

BRAIN-MIND-BODY PRACTICE AND HEALTH

EDITED BY: Gao-Xia Wei, Gangyan Si and Yi-Yuan Tang
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BRAIN-MIND-BODY PRACTICE AND HEALTH

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It is acknowledged that practice could induce rapid change or reorganization of the brain's cellular or neural networks as well as behaviors. Notably, practice relevant to mental or physical approach attracted great attention in this decade. It highlights profound significance both for human evolution and individual development. Specifically, acquiring fine motor skills is a crucial premise for human being to evolve to modern human by using tools in one side. In the other side, numerous evidences indicated that motor learning involved in limb and trunks promotes the development of individual brain in anatomy and functions. Hence, motor learning is also tightly associated with developmental plasticity. These studies on brain-mind-body practice illuminate a promising way in promoting human brain health.

This editorial covers wide range of brain-mind-body practice forms to summarize recent new findings and development from behavioral, physiological, neurobiological and psychological science approaches. In this research topic, we addressed recent findings from theoretical as well as experimental perspective including contributions under the following three headings: 1) intervention studies to investigate the positive effect of brain-mind-body practice on cognition and relevant brain mechanism. The intervention pattern consisted of short-term practice ranging from few hours to several weeks; 2) cross-sectional studies using expert-novice paradigm to explore the behavioral and neural system change induced by extensive brain-mind-body practice; 3) the mediators influence the relationship between practice and health outcomes and 4) new viewpoints on brain-mind-body practice from theoretical perspectives. Here we briefly highlight these articles aiming to provide a deep understanding for the association between practice, plasticity and health for readers. Additionally, it offers new insights for developing possible practice interventions for clinical treatment of neurological dysfunction or disorders.

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Editorial: Brain-Mind-Body Practice and Health

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Editorial on the Research Topic

Brain-Mind-Body Practice and Health

Currently, increasing number of human studies emerged to demonstrate the association between “brain-mind-body practice and health” (Lustig et al., 2009; Bezzola et al., 2011; Wei et al., 2013, 2014), which is of great implication for understanding basic scientific issue “mind and body” and providing efficient strategy for clinical practice and health promotion (Lan et al., 2013; Renoir et al., 2013; Acevedo et al., 2016; Muehsam et al., 2017; Tang, 2017). Here in this research topic, we addressed recent findings from theoretical as well as experimental perspective including contributions under the following three headings: (1) intervention studies to investigate the positive effect of brain-mind-body practice on cognition and relevant brain mechanism. The intervention pattern consisted of short-term practice ranging from few hours to several weeks; (2) cross-sectional studies using expert-novice paradigm to explore the behavioral and neural system change induced by extensive brain-mind-body practice; (3) the mediators influence the relationship between practice and health outcomes and (4) new viewpoints on brain-mind-body practice from theoretical perspectives. Here we briefly highlight these articles aiming to deepen our understanding of relevant development in this topic and establish organic connections among these studies as well as the connections between this topic and broader research field. In addition, it offers an academic insight from theoretical and methodological perspectives for readers. To provide a thorough understanding of these contributions, we classified these publications based on study design.

It is challenging to clarify the influence of brain-mind-body practice on health outcomes because it was misunderstanding that some outcomes could be detectable only after long-term training or practice, which usually takes much time to conduct relevant longitudinal studies. Therefore, researchers found a good solution using acute exercise paradigm only lasting for about 30 min. In this topic, acute practice among healthy and clinical population were also involved. In this topic, Hung et al. employed acute exercise with moderate intensity to investigate its effect on task switching in children with attention-deficit/hyperactivity disorder (ADHD), which indicated that following exercise these children with ADHD exhibited decreased reaction time and increase P3 amplitude compared to control session. This result suggested that single bouts of moderate intensity aerobic exercise might have positive effects on the working memory of children with ADHD. Chen et al. used functional MRI to examine the neural mechanism underlying the effect of acute aerobic exercise on executive function in healthy children. The results also supported the positive effect of aerobic exercise even it only lasted for only 30 min. Moreover, this study found greater activation in brain regions relevant to working memory after exercise compared to resting condition. It is a remarkable attempt to adopt functional MRI technology to explore such effect in spite of the small sample size. Functional MRI was used in another study on motor fatigue (Hou et al.). This study observed the change of brain activation induced by a hand movement task

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between following an exhaustive exercise. It consistently observed significantly decreased activation in subcortical areas (basal ganglia) during motor task and implied the key role of subcortical areas in motor fatigue by disturbing motor control processing. Additionally, traditional Electroencephalogram (EEG) study was also included in this topic. Henz and Schollhorn explored the difference of brain activity by comparing after vs. before practice among Qigong practitioners and found both physical practice and mental practice had similar increased alpha activity, indicating mental practice is also an efficient way to reach the relaxed state.

