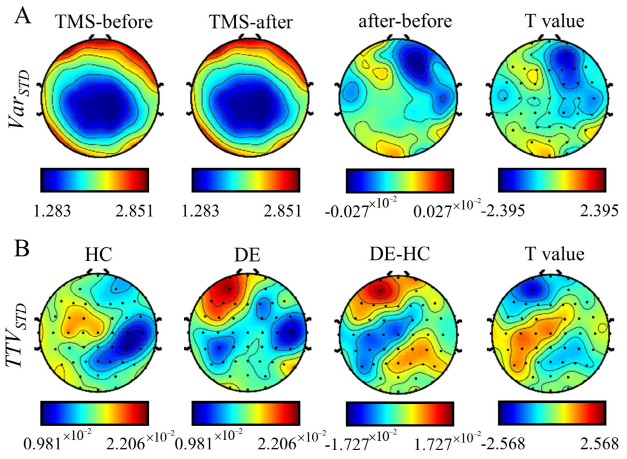

Fig. 2. TTV_{STD} analysis of CD3. (A) analysis of Var_{STD} before and after TMS pulse in HC; (B) Topographic distribution and statistical of analysis of TTV_{STD} between HC and DE in CD3. "×" indicates p<0.025.

III. RESULTS

A. TMS Evoked Potentials

Fig. 1 A and B showed the average TEP and source location analysis of the two groups. After TMS pulse stimulation, all subjects exhibited seven components (P30: 33ms, N45: 44ms, P60: 73ms, N100: 113ms, P200: 171 or 200ms, N280: 276ms, P380: 389ms). In group level, the latency of DE in the P200 component was shorter than that of HC in group level. So the P200 latency of DE were respectively set as 171ms while it was set at 200ms for HC. Statistical results indicated that the P200 amplitudes of channel P3, P5, C5, CP5, T7 and FT7 in DE were significantly larger than those in HC. The detail values were showed in Table II.

B. Trial-by-Trial Variability

In TTV_{STD} method, statistical analysis showed that there was no significant difference in Var_{STD} before and after TMS pulse. For instance, Fig.2A showed the CD3's Var_{STD} value in HC. Compared with HC, DE had lager TTV_{STD} in the left frontal and right central-parietal lobe and smaller TTV_{STD} in the left central, left parietal and middle frontal lobe. However, no significant difference was found between DE and HC (Fig.2D).

Fig. 3 showed the TTV_{ME} results of two groups. First, to investigate the impact of trial number N on TTV_{ME}, we randomly selected 10 to 75 TMS-EEG trials to calculated the TTV_{ME} and observed that the fluctuation of TTV_{ME} remained stable when N was greater than 30 (Fig. 3 A). In order to capture more brain signals, N was set to be 60 for calculating of cross correlation. We also analyzed the real correlated trial number of two groups and found that TMS increased the correlation over trials. For instance, in HC group, TMS significantly increased this value of CD3 in lots of channels and the channel in left central and middle frontal lobe showed the most changes (Fig. 3 B).

TABLE III
THE DETAIL STATISTICAL VALUES OF TTV_{ME}

	Channel	HC(μV)	DE(μV)	T Value	T Value
CD3	T8	0.143±0.246	0.014±0.156	0.021	-2.063
	Pz	0.128±0.194	0.033±0.145	0.010	-2.398
	CP2	0.126±0.164	0.051±0.136	0.020	-2.103
	CP6	0.146±0.299	0.054±0.146	0.024	-2.015
	CP4	0.140±0.234	0.088±0.168	0.023	-2.028
	C6	0.132±0.247	0.052±0.137	0.017	-2.155
	FT8	0.158±0.223	0.059±0.167	0.011	-2.345
	TP8	0.152±0.289	0.045±0.126	0.018	-2.127
	CPz	0.148±0.204	0.057±0.155	0.014	-2.252
CD7	Pz	-0.179± 0.166	-0.065± 0.200	0.005	2.616
	CP1	-0.158± 0.140	-0.073± 0.178	0.013	2.261
	CPz	-0.161± 0.171	-0.082± 0.143	0.018	2.129

Statistical analysis revealed significant differences in TTV_{ME} of CD3 and CD7 between two groups (Fig. 3 C and D). In CD3, HC had a greater TTV_{ME} in left frontal and central lobe while those values of DE had higher values in left central lobe. Compared with HC, DE showed significant smaller values in central-parietal and right temporal lobe. In CD7, HC exhibited higher value in the right frontal and left temporal lobes while DE showed higher values in the right parietal and parietal-occipital lobe. Compared with HC, DE had a significant larger value in the parietal lobe. The detail values were showed in Table III.

