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Event-Related Phase-Amplitude Coupling During Working Memory of Musical Chords

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ABSTRACT Phase-amplitude coupling (PAC) is a well-established concept for evaluating the strength of memory coding within brain regions, and has been shown to possess the characteristic of presenting memory mechanisms. It has been demonstrated that oscillations of theta and gamma brain waves can represent the neural coding structure of memory retrieval. However, most previous studies have presented PAC-related memory mechanisms with visual modalities, and little is known about the influence of auditory stimuli. In this study, 18 participants were recruited and 36-channels electroencephalography (EEG) signals were recorded while they were performing an *n*-back auditory working memory task. There were three experimental conditions with different levels of working memory load. Event-related phase-amplitude coupling (ERPAC) with the advantage of better temporal resolution was used to evaluate the coupling phenomenon from the reconstructed dipole brain sources. We primarily focused on independent components from the frontal and parietal regions, which were reported to be related to memory mechanisms. The results suggest that significant ERPAC was observed in both the frontal and parietal regions. In addition to the coupling between theta (4-7 Hz) and low gamma (30-40 Hz) frequency bands, pronounced high beta oscillations (20-30 Hz) were also observed to be modulated by the phases of theta oscillations. These findings suggest the existence of phase-amplitude coupling in the neocortex during auditory working memory, and provide a highly resolved timeline to evaluate brain dynamics. In addition, the ERPAC results also support the involvement of theta-gamma and theta-beta neural coding mechanisms in cognitive and memory tasks. Collectively, these findings demonstrate the existence of ERPAC within the frontal and parietal regions during an auditory working memory task using complex chords as stimuli, and prompt the use of complex stimuli in studies that are closer to the real-life applications of cognitive evaluations, mental treatments, and brain-computer interfaces.

INDEX TERMS EEG, event-related phase-amplitude coupling, working memory, musical chords.

I. INTRODUCTION

Exploring the mechanism of memory coding with neural oscillations has generated great interest in neuroscience. In the past decade, the coupling mechanisms of working memory (WM) have received increased attention. Modalities such as local field potential (LFP), electroencephalography (EEG), and magnetoencephalography (MEG) have been used to investigate the mechanisms of WM and the functional connection between involved brain regions by cross-frequency coupling (CFC) [1]. Among all the modalities, functional magnetic resonance imaging (fMRI) has a better spatial resolution. However, EEG and LFP have a higher temporal resolution and can evaluate the timing of memory retrieval and the mechanisms of couplings. Previous studies have demonstrated that 7-11 gamma waves are coupled within each theta wave in a memory coding scheme, and the number of gamma waves was proposed to represent the number of memorable items [2]. Distinct methods have been proposed to investigate the strength of coupling during

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cognitive tasks [1] such as phase-phase coupling [3], [4], amplitude-amplitude coupling [5], and phase-amplitude coupling (PAC) [6], [7]. As most past cognitive CFC studies underlined visual stimulation, the functions of auditory WM are still unknown. Additionally, most previous auditory WM studies used relatively simple sound tones as stimuli [8], [9]. The utilization of experimental paradigms closer to real-life situations has become a growing trend in cognitive neuroscience, since complex behaviors are believed to be expressed only within real-life natural contexts [10]. Recent auditory studies have started to use naturalistic stimuli to examine the integration of complex auditory sequences [11]–[15]. Therefore, we primarily focused on the PAC phenomenon during a musical WM task with complex chords as stimuli, as they were closer to real-life situations.

A. NEURAL OSCILLATION, PAC, AND WORKING MEMORY

Neural oscillations of various frequency ranges are associated with cognitive processes. For instance, Jensen and colleagues revealed that the activities of theta bands are related to memory load and maintenance [16]. Additionally, gamma oscillations were found to correlate with memory load and the maintenance of multiple WM information [17]. To study the activation and functional connectivity between brain regions, three crucial factors of neural oscillations, phase, amplitude, and frequency, were evaluated. The phases of neural oscillations reflect the timing of firing [18], the amplitudes represent the strength of brain activations [19], and the frequency represents the corresponding physiological meanings [20]. Several studies have indicated the involvement of gamma band oscillations in numerous cognitive tasks, including perception, attention, memory, and information processing. [4], [21]–[23]. Activation of theta oscillations has been suggested to be involved in WM capacity [24] and memory load [16]. Additionally, beta frequency bands have been linked to encoding, retrieval, and maintenance of memory-related stimuli [25].

The PAC between neuronal oscillations engaged in cognitive tasks has received more attention in the past decade. In particular, PAC is the most reported method for computing neural oscillations and encoding schemes when evaluating the mechanisms of WM. PAC can be observed as the modulation of the amplitudes from high-frequency brain waves by the low-frequency phases and thus plays an essential role in memory stabilization [26]. Moreover, PAC has been associated with memory coding, storage, and retrieval of information [3], [4], [6]. Several studies have suggested the involvement of PAC in the hippocampus [5], [6], [14], [27], frontal [1], [7], [22], [26], [28]–[35], parietal [7], [16], [17], [36], [37], and temporal regions [4], [7], [21], [38] during WM tasks in humans. Previous cognitive studies in rats and humans have observed the engagement of PAC in different frequency bands of phases including alpha, delta, and theta bands as well as frequency bands of amplitudes including gamma, beta, and alpha [3]–[7], [14], [16], [19], [22]–[26], [28], [31]–[34], [36], [37], [39]–[46]. The coupling between theta and gamma oscillations is one of the most well-known coding schemes, and has been suggested to be an important mechanism during memory processes [1]–[3], [5], [6], [14], [18], [19], [22], [27], [28], [34], [35], [41], [44], [47], [48], including memory maintenance [29], [48], formation [30], learning [7], [27], and attention [26]. Moreover, coupling between theta and beta frequency ranges is associated with cognitive tasks such as decision-making [32], memory maintenance [33], and visual attention tasks [34]. The couplings of theta-alpha [24], [36], delta-beta [14], [16], [37], delta-gamma [5], [19], [34], [44], alpha-beta [18], [34], and alpha-gamma [14], [20], [28], [32], [46] have been proposed in previous studies involving memory tasks. Additionally, in the past few years, PAC has also been associated with clinical conditions and diseases related to cognitive impairment [49]–[55].

However, most previous cognitive studies have utilized traditional PAC methods to evaluate the strength of coupling. In contrast, event-related phase-amplitude coupling (ERPAC) is a novel approach proposed by Voytek to evaluate the strength of coupling with better temporal resolution [36]. Therefore, the precise timing of neural firing and memory retrieval can be quantitatively evaluated with ERPAC by analyzing the strength of couplings between the phases and amplitudes of neural oscillations.