Another solution to detect the effect of practice on health is adopting cross-sectional study to compare the differences in behavioral and neural level between experienced practitioners and controls. Wei et al. used functional MRI to investigate the neural correlates underlying the effect of extensive mind-body practice on cognitive control in large-scaled brain network perspective. They recruited Tai Chi Chuan (TCC) practitioners for 10 years of experience as expert group and controls as novice group to scan their brain function during resting state. The results showed that compared to control group, TCC group had significantly decreased fractional Amplitude of Low Frequency Fluctuations (fALFF) in bilateral frontoparietal network (FPN), which was found to be associated with TCC experience as well as cognitive control performance. This study highlights the functionally plastic role of the frontoparietal network in the context of the “immune system” of mental health recently developed in relation to flexible hub theory. Professional athlete is a typical human model to detect the influence of long-term practice on cognition. Here Xu et al. and her colleagues also used functional MRI to scan task-evoked brain activation during action anticipation task. Greater activation in medial frontal cortex was observed in badminton players relative to control group, which addressed the crucial role of medial frontal cortex in perception anticipation. As mentioned above, the consistent findings in prefrontal cortex possibly supported improved top-down regulation benefited from extensive practice, which might be meaningful for treatment of mental disorders. The limitation of this paradigm is that it is not clear if the differences of behaviors or brain between these groups are induced by nature or nurture (practice) (Shors et al., 2014). It is possible that some practitioners have featured brain tissue sensitive to training and they undoubtedly presented better performance in behaviors. However, these original studies offer more insights for further investigations in potential change of behaviors and neural circuits induced by long-term practice.

In view of this limitation, researchers in the field of brain-mind-body practice utilized short-term intervention period for about several weeks to examine the effect of practice in order to exclude the influence of nature or heredity. Baduanjin, a form of Qigong, was employed as an intervention tool to investigate behavioral change including mood and executive control induced by it. The intervention protocol lasted for 8 weeks. As predicted, Chen et al. detected significant improved mood state and executive function. Moreover, an increase in oxygenated hemoglobin in the left prefrontal cortex was observed during the Incongruent Trails test only after exercise intervention. Similarly,

Ma et al. observed that 8-week diaphragmatic breathing without any explicit movement was observed to improve cognitive function and negative mood as well as decreased stress level among healthy adults. A combined cognitive training consisting of memory strategy and executive function was examined in this topic. The results demonstrated the effects of cognitive training on both intention-based and stimulus-based actions, which supported the role of mental training on action operation (Niu et al.). Intriguingly, Luo et al. investigated the effects of working memory capacity (WMC) and state anxiety (SA) on attentional control, which supported working memory training benefited to improve attentional control. And the relation between state anxiety and attentional control was also discussed in this paper.

Although most evidences supported the association between exercise and health-related outcomes, the variables mediating such relationship still remains largely unknown. Relevant questions were discussed in this heading. Cardiovascular fitness level, regarded as an important mediator, was confirmed to associate with cognitive performance, which were involved in two separate studies. Song et al. mainly demonstrated how obesity and cardiovascular fitness are associated with the inhibition aspect of executive function from behavioral and electrophysiological perspectives. What makes that all the more remarkable is adopting randomized control observation design to examine the cognitive difference and simultaneously recorded participants' brain activity during operating Stroop task. The results confirmed the hypothesis that the status of being both normal weight and having high cardiovascular fitness is associated with better behavioral and later stages of electrophysiological indices of cognitive function. Regarding the cognitive component benefited from cardiovascular fitness, Chu et al. used event-related desynchronization (ERD) and event-related synchronization (ERS) to explore group difference of the same cognitive task. This findings finally suggested that such cognitive advantage is related to the inhibition of task-irrelevant information and those processes required the devotion of greater amounts of attentional resources to a given task. Moreover, trait self-control is another factor to influence the relationship between motivation toward exercise and subjective wellbeing by using structural equation modeling (Briki), which attached importance for motivation during exercise and provided important insight for effective exercise instruction.