C. Correlation Between TTV and HAMD-17

The correlation analysis showed there were significant correlations between TTV_{ME} and depressive symptoms of DE (Fig. 4). In CD3, the TTV_{ME} values of some channels were negatively correlated with HAMD-17 scale. Those channels were distributed in two clusters: cluster 1 (left parietal region), C4, C6, FC6 and FC4; cluster 2 (left parietal region), P3, P5, P7 and CP5. The detail values were as shown in TABLE IV.

IV. DISCUSSION

A. TMS Evoked Potentials

TEPs, which are evoked by TMS on DLPFC, are typically defined as P30, N45, P60, N100, and P200 [47]. Our study also found these components using supraliminal stimulation

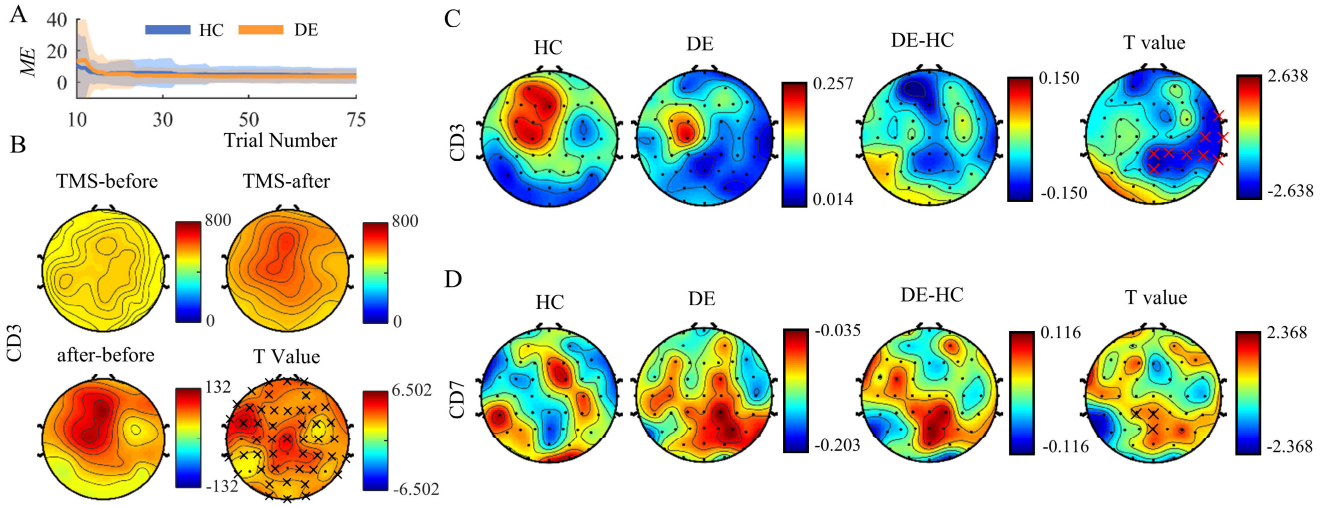

Fig. 3. TTV_{ME} analysis. (A) The TTV_{ME} value of CD3 for different trial number in channel Cz. (B) analysis of real correlated trial number over TMS-EEG trials before and after TMS pulse in HC. (C) and (D) Topographic distribution and statistical analysis of CD3 and CD7's TTV_{ME}. "×" indicates $p<0.025$.