B. NEUROMUSIC AND AUDITORY MEMORY

An increasing number of recent studies have explored the relationship between auditory WM and brain mechanisms [8], [14], [20], [22], [56], occasionally in comparison to visual WM [6], [57]. Neuroscience studies on neuromusic-related topics have examined various factors, including memory and learning [8], musical emotion perception [58], recognition of musical items [37], and musical training [56], [59]. A neuromusic study by Pallesen and colleagues on non-musicians and musicians during musical WM found that musicians perform better during enhanced auditory WM tasks. It is believed that this superior memory performance in musicians is the result of long-term musical training [56]. Additionally, fMRI results have confirmed a positive correlation between neural activity and WM load [56]. Previous fMRI studies have reported that different chords and pitches result in activation and connectivity between brain regions linked through the function of language, perception, emotion, memory, and cognition [28], [60]. PET studies have also suggested a correlation between pitch memory and the frontal, temporal, and parietal regions [3].

Neuroimaging studies have proposed the engagement of the frontal and parietal regions in auditory WM and language processes [33]. Previous MEG studies have identified brain activation in the superior temporal gyrus and inferior frontal gyrus during short-term WM tasks [61]. Additionally, fMRI studies have established the engagement of the parietal, frontal, prefrontal, and temporal brain regions during auditory stimuli, such as sound recognition [3], [62] and auditory WM tasks [62], [63]. EEG research on auditory WM has also suggested the involvement of the frontal and parietal regions [14]. Moreover, several results of auditory stimuli were reported to be consistent with those previously described for visual stimuli [19], [28], [32], [34], [64]. Although it is believed that more complex tasks involving auditory stimuli can be built up from basic auditory memory processes, no consensus has yet been reached on musical WM in neuroscience.

C. PAC METHODS IN EVALUATING THE MECHANISMS OF WORKING MEMORY

Numerous studies have been conducted on auditory WM tasks. However, most of these studies utilized fMRI as a recording modality. Although fMRI provides better spatial resolution to localize brain activity in distinct regions, it does not offer sufficient temporal resolution. In comparison, other neurophysiological techniques, such as EEG and MEG, provide better temporal resolution during task execution. Several methods have been reported for evaluating the traditional PAC of EEG or MEG signals under different experimental conditions. In 2004, Bruns and Eckhorn calculated the strength of coupling by computing the correlation between high-frequency envelopes and low-frequency phases [33]. In 2005, Lakatos and colleagues proposed the heights ratio to evaluate the strength of coupling by calculating the difference between the maximum and minimum amplitudes at the same phase [44]. Canolty *et al.* observed the mean vector length by computing the distance and vector angles from the center, thereby obtaining instantaneous amplitudes and phases in a complex plane [35]. Later in 2008, Cohen claimed that the power spectral density of instantaneous amplitudes has the advantage of synchronously measuring the PAC with fixed amplitudes across multiple phases [20]. In 2008, Penny introduced a new approach to enhance the accuracy of calculating coupling by adding cosine or sine when computing the correlation [32]. In 2010, the modulation index was used to compute the strength of coupling by determining the Kullback-Leibler distance between the normalized amplitudes and uniform distributions [27]. However, PAC has several deficiencies, such as a lower temporal resolution [36]. Therefore, a new approach, ERPAC, which underlines the function of event-related coupling, was proposed by Voytek *et al.* [36]. ERPAC can compensate for the deficiencies of traditional PAC, as it possesses higher temporal resolution.

In this study, ERPAC was used to evaluate the strength of the coupling of EEG oscillations during a musical WM task. By clarifying the inter-regional brain activities, the aim of this study was to address the following hypotheses: (1) the brain regions involved in auditory WM; (2) the existence of ERPAC in the neocortex as an indicator of neural coding during auditory WM processes; and (3) the temporal dynamics of ERPAC after the onset of auditory memory retrieval. EEG signals were recorded when participants performed an *n*-back auditory WM task with musical chords as stimuli. We focused on the memory mechanisms aroused by musical stimulation and expected that coupling can be observed during auditory memory encoding and retrieval. Furthermore, with the advantage of ERPAC, the precise timing of memory retrieval can be detected. Based on previous findings on auditory WM, we proposed a novel approach using complex chord stimuli and ERPAC to study EEG dynamics and neural coding during working memory processes.

II. MATERIALS AND METHODS

This topic addresses the existence of ERPAC among brain sources during auditory memory processing. Thus, the following procedures are conceived to reveal the source patterns and strength of coupling. First, we described the participants recruited in this experiment and the experimental design of the complex auditory WM task. Second, the details of EEG recording and data preprocessing are illustrated, followed by an advanced source reconstruction method of independent component analysis (ICA). The clustered independent components (ICs) were then further analyzed by an event-related PAC method to evaluate the strength of coupling, which is correlated to the involvement of neural coding. Finally, group analysis and statistical methods are described.

A. PARTICIPANTS

Twenty-six right-handed subjects without any brain disorders, injuries, or brain implants were recruited for this study. The participants were instructed to fill in two forms: a consent form and a rating scale of music familiarity prior to the experiment. After being informed of the procedure and the risks of human subject research, all participants were asked to sign a consent form in accordance with the requirements of the Human Subject Research Ethics Committee of the Institutional Review Board [65] at Fu Jen Catholic University, Taiwan. The rating scale of music familiarity was used to help acquire a basic understanding of the participants' musical knowledge. None of the participants were experts. The participants were also instructed to report their preferences in music types, such as popular, rock, jazz, country, or classical music. The experiment was performed in an isolated environment where the vision and hearing of the participants were blocked from the outside environment to prevent any potential influencing factors. The performance of participants with task accuracy less than 60% was excluded from further analysis, as well as those who lost dipole sources in their data, especially in the frontal and parietal regions.

B. EXPERIMENTAL PARADIGM

An *n*-back auditory WM task was used in this study which was a modified version of the paradigm proposed by Pallesen and colleagues [56]. The experimental design is illustrated in Figure 1, and it took 40 min to perform the entire experiment. There were a total of 36 blocks in one experiment divided into two sessions (Figure 1A). The length of each session was 19 minutes, with a 2-minute break between

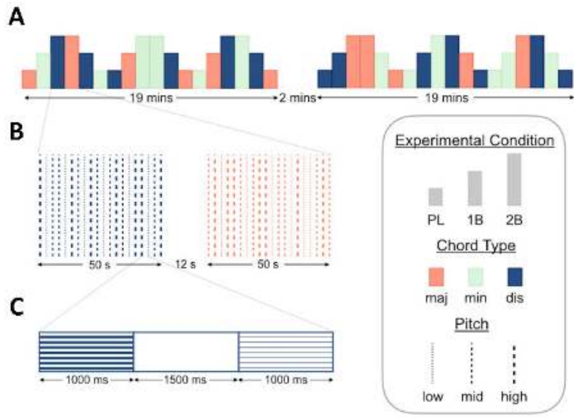

FIGURE 1. Experimental design of the *n*-back auditory working memory task. (A) Three experimental conditions including passive listening (PL), 1B and 2B were distributed in 36 blocks and divided into 2 sessions. (B) Each block consisted of 20 trials with a 12-second rest between blocks. Each trial of 50-second length with different pitch types was marked by 3 kinds of line styles. (C) The duration of each musical chord was 1000-ms with a 1500-ms rest between each stimulus.

sessions. Each block contained 20 trials of 50-second length with a 12-second rest between the blocks (Figure 1B). There were three experimental conditions, including passive listening (PL), 1-back (1B), and 2-back (2B), with three categories of sound combinations: major, minor, and dissonant chords. All chord types were randomly distributed throughout the trials. The participants were requested to press a button after the onset of each stimulus, depending on the experimental conditions. In the PL condition, participants were instructed to press the button when they heard a chord. In the 1B condition, the participants pressed the button if the chord matched the previous one; otherwise, they pressed another button for control. In the 2B condition, the participants pressed a button if the chord corresponded to one of the latest two chords. Under each experimental condition, the participants underwent memory retrieval with different memory loads.

The three kinds of musical chords were programmed and recorded using the Overture 4.0 software (GenieSoft, USA) at the same volume and duration of 1000 ms, as shown in Figure 1C. The major chords contained A, C#, E, A, and C#, with four pitches starting at A3, C#4, E4, and A4. The minor chords were composed of A, C, E, A, and C, with four pitches starting at A3, C4, E4, and A4, which can be viewed as imperfect consonance in music theory. The dissonant chords comprised of A, Bb, G, Ab, and C, with four pitches starting at A3, Bb3, G4, and Ab4. The influence of different types of chords and pitches was not addressed in this study.

All the stimuli were presented using Psychophysics Toolbox Version 3 [66]–[68] and MATLAB R2013b (The MathWorks, Inc., Natick, Mass, USA) software to control the duration and sequence of musical chords, and to send trigger signals during EEG recording. During the experiment, participants wore ATH-MSR7 headphones (Audio-Technica Taiwan Co., Ltd., Taiwan) to listen to the auditory stimuli. The sound stimuli were displayed with the volume adjusted by the participants to a comfortable level. The participants sat in a soundproof chamber, and EEG signals were recorded throughout the experiment. The participants were asked to keep their eyes open, and all instructions were delivered by an LCD monitor.

C. EEG RECORDING AND DATA PREPROCESSING

EEG signals were recorded with a 36-channel electrode cap (Nuamp, NeuroScan, Inc., Charlotte, NC), which conformed to the international standard of a 10-20 electrode placement system [69]. The channels used in this study were O2, Oz, O1, T6, P4, Pz, P3, T5, TP8, CP4, CPZ, CP3, TP7, T4, C4, Cz, C3, T3, FT8, FC4, FCZ, FC3, FT7, F8, F4, Fz, F3, F7, VOEL, VEOU, Fp2, Fp1, HEOR, HEOL, A1, and A2. The impedance of each electrode was maintained under 5 kΩ to enhance signal quality. Software Scan4.5 (Advanced Medical Equipment Ltd., UK) was used to record and receive the EEG signals. The average signals from electrodes A1 and A2 were used as a reference for the EEG signals. The signals were digitized at a sampling rate of 500 Hz and filtered with a frequency band between 0.1 and 50 Hz. The EEGLAB toolbox [70] in MATLAB was used to analyze the brain signals collected by the electrode cap. For further analysis, the continuous EEG data were segmented into epochs of −1500 to 2000 ms following stimulus onset. Trials with incorrect answers and noisy EEG signals with absolute values greater than 100 μV were removed. Artifacts including eye movement, blinking, muscle tension, and heart rate signals, were estimated and removed using an ICA procedure based on the power spectra and the reference signals of eye movement from VOEL, VEOU, HEOR, and HEOL [71].

D. SOURCE RECONSTRUCTION USING INDEPENDENT COMPONENT ANALYSIS

The DIPEFIT2 plug-in [72] in the EEGLAB toolbox [70] was used to locate the source of the signals from the sensor to source level. ICs were applied to DIPFIT2 to obtain the location of source dipoles with a standard boundary element head model (BEM) used for head modeling [73]. Furthermore, back projection was used to average multiple similar ICs into a new averaged-IC [70]. Group analysis was then performed by registering the individual data to a brain template provided by the Brain Imaging Center, Montreal Neurological Institute (MNI) [74] with the locations co-registered and mapped to a 3D brain image. ICs with equivalent dipoles located outside the modeled brain volume and residual variance larger than 20% [75] were excluded. According to the spectral characteristics and dipole locations, the ICs from all participants were clustered into the frontal and parietal regions. In this study, the dipole sources from these two regions were identified for further analysis. However, previous EEG studies using ICA and dipole fitting [76]–[78] mentioned that some participants may lack one or more ICs after component selection during clustering analysis. Therefore, the ICs of the loose dipole fitting were excluded from further analysis. The EEG source components from all participants were finally clustered into the frontal and parietal regions for further analysis of ERPAC.

E. EVENT-RELATED PHASE-AMPLITUDE COUPLING

ERPAC [36] has been proposed to overcome the problems encountered in traditional PAC, including the lack of temporal resolution and the precondition of defining the time window prior to the experiment. With the modification of the correlation coefficient method, ERPAC possesses the merits of both temporal and spatial resolutions. Instead of calculating the strength of coupling from each trial, ERPAC measures coupling over trials at each time point and thus provides better temporal resolution.

ERPAC was used to measure the strength of the coupling across the trials in this study. EEG signals from ICs located in the frontal and parietal regions were used to compute the strength of the ERPAC. To measure the level of ERPAC, the raw EEG signals (X_{raw}) from an IC were band-pass filtered into two different frequency bands, $X_p(t)$ and $X_q(t)$ at time t. Hilbert transform (HT) was then used to obtain the instantaneous phase, $\phi_p(t)$, from the lower frequency band p and the envelopes of amplitudes, $A_q(t)$, from the higher frequency band q. The definition of HT is:

$$HT(X_{raw}(t)) = \frac{1}{\pi} P.V. \int_{-\infty}^{\infty} \frac{X_{raw}(\tau)}{t-\tau} d\tau. \tag{1.1}$$

HT can be viewed as the convolution of $X_{raw}(t)$ and $h(t) = 1/\pi t$, and P.V. expresses the Cauchy principal value. The analytic form of the time series can be obtained by HT as

$$Z_p(t) = X_p(t) + jHT(X_p(t)) = A_p(t)\, e^{i\phi_p(t)}. \tag{1.2}$$

Envelopes of amplitudes are defined as,

$$A_p(t) = \sqrt{X_p^2(t) + HT^2(X_p(t))}. \tag{1.3}$$

The instantaneous phase was therefore denoted as,

$$\phi_p(t) = arctan\left(\frac{HT(X_p(t))}{X_p(t)}\right), \tag{1.4}$$

with $\phi_p(t) \in [-\pi, \pi)$. The $\phi_p(t)$ and $A_q(t)$ of $X_{raw}(t)$ can be computed using (1.3) and (1.4), respectively.