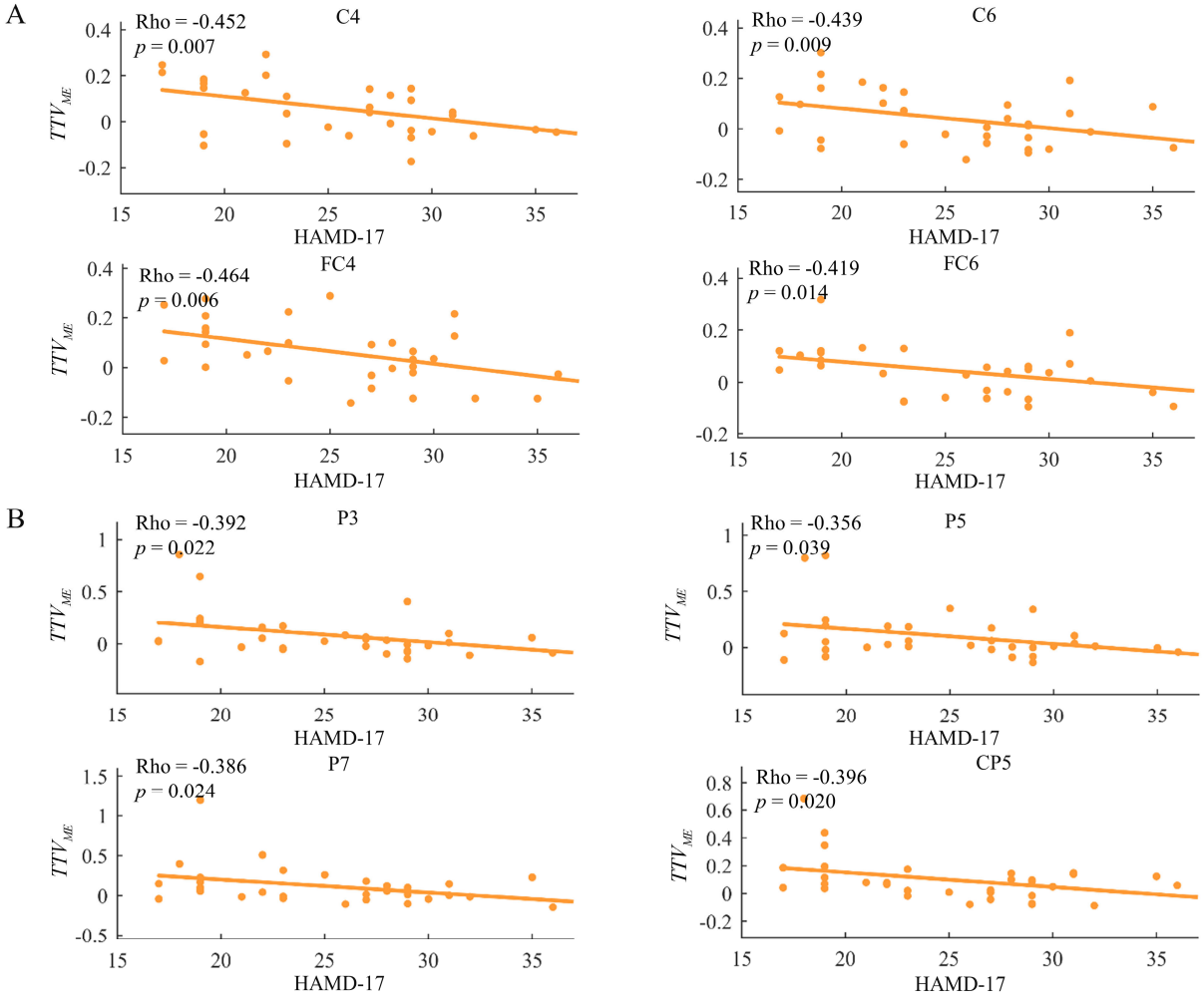

Fig. 4. Correlation analysis between TTV_{ME} of CD3 and clinical HAMD-17. (A) Cluster 1: right central region; (B) Cluster 2: left parietal region.

patterns of TMS pulses. The generation of TEPs is associated with the spatial and temporal summation of postsynaptic potentials which originate from a large number of pyramidal neurons and interneurons. These components are thought to

TABLE IV
THE DETAIL STATISTICAL VALUES OF TTV_{ME}

Channel		RHO	p Value	F Value
Cluster 1	C4	-0.452	0.007	-2.868
	C6	-0.439	0.009	-2.762
	FC4	-0.464	0.006	-2.959
	FC6	-0.419	0.014	-2.612
Cluster 2	P3	-0.392	0.022	-2.409
	P5	-0.356	0.039	-2.154
	P7	-0.386	0.024	-2.366
	CP5	-0.396	0.020	-2.442

be important indices of cortical excitability. For instance, early TEPs (such as N45 and P60) are associated with postsynaptic inhibitory Gamma aminobutyric acid A receptors and glutamatergic activity, while the N100 component is involved in cortical inhibition mediated by presynaptic and postsynaptic Gamma aminobutyric acid B receptors [48], [49].