The correlation coefficient was then used to calculate the PAC strength. However, the correlation coefficient method failed to detect coupling at cycles 1/4 and 3/4, for instance, $cos((1/4)2\pi) = 0$, so the values might have been missed and the correlation result would be influenced by some specific situations. Hence, the correlation coefficient was modified to a circular-linear correlation to obtain the ERPAC, which can be denoted as [36], [79], [80],

$$ERPAC(p, q, t) = \sqrt{\frac{r_{ca}^2 + r_{sa}^2 - 2r_{ca}r_{sa}r_{cs}}{1 - r_{cs}^2}}, \tag{1.5}$$

where

$$r_{ca} = Corr(cos(\phi_p(t)), A_q(t)), \tag{1.6}$$

$$r_{sa} = Corr(sin(\phi_p(t)), A_q(t)), \quad \text{and} \tag{1.7}$$

$$r_{cs} = Corr(sin(\phi_p(t)), cos(\phi_p(t))). \tag{1.8}$$

In this study, ERPAC was calculated in the frequency range of the EEG phases from 1 to 15 Hz and amplitudes from 12 to 50 Hz. In this specific frequency range, ERPAC was calculated for each frequency bin across trials and at each time point. A narrow-band convolution filter was used to filter the EEG signals in the frequency range with a bandwidth of 2 Hz for each frequency bin. After calculating the ERPAC at each frequency bin and time point, the ERPACs were averaged over every 100 ms.

According to previous studies [35], [36], [47], [81], the significance of PAC is usually performed using a surrogate test with random-permuted surrogate distributions. Therefore, we evaluated the significance of each experimental condition by comparing the ERPAC values with the surrogated EEG data. The surrogated data were obtained by fixing the phase values, but randomly permuting the amplitude values 50 times. The significance of the averaged ERPACs was calculated over each time interval, and then the frequency bands with more than 80% of pronounced ERPACs were selected as the frequency bands of interest (FOI). After the FOIs were selected, we re-calculated the ERPAC of each subject over the desired frequency ranges and compared them with the surrogate distribution by randomly permuting the data 1000 times. The results of pronounced ERPAC were compared to the baseline and presented by Brainnet (NeuroImaging Tools & Resources Collaboratory, NITRC) [82].

F. STATISTICAL ANALYSIS

Statistical analysis was conducted using MATLAB R2013b. The surrogate procedure described in the previous section was used to measure the p-values by comparing the ERPACs with the surrogate distribution under each experimental condition and each IC from the frontal and parietal regions. ERPACs with p-values < 0.05, were defined as significant. Group analysis of ERPAC was performed by averaging the ERPAC data from each subject and comparing it with the surrogate distribution.

III. RESULT

A. BEHAVIOR RESULT

EEG signals from 26 participants were collected during the n-back auditory WM task. Data from eight participants were excluded due to low task accuracy (i.e., <60%) or the absence of ICs from the frontal or parietal regions. The age distribution of the remaining 18 participants (11 females) was between 18 and 42 years with an average age of 23.72 years (SD = 5.15). The average task accuracy of the data sample was 75% (SD = 7.57). The average response time of all participants was 1279 ms in the PL condition, 1511 ms in 1B, and 1517 ms in 2B. Most participants reported the habit of listening to music for two hours per day on average. None of them had expertise in music, although many had access to instruments for years under either formal lessons or self-study.

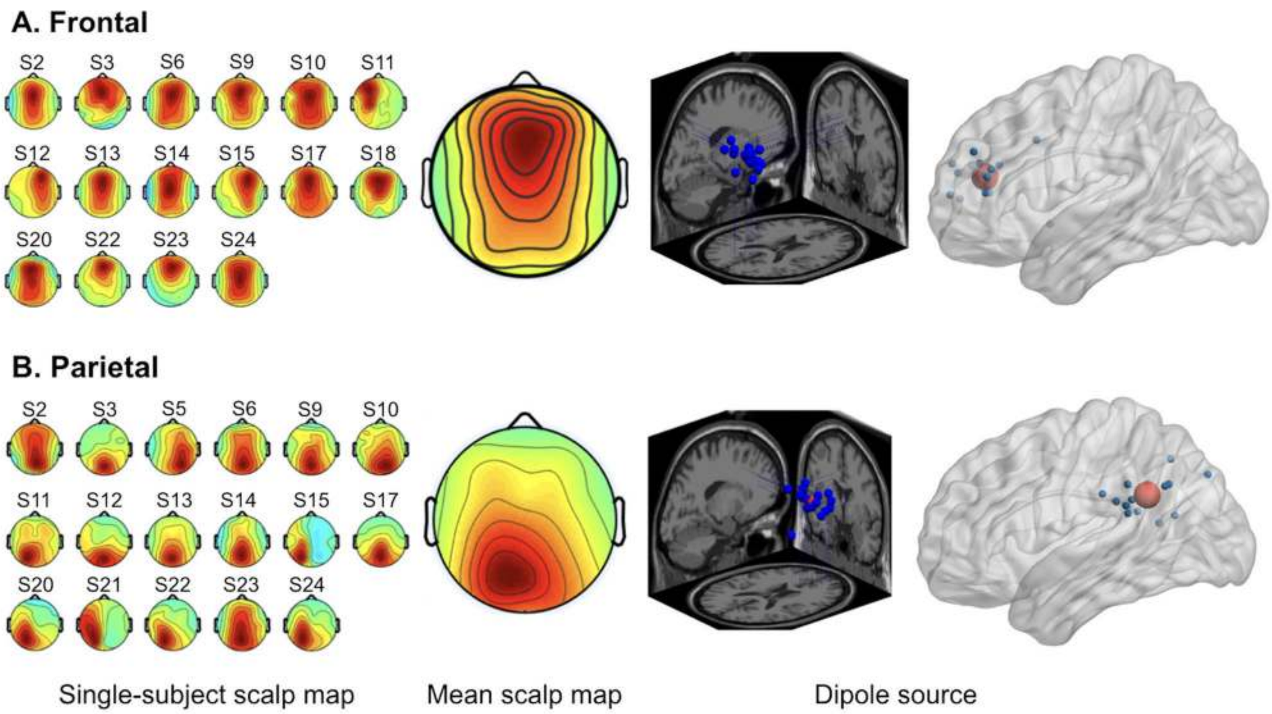

FIGURE 2. The individual scalp maps, mean scalp maps, and dipole location of the frontal (A) and parietal (B) regions. The single-subject scalp map illustrates the independent components of each subject used in analysis, and mean scalp map is the averaged ICs of all subjects. The dipole locations of each subject were shown in blue points and the averaged dipoles were shown in red.

TABLE 1. Independent component clusters and the centroid of their source distributions.

Brain region	# of components	Mean residual variance (%)	Talairach coordinates		
			x	y	z
Frontal	16	11.88	2	38	16
Parietal	17	8.6	-12	-50	30

B. INDEPENDENT COMPONENT CLUSTERS

Two IC clusters were obtained from 18 subjects in this study. Individual scalp maps, mean scalp maps, and dipole locations of the ICs from the frontal and parietal regions are presented in the left, middle, and right panels of Figure 2, respectively. Individual scalp maps of each subject were obtained from component maps in 2-D using the EEGLAB toolbox after ICA. The mean scalp map was computed after averaging the individual scalp maps of all the ICs. The precise location of the source signal after dipole fitting is illustrated in the right two panels of Figure 2. Data samples without frontal or parietal IC were excluded. Sixteen ICs from the frontal region and 17 ICs from the parietal region were obtained from the participants. The dipole information of the ICs clustered into the frontal and parietal brain regions are listed in Table 1, including the number of contributing participants, mean residual variance, and the Talairach coordinates of the central dipoles. According to the Talairach coordinates, the dipoles from the two regions were source localized around the anterior cingulate cortex (ACC, Brodmann area 32) in the frontal region and the precuneus (Brodmann area 31) in the parietal region.