Previous studies have demonstrated that the TMS-EEG excitability can not only be used as a diagnostic biomarker for depression but also to monitor biological responses to treatment. For instance, compared with HC, major depressive disorder patients exhibit lower N100 and N45 amplitudes and greater P200 amplitudes [10], [50]. Additionally, Theta burst stimulation can reduce the N45 in the right intraparietal lobules, while the administration of Baclofen (a GABAB agonist), can increase the N100 amplitude in healthy participants [48], [51]. Unfortunately, our study only found significant greater P200 amplitude of DE in channel P3, P5, C5, CP5, T7 and FT7, while no significant difference was observed in N45 or N100. Previous studies, which set the intensity of TMS pulse to less than 100%RMT, have reported significant differences in the early TEP between DE and HC. However, such differences are rarely reported when setting the intensity of TMS pulse to larger than 100%RMT [50], [52]. Only Dhami found a significant P30 difference among youth with depression [51]. Therefore, we hypothesis that there are two possible reason. One possible reason may be the different clinical condition. For example, other studies usually recruited young, fist-episode or severely suicidal patients which this paper are not identical with. In addition to the clinical factors of patients, the TMS pulse intensity (110% RMT) may be so large that it failed to evoked the a significant different early TEP between the DE and HC.

B. TTV_{STD} vs TTV_{ME}

Neural activity is variable over time and across stimulation trials. Among the trials with identical stimulation, neural variability is small before stimulus onset and significantly increased after stimulus presentation. In traditional event-related potential experiments, the standard deviation method is usually used to describe neural variability of cognitive processing pathways after the brain receives external stimuli through the sensory organ [53]. In this study, we first studied the TTV in the TMS-EEG data and developed a new method to assess the neural variability of TMS-EEG data. The results showed that TTV_{ME} were changed after TMS stimulation. From this phenomenon, we assume that the reduced reproducibility of neural activity may be attributed to the TMS stimulation on DLFPC leading to the depolarization of neurons and evoked action potentials, which phenomenon can also be observed in other event-related potential studies and EEG calculation methods [9], [54], [55]. Our results indicate that TTV_{ME} is more sensitive for representing abnormal in DE patients and reducing falsity of the TTV index than TTV_{STD}.

C. Trial-by-Trial Variability and Depressive Disorder

In the pathophysiology of clinical disorders, oscillations can provide important information about the effects of plasticity-inducing protocols on brain activity. For instance, specific frequency bands can be associated with specific behaviors or cognition [56]. Gamma band activities are commonly used to characterize depressive disorder and schizophrenia patients [57], [58], [59]. Event-related potential and resting-state EEG studies have shown that Gamma oscillations have a good diagnostic capability for DE [3], [60]. Unlike traditional event-related potential, TMS–EEG offers an opportunity to measure those activities in different frequency bands of the brain without involving the processing of sensory pathways. Our results revealed that DE patients had significantly smaller variability of CD3, which mainly reflects the gamma band information (32∼64Hz), than HC. Pizzagalli and Isomura both found that Gamma activities, which was evoked by auditory steady-state responses or Flanker task, can be used to distinguish depressed patients from healthy controls, even unipolar depression and bipolar disorder in their own experiments [54], [61]. In clinical studies, Gamma oscillations can not only be used to distinguish the depressive disorder, but also can be the potential biomarker for the treatment. For instance, Pellicciari and his colleague found the gamma oscillation, which were evoked by TMS in DE patients, was asymmetrical in bilateral DLPFC, and a 10-day Theta Burst Stimulation can reduce this abnormal phenomena [62]. In a paired associative stimulation studies, Noda found that compared to HC, the changes of Gamma power and Theta-Gamma coupling activities were significantly lower in DE [64]. Our results provide another important evidence for the abnormal Gamma brain activity in depressed patients.

D. Correlation Between TTV and HAMD-17

In this study, we discovered a significant negative correlation between the HAMD-17 and TTV_{ME} of CD3 which mainly reflecting the Gamma(32∼64Hz) band information. Similar results have been found in previous studies of patients with DE. For instance, one study reported a 50% reduction of Gamma aminobutyric acid in the occipital cortex of major depressive disorder patients while it is also significantly associated with the severity of depression [63]. High-frequency activity is known to be associated with the continuation of the cognitive setting and the dominance of endogenous top-down influences, which override the effect of potentially

novel, or unexpected, external events [64]. Our results indicated TTV_{ME} had a negative correlation with HAMD-17. We hypothesis that our findings may be related to a defect in receiving and processing new information in depressed patients. Future experiments need to be designed to investigate the difference between TMS-EEG and other states.