C. EVENT-RELATED PHASE-AMPLITUDE COUPLING

An example of ERPAC in the parietal region from a single subject during the 2B task between 200 and 300 ms after the stimulus onset is illustrated in Figure 3. ERPAC was calculated across trials at each frequency bin ranging from a phase of 1-15 Hz and an amplitude of 12-50 Hz. The upper panel (Figure 3A) shows the strength of coupling, and the lower panel (Figure 3B) shows the significance level of coupling after t-test and random permutation for 50 times. As shown in Figure 3, pronounced theta-beta and theta-gamma coupling can be observed with an ERPAC significance level (p) lower than 0.03.

Figure 4 and 5 illustrate the group significance of ERPAC in the frontal (Figure 4) and parietal (Figure 5) regions after computing the t-test and surrogation 1000 times over the group ERPAC. The frequency range of the phase was 4 to 7 Hz (theta) and 8 to 12 Hz (alpha), and the frequency bands of amplitudes ranged from 24 to 27 Hz, 20 to 30 Hz (high beta), and 30 to 40 Hz (low gamma).

As demonstrated in Figure 4, pronounced ERPAC with theta-low gamma coupling was observed between 700 and 900 ms after the onset of stimuli in the frontal region during the 1B task. In the PL task, significance existed later from 900 to 1000 ms, close to the end of the stimuli. However,

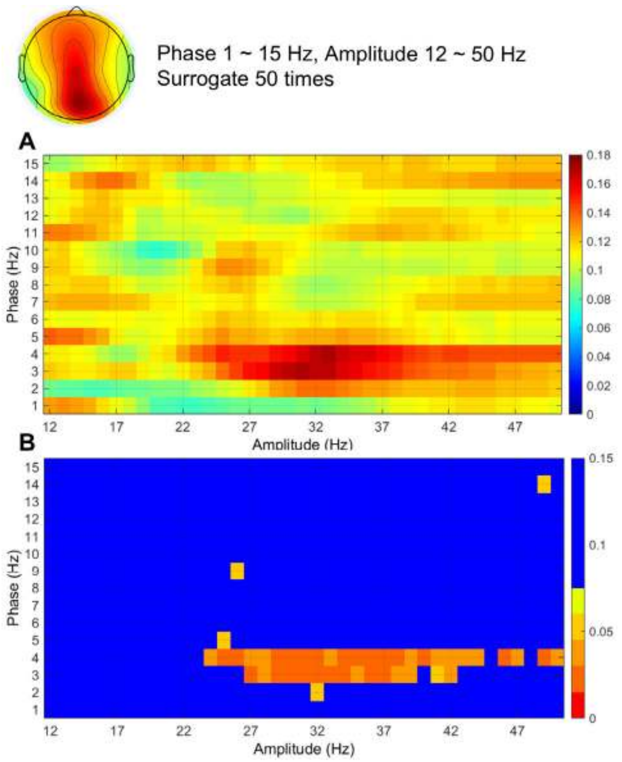

FIGURE 3. **Example of ERPAC in the parietal region from a single subject. ERPACs were computed at every frequency bin in the frequency ranges from an example of 2B condition with 152 trials. The strength of ERPAC is presented in A, and the significant levels (*p* values) of ERPACs are illustrated in B. Observing from A and B, pronounced theta-beta and theta-gamma coupling can be detected.**

no significant theta-low gamma coupling was found in the 2B task. In addition to theta-low gamma coupling, pronounced high beta amplitudes were modulated by theta phases in the frontal regions during musical WM tasks. Pronounced ERPAC with theta to high beta coupling can be observed between 100 and 200 ms after the onset of stimuli in the frontal region (Figure 4B) during the 2B task. Later, between 700 and 800 ms, coupling was found during the 1B task. In the PL task, significance existed later in 900 ms close to the end of the stimuli.

The significance of theta and alpha to high beta ERPACs in the parietal regions compared with 1000 times surrogation is shown in Figure 5. Pronounced ERPAC with theta to high beta coupling can be observed from 0 to 100 and 400 to 500 ms after the onset of stimuli in the parietal region (Figure 5A) during the 2B task. In the 1B task, significant theta-high beta ERPACs existed from 800 to 900 ms. No significant theta to high beta coupling was found in the PL task. Pronounced high beta (24-27 Hz) amplitudes were also found to be modulated by alpha phases in the parietal regions during musical WM tasks. In the 2B task, significant theta and high beta ERPACs were found from 100 to 200 ms, as shown in Figure 5B. Pronounced ERPAC can also be observed from 800 to the end of the stimuli during 1B task. In the PL task, significance existed between 600 and 700 ms.

Our results demonstrate the existence of ERPACs in the frontal and parietal regions during auditory WM tasks. Pronounced ERPACs indicate that the amplitudes of the gamma and beta frequency bands are modulated by the phases of theta and alpha frequency bands. Significance was found between 100 and 500 ms in the 2B condition, 700 and 1000 ms in the 1B condition, and after 600 ms in the PL condition. Pronounced ERPAC is observed earlier before 500 ms at a higher WM load (2B) than in passive listening, and is proposed to be involved in auditory memory retrieval processes.

IV. DISCUSSION

The aim of this study was to discover neural oscillations and coding correlated with musical WM load. Numerous studies have implicated the important phenomenon of high-frequency EEG amplitudes modulated by low-frequency phases across various frequency bands in cognitive processes. Particularly, PAC has been proposed to play a crucial role in memory procedures during the encoding, maintenance, and retrieval of information [61], [62], [83]. Fell and colleagues characterized the relationship between PAC and memory maintenance, retrieval, and recall, especially between theta and gamma frequency bands [84]. Lega and colleagues have reported that theta-gamma coupling is engaged in the formation of novel memories and the prediction of successful memory encoding [62]. Axmacher and colleagues have also indicated the involvement of theta and beta/gamma coupling during WM maintenance [63]. Therefore, PAC is essential in cognitive tasks such as auditory [14], [57] and visual short-term memory [22], [28], [33], [41], [42], [57], [62], [85], [86], attention [34], [36], [56], perception [14], [19], [69], [71], learning [18], [36], [47], [75], [87], decision making [36], and other memory-related tasks [6], [46], [79], [82]. Additionally, it is suggested that PAC is involved in memory processes through various mechanisms including WM performance [4], [22], [47], memory maintenance [14], [33], [43], [47], recall [18], [22], [43], storage [18], [22], [43], encoding [28], [33], retrieval [14], [34], formation [28], [42] and cognitive control [4], [34], [88].

In accordance with the previous EEG and MEG findings [3], [4], [14], [16], [18], [22], [40], [42], [87], our ICA and dipole fitting results showed the existence of neural oscillations in similar regions including the frontal and parietal areas, which have been reported to be engaged in cognitive tasks [16], [18]–[20], [22], [24], [31]–[33], [35], [41], [89]. This study reported three main findings. First, elevated couplings between low-frequency phases and high-frequency amplitudes were observed during memory retrieval and found earlier after the onset of stimuli as the WM load increased. Second, significant neural oscillations can be found during the first 1000 ms after the onset of stimuli in the auditory WM task compared to the baseline. Third, the results of ERPAC revealed the existence of pronounced theta-beta, theta-gamma, and alpha-beta couplings during musical stimuli. These findings suggest that ERPACs in the frontal and

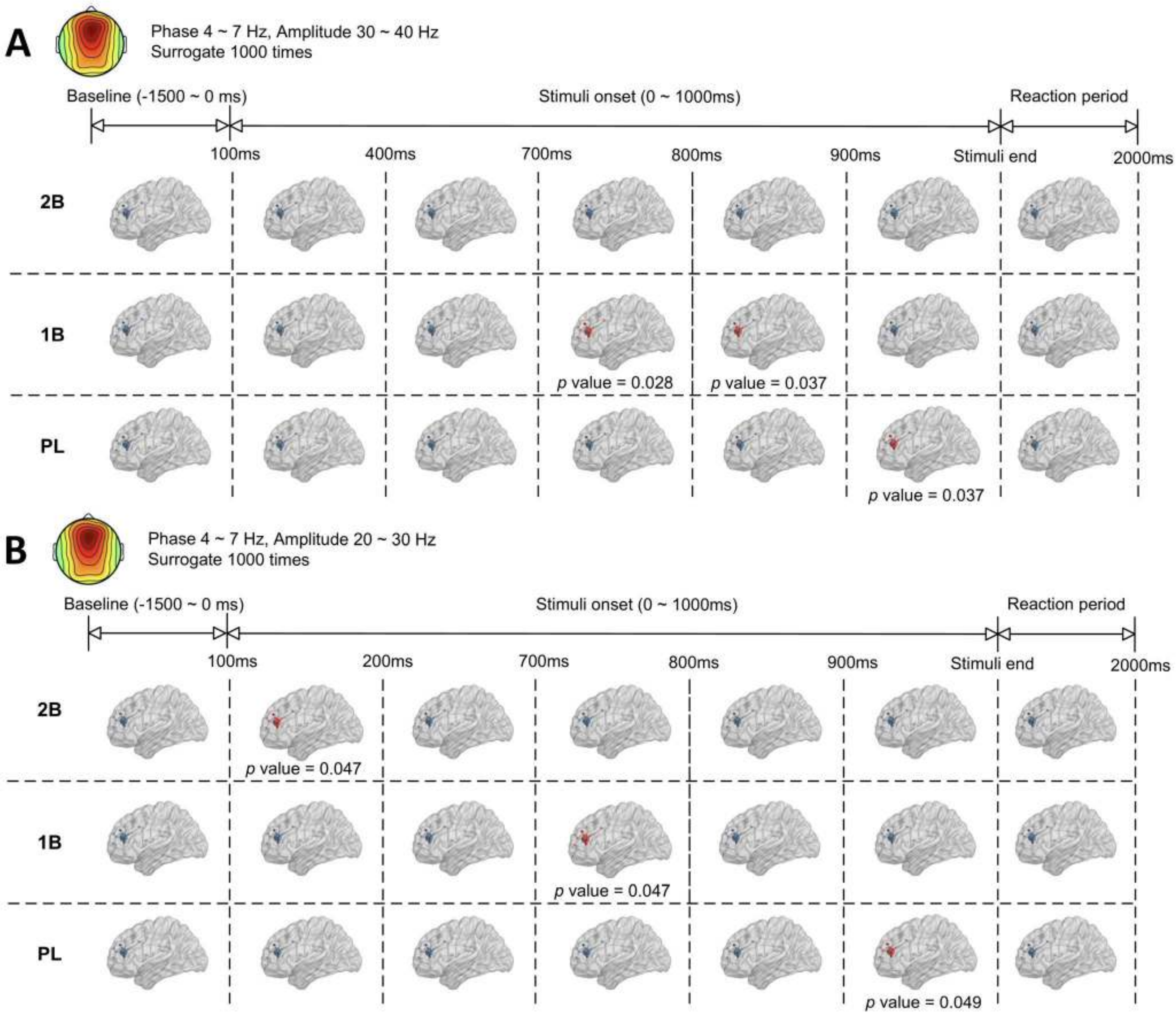

FIGURE 4. The significance of (A) theta-low gamma and (B) theta-high beta couplings in the frontal regions compared with 1000 times surrogation. The significance level of each experimental condition can be found in corresponding time periods with *p* values. The small and large points are representative of individual and averaged dipole locations with significant ERPACs (red point) and non-significant ERPACs (blue point), respectively.

parietal regions can contribute to memory retrieval, identification of musical items, and decision-making. Moreover, the findings indicate that ERPACs in these brain regions might be related to memory retrieval processes and contribute to successful auditory memory recall.

A. EVENT-RELATED PHASE-AMPLITUDE COUPLING IN THE FRONTAL AND PARIETAL BRAIN REGIONS

Previous observations suggest that coupling between phase and amplitude in the frontal and parietal regions is involved in cognitive processes [3], [14], [19], [22], [31]–[33], [35], [40], [46]. Previous PAC findings underlying cognitive procedures in humans are listed in Table 2, including the related brain regions, modalities, and frequency bands involved in PAC.

Outcomes from previous neuroscience studies have demonstrated results similar to those of the present study. First, the frontal [3], [14], [16], [19], [20], [22], [32], [46], [89] and parietal [3], [14], [19], [24], [32], [40] regions have been proposed to be engaged in WM processes including sound recognition [14], auditory WM [28], [56], memory retrieval [14], [32], reward procedure [46], decision making [3], and WM capacity [19]. Consistent with previous findings, our EEG results also suggest the presence of ERPACs in both the frontal and parietal regions when performing musical cognitive tasks during memory retrieval.