V. Conclusion

In this study, we not only proposed a new, more sensitive, and reliable method to assess TTV based on the maximum eigenvalue of the cross-correlation matrix, but also conducted the first study of TTV in TMS-EEG data. Additionally, we observed significantly smaller TTV in Gamma band and greater TTV in Delta band in DE patients. Furthermore, the HAMD-17 scores were negatively correlated with TTV values in Gamma band. Our findings may contribute to a better understanding of the capabilities of TMS-EEG technology and the specific characteristics associated with depressive disorders for researchers and medical professionals. However, our study still has some limitations: 1. Lack of MRI data to help localize the M1 motor region and DLPFC; 2. Lack of a paired-pulse TMS–EEG experiment (e.g. long interval intracortical inhibition) to examine the inhibition of neural variability; 3. No intensity lower than 100% RMT were used as the TMS pulse intensity. In future studies, we will recruit more DE patients to validate the findings and apply this method to other patients with different brain diseases. Moreover, new studies should be designed to resolve these limitations.

Acknowledgment

The authors are particularly thankful to Beijing Anding Hospital and Beijing Normal University for their cooperation in recruiting the HC and DE patients. They would also like to thank Xin Liu for their valuable helps on grammar of manuscript.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this article.

References

[1] R. S. McIntyre et al., "Treatment-resistant depression: Definitions, review of the evidence, and algorithmic approach," (in English), *J. Affect. Disorders*, vol. 156, no. 1, pp. 1–7, Mar. 2014.

[2] *The ICD-10 Classification of Mental and Behavioural Disorders*, WHO, Geneva, Switzerland, 1993.

[3] F. S. de A. Neto and J. L. G. Rosa, "Depression biomarkers using non-invasive EEG: A review," *Neurosci. Biobehavioral Rev.*, vol. 105, pp. 83–93, Oct. 2019.

[4] S. Yasin, S. A. Hussain, S. Aslan, I. Raza, M. Muzammel, and A. Othmani, "EEG based major depressive disorder and bipolar disorder detection using neural networks: A review," (in English), *Comput. Methods Programs Biomed.*, vol. 202, Apr. 2021, Art. no. 106007.

[5] H. Zhang et al., "The applied principles of EEG analysis methods in neuroscience and clinical neurology," *Mil. Med. Res.*, vol. 10, no. 1, p. 67, Dec. 2023.

[6] C. R. C. Fatt et al., "Dorsolateral prefrontal cortex and subcallosal cingulate connectivity show preferential antidepressant response in major depressive disorder," *Biol. Psychiatry, Cognit. Neurosci. Neuroimaging*, vol. 6, no. 1, pp. 20–28, Jan. 2021.

[7] P. Canali, "A role for TMS/EEG in neuropsychiatric disorders," *Neurol., Psychiatry Brain Res.*, vol. 20, no. 2, pp. 37–40, Jun. 2014.

[8] P. Canali et al., "Shared reduction of oscillatory natural frequencies in bipolar disorder, major depressive disorder and schizophrenia," *J. Affect. Disorders*, vol. 184, pp. 111–115, Sep. 2015.

[9] Z. Ding, L. Guan, W. He, H. Gu, Y. Wang, and X. Li, "Spatial characteristics of closed-loop TMS-EEG with occipital alpha-phase synchronized," (in English), *Biomed. Signal Process. Control*, vol. 83, May 2023, Art. no. 104650.

[10] D. Voineskos et al., "Altered transcranial magnetic stimulation–electroencephalographic markers of inhibition and excitation in the dorsolateral prefrontal cortex in major depressive disorder," (in English), *Biol. Psychiatry*, vol. 85, no. 6, pp. 477–486, Mar. 2019.

[11] P. Belardinelli et al., "TMS-EEG signatures of glutamatergic neurotransmission in human cortex," *Sci. Rep.*, vol. 11, no. 1, p. 8159, Apr. 2021.

[12] Y. Sun et al., "Indicators for remission of suicidal ideation following magnetic seizure therapy in patients with treatment-resistant depression," (in English), *JAMA Psychiatry*, vol. 73, no. 4, p. 337, Apr. 2016.

[13] E. Miyauchi, M. Ide, H. Tachikawa, K. Nemoto, T. Arai, and M. Kawasaki, "A novel approach for assessing neuromodulation using phase-locked information measured with TMS-EEG," (in English), *Sci. Rep.*, vol. 9, no. 1, p. 428, Jan. 2019.

[14] M. M. Churchland et al., "Stimulus onset quenches neural variability: A widespread cortical phenomenon," *Nature Neurosci.*, vol. 13, no. 3, pp. 369–378, Mar. 2010.