Additionally, cognitive PAC studies have reported that the theta phases and gamma power oscillations in the parietal region during an averaged epoch lasted from 400 to 1300 ms [22] and 100 to 600 ms post-stimuli [32],

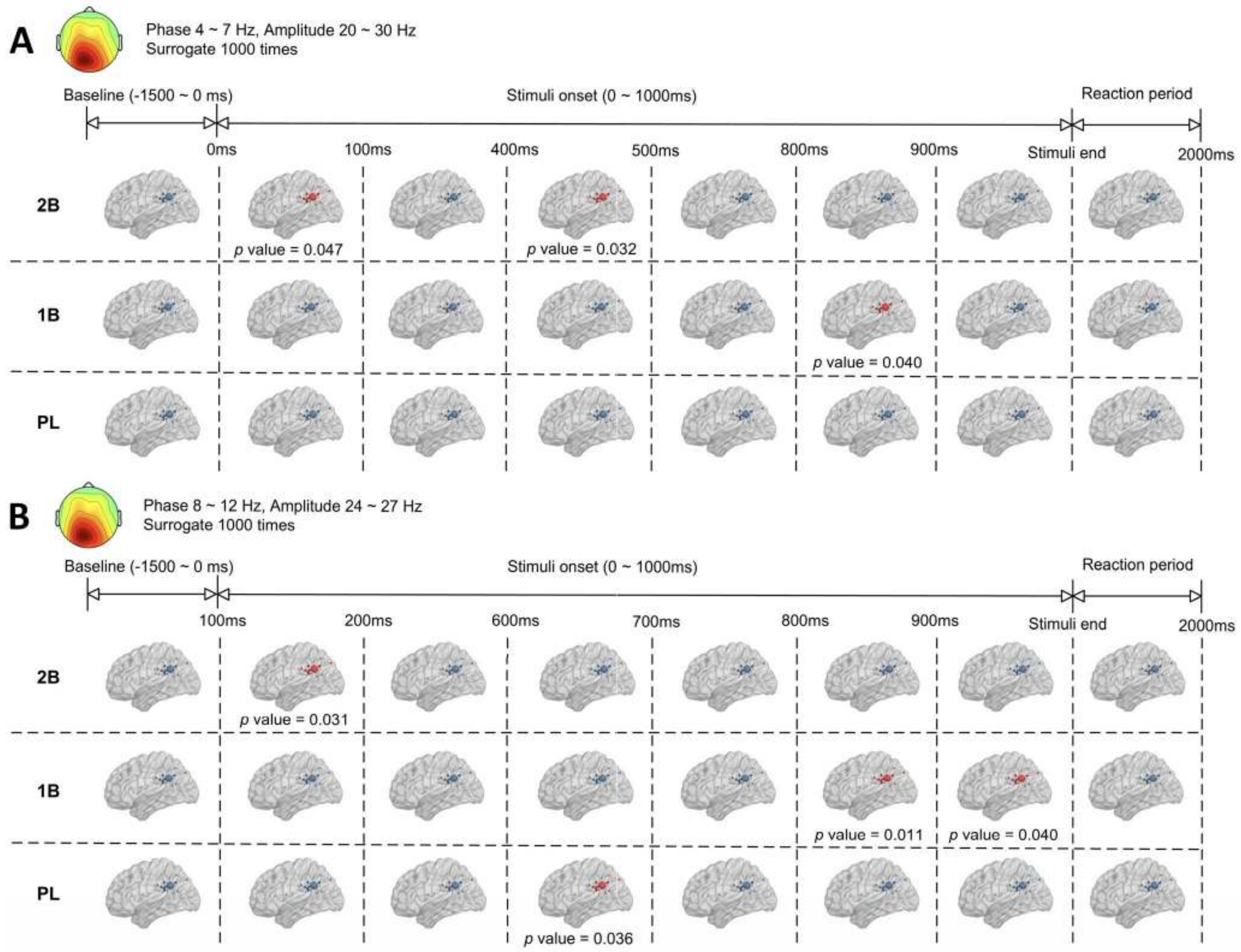

FIGURE 5. **The significance of (A) theta-high beta and (B) alpha-high beta couplings in the parietal regions compared with 1000 times surrogation. The significance level of each experimental condition can be found in corresponding time periods with *p* values. The small and large points are representative of individual and averaged dipole locations with significant ERPACs (red point) and non-significant ERPACs (blue point), respectively.**

respectively. Another PAC study reported that beta oscillations were modulated by theta/alpha phases during a delay period (500-3000 ms) [33].

Various studies have reported that theta-gamma PAC and theta-beta PAC were related to cognitive tasks in humans [3], [14], [19], [22], [25], [28], [31], [32], [34], [35], [43], [48] and animals [2], [3], [5], [14], [18]–[20], [28], [32], [84], [90]–[96]. However, most of these studies were performed using traditional PAC. Although traditional methods of measuring the strength of PAC are precise and intuitive, these methods lack temporal resolution. Moreover, a predefined time window is required, which may result in several limitations [36]. Therefore, it is believed that ERPAC with better temporal resolution can provide a more precise analysis of the timing and function of neural oscillations during cognitive processes. In the present work, by applying ERPAC to the frontal and parietal EEG components, the precise timing of coupling can be derived. Pronounced ERPACs were mainly observed between 100 and 500 ms in the 2B condition with higher WM load, 700 and 1000 ms in the 1B condition, and after 600 ms in the PL condition. Significance was found between 0 and 900 ms after the stimulus onset, while weak coupling was detected after 1000 ms during the late period around the reactions. Although there are still few studies examining the time-resolved or event-related PAC of auditory memory, our results are in line with previous findings [14], [32]. A recent ERPAC study on auditory odd-ball also mentioned the existence of ERPAC in the first 600 ms after the stimulus onset [32]. Furthermore, our previous results on musical memory retrieval indicated that elevated theta-gamma coupling during memory retrieval was observed at the beginning of each trial [14]. This study further suggested that the timing of ERPAC may be correlated with cognitive load. With a higher memory load, the observation of ERPAC may be earlier after the occurrence of stimuli.

B. COUPLING OF VARIOUS FREQUENCY BANDS IN THE NEOCORTEX

Neuronal dynamics data have demonstrated the presence of varying frequency bands in cognitive tasks, especially

TABLE 2. Previous cognitive task findings with involved brain regions and coupling frequency band.

Study	Task	Brain Regions	Modality	Coupling Frequency Band
Canolty (2006) [34]	Cognitive tasks	Frontal, Temporal	ECoG	Theta-Gamma
Cohen (2008) [18]	Decision making	Frontal	EEG	Theta-Alpha, Theta-Beta
Cohen (2009) [46]	Reward	Frontal	Intracranial EEG	Alpha-Gamma
Handel (2009) [19]	Visual sensory	Frontal, Temporal	MEG	Delta-Gamma
Axmacher (2010) [47]	Working memory	Hippocampus	Intracranial EEG	Theta-Gamma
Voytek (2010) [18]	Visual attention	Posterior visual cortex	EEG	Alpha-gamma
Friese (2013) [22]	Memory encoding	Frontal Posterior	EEG	Theta-Gamma
Roux (2014) [24]	Working memory	Frontal, Parietal	EEG	Theta-Alpha, Alpha-Gamma
Arnal (2014) [40]	Auditory temporal prediction	Temporal, Parietal	MEG	Delta-Beta
Lega (2014) [22]	Memory formation	Frontal Temporal	ECoG	Theta-Gamma, Alpha-Gamma
Szczepanski (2014) [28]	Visual attention	Frontal, Parietal	EcoG	Theta-gamma, Delta-gamma
Nakatani (2014) [34]	Visual attention blink	Posterior, midline-parietal	EEG	Theta-Gamma, Theta-Beta, Delta-Beta, Delta-Gamma
Voytek (2015) [31]	Information encoding	Prefrontal	ECoG	Theta-Gamma
Rajji (2016) [89]	Visual working memory	Frontal	EEG	Theta-Gamma
Park(2016) [42]	Visual memory	Visual cortex	MEG	Alpha-Gamma
Daume (2017) [33]	Visual working memory	Left Inferior Temporal	MEG	Theta-Beta, Alpha-Beta
Bachiller (2017) [32]	Auditory oddball task	Parietal	EEG	Alpha-Beta, Alpha-Gamma
Morillon (2017) [97]	Pitch categorization task	Frontal, Temporal	MEG	Delta-Beta
Kikuchi (2017) [98]	Sequences of nonsense words	Auditory cortex	Intracranial recordings	Theta-Gamma
Köster (2018) [99]	Associative memory	Frontal	EEG	Theta-Gamma
Hirano (2018) [54]	Auditory steady-state response	Left hemisphere	EEG	Theta-Gamma
Keitel (2018) [100]	Speech	Central	MEG	Delta-Beta
Köster (2019) [101]	Visual memory entrainment	Left temporal, Centro-frontal	EEG	Theta-Gamma
Tseng (2019) [14]	Auditory memory	Frontal, Parietal	EEG	Delta-Beta, Theta-Gamma
Lee (2020) [102]	Natural sounds, emotion	Left frontal	EEG	Delta-Alpha
Davoudi (2021) [103]	Visual attention	Frontal, Temporal	EEG	Theta-Gamma, Alpha-Beta, Alpha-Gamma
Liang (2021) [104]	Visual working memory	Mid-frontal and right-parietal	EEG	Theta-Beta
Vivekananda (2021) [105]	Spatial memory	Hippocampus	Intracranial EEG	Theta-Gamma
Low (2021) [106]	Emotional prosody	Frontal, Temporal	MEG	Theta-Gamma, Alpha-Gamma