[15] B. J. He and J. M. Zempel, "Average is optimal: An inverted-U relationship between trial-to-trial brain activity and behavioral performance," (in English), *PLoS Comput. Biol.*, vol. 9, no. 11, Nov. 2013, Art. no. e1003348.

[16] A. Arazi, G. Gonen-Yaacovi, and I. Dinstein, "The magnitude of trial-by-trial neural variability is reproducible over time and across tasks in humans," *eneuro*, vol. 4, no. 6, p. e0292, Nov. 2017.

[17] S. Yun and B. Jeong, "Aberrant EEG signal variability at a specific temporal scale in major depressive disorder," *Clin. Neurophysiol.*, vol. 132, no. 8, pp. 1866–1877, Aug. 2021.

[18] Z. Niu et al., "Scale-free dynamics of microstate sequence in negative schizophrenia and depressive disorder," *Comput. Biol. Med.*, vol. 143, Apr. 2022, Art. no. 105287.

[19] N. Jaworska, H. Wang, D. M. Smith, P. Blier, V. Knott, and A. B. Protzner, "Pre-treatment EEG signal variability is associated with treatment success in depression," (in English), *NeuroImage, Clin.*, vol. 17, pp. 368–377, Oct. 2018.

[20] J. S. Lee, B. H. Yang, J. H. Lee, J. H. Choi, I. G. Choi, and S. B. Kim, "Detrended fluctuation analysis of resting EEG in depressed outpatients and healthy controls," *Clin. Neurophysiol.*, vol. 118, no. 11, pp. 2489–2496, Nov. 2007.

[21] M. Bachmann, J. Lass, A. Suhhova, and H. Hinrikus, "Spectral asymmetry and Higuchi's fractal dimension measures of depression electroencephalogram," (in English), *Comput. Math. Methods Med.*, vol. 2013, 2013, Art. no. 251638.

[22] K. Kalev, M. Bachmann, L. Orgo, J. Lass, and H. Hinrikus, "Lempel-ziv and multiscale lempel-ziv complexity in depression," (in English), in *Proc. 37th Annu. Int. Conf. IEEE Eng. Med. Biol. Soc. (EMBC)*, Aug. 2015, pp. 4158–4161.

[23] J. I. Aunon, C. D. Mcgillem, and D. G. Childers, "Signal-processing in evoked-potential research—Averaging and modeling," (in English), *Crit. Rev. Bioeng.*, vol. 5, no. 4, pp. 323–367, 1981.

[24] P. D. Pelo, M. D. Tommaso, A. Monaco, S. Stramaglia, R. Bellotti, and S. Tangaro, "Trial latencies estimation of event-related potentials in EEG by means of genetic algorithms," *J. Neural Eng.*, vol. 15, no. 2, Apr. 2018, Art. no. 026016.

[25] A. Arieli, A. Sterkin, A. Grinvald, and A. Aertsen, "Dynamics of ongoing activity: Explanation of the large variability in evoked cortical responses," *Science*, vol. 273, no. 5283, pp. 1868–1871, Sep. 1996.

[26] M. N. Shadlen and W. T. Newsome, "The variable discharge of cortical neurons: Implications for connectivity, computation, and information coding," *J. Neurosci.*, vol. 18, no. 10, pp. 3870–3896, May 1998.

[27] A. Gramfort, R. Keriven, and M. Clerc, "Graph-based variability estimation in single-trial event-related neural responses," *IEEE Trans. Biomed. Eng.*, vol. 57, no. 5, pp. 1051–1061, May 2010.

[28] S. Musall, M. T. Kaufman, A. L. Juavinett, S. Gluf, and A. K. Churchland, "Single-trial neural dynamics are dominated by richly varied movements," *Nature Neurosci.*, vol. 22, no. 10, pp. 1677–1686, Oct. 2019.

[29] A. Schurger, I. Sarigiannidis, L. Naccache, J. D. Sitt, and S. Dehaene, "Cortical activity is more stable when sensory stimuli are consciously perceived," (in English), *Proc. Nat. Acad. Sci. USA*, vol. 112, no. 16, pp. E2083–E2092, Apr. 2015.

[30] J. F. Mitchell, K. A. Sundberg, and J. H. Reynolds, "Differential attention-dependent response modulation across cell classes in macaque visual area V4," *Neuron*, vol. 55, no. 1, pp. 131–141, Jul. 2007.

[31] S. Liu et al., "Neurophysiological markers of depression detection and severity prediction in first-episode major depressive disorder," *J. Affect. Disorders*, vol. 331, pp. 8–16, Jun. 2023.

[32] A. H. Kemp, M. A. Gray, R. B. Silberstein, S. M. Armstrong, and P. J. Nathan, "Augmentation of serotonin enhances pleasant and suppresses unpleasant cortical electrophysiological responses to visual emotional stimuli in humans," *NeuroImage*, vol. 22, no. 3, pp. 1084–1096, Jul. 2004.

[33] S. Tremblay et al., "Clinical utility and prospective of TMS-EEG," *Clin. Neurophysiol.*, vol. 130, no. 5, pp. 802–844, 2019.

[34] K.-X. Cao et al., "TMS-EEG: An emerging tool to study the neurophysiologic biomarkers of psychiatric disorders," *Neuropharmacology*, vol. 197, Oct. 2021, Art. no. 108574.

[35] E. Kallioniemi, J. Saari, F. Ferreri, and S. Määttä, "TMS-EEG responses across the lifespan: Measurement, methods for characterisation and identified responses," (in English), *J. Neurosci. Methods*, vol. 366, Jan. 2022, Art. no. 109430.

[36] R. Sparing, D. Buelte, I. G. Meister, T. Pauš, and G. R. Fink, "Transcranial magnetic stimulation and the challenge of coil placement: A comparison of conventional and stereotaxic neuronavigational strategies," *Human Brain Mapping*, vol. 29, no. 1, pp. 82–96, Jan. 2008.

[37] E. Wassermann, "Variation in the response to transcranial magnetic brain stimulation in the general population," *Clin. Neurophysiol.*, vol. 113, no. 7, pp. 1165–1171, Jul. 2002.

[38] A. Mir-Moghtadaei et al., "Concordance between BeamF3 and MRI-neuronavigated target sites for repetitive transcranial magnetic stimulation of the left dorsolateral prefrontal cortex," (in English), *Brain Stimulation*, vol. 8, no. 5, pp. 965–973, Sep. 2015.

[39] L. M. Koponen, S. M. Goetz, and A. V. Peterchev, "Double-containment coil with enhanced winding mounting for transcranial magnetic stimulation with reduced acoustic noise," *IEEE Trans. Biomed. Eng.*, vol. 68, no. 7, pp. 2233–2240, Jul. 2021.

[40] N. C. Rogasch et al., "Analysing concurrent transcranial magnetic stimulation and electroencephalographic data: A review and introduction to the open-source TESA software," (in English), *NeuroImage*, vol. 147, pp. 934–951, Feb. 2017.

[41] Y. Wang et al., "Application of fast perturbational complexity index to the diagnosis and prognosis for disorders of consciousness," (in English), *IEEE Trans. Neural Syst. Rehabil. Eng.*, vol. 30, pp. 509–518, 2022.

[42] Y. Wang et al., "Comparison of synchrosqueezing transform to alternative methods for time-frequency analysis of TMS-evoked EEG oscillations," (in English), *Biomed. Signal Process. Control*, vol. 70, Sep. 2021, Art. no. 102975.

[43] F. Tadel, S. Baillet, J. C. Mosher, D. Pantazis, and R. M. Leahy, "Brainstorm: A user-friendly application for MEG/EEG analysis," (in English), *Comput. Intell. Neurosci.*, vol. 2011, pp. 1–13, May 2011.

[44] A. Gramfort, T. Papadopoulo, E. Olivi, and M. Clerc, "OpenMEEG: Opensource software for quasistatic bioelectromagnetics," (in English), *Biomed. Eng. OnLine*, vol. 9, no. 1, p. 45, Sep. 2010.

[45] M. I. Dutt and W. Saadeh, "Monitoring level of hypnosis using stationary wavelet transform and singular value decomposition entropy with feedforward neural network," *IEEE Trans. Neural Syst. Rehabil. Eng.*, vol. 31, pp. 1963–1973, 2023.

[46] E. Maris and R. Oostenveld, "Nonparametric statistical testing of EEG- and MEG-data," *J. Neurosci. Methods*, vol. 164, no. 1, pp. 177–190, Aug. 2007.

[47] P. Lioumis, D. Kič ić, P. Savolainen, J. P. Mäkelä, and S. Kähkönen, "Reproducibility of TMS—Evoked EEG responses," *Hum. Brain Mapping*, vol. 30, no. 4, pp. 1387–1396, Apr. 2009.