the engagement of theta, gamma, and beta band oscillations. The first indication of theta-gamma neuronal code is widely believed to exist in the subcortical region of primates. It was initially demonstrated to be activated within the hippocampus in animal experiments reported by Tort and colleagues during the T-maze task [33], and the related research and mechanisms were organized by Lisman and Jensen [5]. The existence of PACs was later demonstrated in the neocortex region by EEG and MEG [3], [14], [16], [22], [25], [28], [33], [34], [40], [47], [87], [88]. Additionally, the involved oscillatory frequency bands included not only theta and gamma bands but also delta, alpha, and beta frequency bands. As noted by Lisman, the occurrence of varied frequency bands might be due to the mixing of frequencies, which means that several frequencies might be lumped together when coupling was observed in the neocortex. Moreover, neuronal responses in one frequency band might increase in some brain regions and decrease in others during cognitive tasks [5]. Because of the engagement of varied frequency bands in the neocortex, they are proposed to be engaged in memory processes. For instance, theta-band oscillations are widely believed to exist in memory processes [5], [43], and gamma frequency oscillations have been proposed to be involved in attention and memory [5], [90]. Additionally, beta oscillations are associated with attention, memory retrieval, alertness, and perception [107, 108]. Enhanced neuronal coupling between theta phases and gamma amplitudes [2], [3], [5], [14], [16], [18], [20], [22], [28], [32], [34], [36], [37], [41], [47], [48], [87], [88], [90], [109], as well as theta phases and beta amplitudes [19], [33], [34] have been linked to memory tasks and cognitive functions.

In 2001, Kahana and colleagues proposed that theta-gamma coupling in humans and rodents is involved in memory coding and WM mechanisms [5]. Additionally, Canolty and colleagues observed the involvement of theta-gamma coupling in cortex communication during cognitive procedures [110]. In 2008, Lisman and Buzsaki introduced theta phases and gamma powers as important factors in memory neural coding schemes [2]. Tort and colleagues also showed an association between theta-gamma PAC and learning, memory recall, and long-term memory in 2009 [5]. Axmacher and colleagues proposed the engagement of theta-gamma PAC in WM performance in 2010 [39]. Frise and colleagues indicated the relevance between theta and gamma oscillations and memory storage in 2013 [22]. Lega and colleagues have also pointed out the involvement of theta-gamma PAC in memory formation in the human hippocampus [41]. Tseng and colleagues reported an association between theta-gamma ERPACs and musical memory retrieval in 2019 [14]. As for theta-beta oscillations, Cohen and colleagues observed theta-beta coupling during decision making in 2008 [3]. In 2004, Nakatani and colleagues showed an enhanced PAC between the theta phase and beta amplitude in visual attention blink tasks [34]. Later in 2017, Daume and colleagues reported an oscillatory correlation between the theta/alpha phase and beta amplitude during visual WM maintenance [33]. Additionally, although Axmacher and colleagues [5] reported theta-gamma coupling, the amplitude of the active frequency band was between 14 and 50 Hz, which also includes the beta frequency band. Therefore, the observed theta-gamma and theta-beta couplings in this study were in line with previous findings.

In agreement with previous studies, pronounced coupling frequency bands were observed between the theta phases and beta/gamma amplitudes. Oscillations across other frequency bands can also be detected in different subjects. However, after averaging in the group analysis and surrogating, the signals were not significant. Despite the restriction of small sample size, ERPAC, an improved temporal-resolved method, was applied to analyze EEG signals in the present study. All results were analyzed in compliance with the statistical method of significance. Furthermore, our ERPAC results can provide a more precise timing of neuronal oscillations.

Although this study demonstrated the existence of PAC in the neocortex as an indicator of neural coding scheme formed by EEG oscillations during complex auditory WM processes of musical chords, there is still a huge gap in the study of PAC during auditory cognitive processes. Since there are many different kinds of auditory stimuli involved in real-life cognitive processes such as sound tones, chords, speech, or even complex melodies, previous studies using PAC to investigate the neural mechanisms of auditory cognition have utilized many different types of experimental designs. Different types of auditory stimuli with simple tones [40], [54], [97] or more complex auditory tasks [98], [100], [102], [106] have been widely observed. The utilization of these diverse types of experimental designs made the findings too difficult to converge. Moreover, even the results of the aforementioned auditory tasks using simple tones as stimuli have not provided a convergent and comprehensive perspective of auditory memory mechanisms [32], [40], [54], [97]. Therefore, future efforts should be devoted to systematically decompose these complex auditory stimuli and make sound properties comparable with traditional experimental designs for studying the underlying neural mechanisms.

V. CONCLUSION

This study encompasses an experimental paradigm involving musical chords. The utilization of a certain kind of experimental design that is closer to real-life situations has become a growing trend in cognitive neuroscience. An advanced signal processing technique, ERPAC, was applied to analyze the EEG signals recorded from the complex paradigm of musical chords. The results provide a more precise timeline for observing oscillations and ERPACs in brain regions. The findings of this study suggest that elevated theta-gamma coupling was observed in the first 1000 ms, and the timing of ERPAC may be correlated with cognitive load. We demonstrated the existence of PAC in the neocortex as an indicator of the neural coding scheme formed by EEG oscillations during complex auditory WM processes of musical chords. The present work might help in establishing the mechanisms of neural oscillations and the scheme of memory encoding during musical WM. It demonstrated not only the relevance of auditory WM tasks and neuronal oscillations, but also revealed a correlation between PAC and cognitive load. Furthermore, this study suggests possible clinical applications for effective learning and memory training.

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