[48] I. Premoli et al., "TMS-EEG signatures of GABAergic neurotransmission in the human cortex," (in English), *J. Neurosci.*, vol. 34, no. 16, pp. 5603–5612, Apr. 2014.

[49] Y. Noda et al., "Resting-state EEG gamma power and theta–gamma coupling enhancement following high-frequency left dorsolateral prefrontal rTMS in patients with depression," *Clin. Neurophysiol.*, vol. 128, no. 3, pp. 424–432, Mar. 2017.

[50] P. Dhami et al., "Prefrontal cortical reactivity and connectivity markers distinguish youth depression from healthy youth," (in English), *Cerebral Cortex*, vol. 30, no. 7, pp. 3884–3894, Jun. 2020.

[51] P. Dhami et al., "Neurophysiological markers of response to theta burst stimulation in youth depression," *Depression Anxiety*, vol. 38, no. 2, pp. 172–184, Feb. 2021.

[52] N. Eshel et al., "Global connectivity and local excitability changes underlie antidepressant effects of repetitive transcranial magnetic stimulation," *Neuropsychopharmacology*, vol. 45, no. 6, pp. 1018–1025, May 2020.

[53] M. S. Hoseini, N. C. Wright, J. Xia, W. Clawson, W. Shew, and R. Wessel, "Dynamics and sources of response variability and its coordination in visual cortex," (in English), *Vis. Neurosci.*, vol. 36, p. E012, Dec. 2019.

[54] S. Isomura et al., "Differentiation between major depressive disorder and bipolar disorder by auditory steady-state responses," *J. Affect. Disorders*, vol. 190, pp. 800–806, Jan. 2016.

[55] G. Pfurtscheller and F. H. Lopes da Silva, "Event-related EEG/MEG synchronization and desynchronization: Basic principles," *Clin. Neurophysiol.*, vol. 110, no. 11, pp. 1842–1857, Nov. 1999.

[56] G. Thut and A. Pascual-Leone, "A review of combined TMS-EEG studies to characterize lasting effects of repetitive TMS and assess their usefulness in cognitive and clinical neuroscience," *Brain Topography*, vol. 22, no. 4, pp. 219–232, Jan. 2010.

[57] P. J. Fitzgerald and B. O. Watson, "Gamma oscillations as a biomarker for major depression: An emerging topic," *Transl. Psychiatry*, vol. 8, no. 1, p. 177, Sep. 2018.

[58] S. J. Haghighi, M. Komeili, D. Hatzinakos, and H. E. Beheiry, "40-hz ASSR for measuring depth of anaesthesia during induction phase," (in English), *IEEE J. Biomed. Health Inf.*, vol. 22, no. 6, pp. 1871–1882, Nov. 2018.

[59] C. Spironelli et al., "Evidence of language-related left hypofrontality in major depression: An EEG beta band study," (in English), *Sci. Rep.*, vol. 10, no. 1, p. 8166, May 2020.

[60] Y. He, W. Guo, Z. Ren, J. Liang, S. Liu, and D. Ming, "Gamma auditory steady-state response as a promising electrophysiological biomarker for depression: An in vivo study with chronic unpredictable mild stress (CUS)-induced rats," *Cerebral Cortex*, vol. 33, no. 12, pp. 7741–7753, Jun. 2023.

[61] D. A. Pizzagalli, L. A. Peccoralo, R. J. Davidson, and J. D. Cohen, "Resting anterior cingulate activity and abnormal responses to errors in subjects with elevated depressive symptoms: A 128-channel EEG study," *Hum. Brain Mapping*, vol. 27, no. 3, pp. 185–201, Mar. 2006.

[62] M. C. Pellicciari, V. Ponzo, C. Caltagirone, and G. Koch, "Restored asymmetry of prefrontal cortical oscillatory activity after bilateral theta burst stimulation treatment in a patient with major depressive disorder: A TMS-EEG study," *Brain Stimulation*, vol. 10, no. 1, pp. 147–149, Jan. 2017.

[63] G. Sanacora et al., "Subtype-specific alterations of γ-aminobutyric acid and glutamate in patients with major depression," (in English), *Arch. Gen. Psychiatry*, vol. 61, no. 7, pp. 705–713, Jul. 2004.

[64] A. K. Engel and P. Fries, "Beta-band oscillations—Signalling the status quo?" *Current Opinion Neurobiol.*, vol. 20, no. 2, pp. 156–165, Apr. 2010.