Theoretical approach on the relationship between brain-mind-body practice and health is also addressed in this topic. Shen et al. elaborated the theoretical framework of triadic interaction of brain-mind-body (TIBMB) for creative insight. In the opinion, it is emphasized that the brain is separated from the body, which can benefit identifying the role of body components in different psychobiological/biopsychological activities and also manifest the zeitgeist of the embodied approach. By contrast, Tang et al. suggested in his opinion that mind and body have to be integrated to explain the effect of mindfulness practice because mindfulness meditation includes three components that interact closely to constitute a process of enhanced self-regulation: enhanced attention control, improved emotion regulation and altered self-awareness. Mind-body practice is a

holistic system and works through the integration of different ingredients rather than separated components to achieve the beneficial effects. Moreover, an interesting approach in mind-body-brain practice is optimal performance. Regarding the association between body and brain, Cheron (2016) put forward a new neuroscience perspective to investigate one form of optimal experience—"flow" state, which pointed out the possible way to measure psychological "flow" with EMG and EEG technology. Similarly, another form of optimal experience—clutch state was attached great importance in investigating mind-body-brain association (Swann et al., 2017). In this issue, it is firstly demonstrated the relationship among optimal performance and mindfulness training (Tang and Bruya). Also the necessity is addressed to explore underlying mechanisms (e.g., key biomarkers) of mind-body interaction and optimal performance.

In summary, these contributions in this research topic cover majority forms of brain-mind-body practice including mindfulness, Tai Chi Chuan, Qigong, cognitive training and aerobic exercise. Readers who are interested in any brain-mind-body practice could have access to basic knowledge on theoretical background and get a glimpse of new development and research findings. The publications in this special issue

involve multi-modal techniques to uncover the effect of practice in psychological, physiological, neurobiological, and immunological levels. Thus, all these studies enrich our understanding of neural mechanisms underlying healthy behaviors as well as the association between mind, body and brain, and offer new insights for developing possible behavioral practice interventions in subjects with neurological or mental dysfunction.

AUTHOR CONTRIBUTIONS

GW drafted and revised the manuscript. GS and YT finalized and revised the manuscript.

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Neuroelectric and Behavioral Effects of Acute Exercise on Task Switching in Children with Attention-Deficit/Hyperactivity Disorder

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The main purpose of this two-part study was to examine the effects of acute, moderate intensity exercise on task switching in children with attention-deficit/hyperactivity disorder (ADHD). In Study 1, we compared the task switching performance of children with and without ADHD. Twenty children with ADHD and 20 matched controls performed the task switching paradigm, in which the behavioral indices and P3 component of event-related potentials elicited by task-switching were assessed simultaneously. The amplitude and latency of P3 reflected the amount of attention resource allocated to task-relevant stimulus in the environment and the efficiency of stimulus detection and evaluation, respectively. The task switching included two conditions; the pure condition required participants to perform the task on the same rule (e.g., AAAA or BBBB) whereas the mixed condition required participants to perform the task on two alternating rules (e.g., AABBA...). The results indicated that children with ADHD had significantly longer RTs, less accuracy, and larger global switch cost for accuracy than controls. Additionally, ADHD participants showed smaller amplitudes and longer P3 latencies in global switch effects. In Study 2, we further examined the effects of an acute aerobic exercise session on task switching in children with ADHD. Thirty-four children with ADHD performed a task switching paradigm after 30 min of moderate-intensity aerobic exercise on a treadmill and after control sessions (watching videos while seated). The results revealed that following exercise, children with ADHD exhibited smaller global switch costs in RT compared with after control sessions. The P3 amplitude only increased following exercise in the mixed condition relative to the pure condition, whereas no effects were found in the control session. These findings suggest that single bouts of moderate intensity aerobic exercise may have positive effects on the working memory of children with ADHD.

Keywords: ERP, working memory, switch cost, executive function, physical activity

INTRODUCTION

Attention-deficit/hyperactivity disorder (ADHD) is one of the most common disorders in children, present in approximately 8.7% of children in the U.S. (Froehlich et al., 2007), and approximately 50% of children with ADHD experience symptoms that persist into adulthood (Lara et al., 2009). ADHD can cause numerous impairments in social, academic, and occupational functioning (Bledsoe et al., 2010), resulting in a substantial economic impact on society (Pelham et al., 2007). Medication, behavioral treatment, combinations of medication and behavioral treatment, and community care are four distinct treatment strategies for ADHD (Molina et al., 2009). Of these, the most widely and effectively used treatment for ADHD centers on pharmacotherapy, namely stimulants, as the first-line treatment in children with ADHD (Craig et al., 2015). Despite the positive effects of pharmaceutical treatment on the behavioral symptoms of ADHD, some potential side effects may occur, such as insomnia, appetite reduction, moodiness, and headaches (Schachter et al., 2001), and this type of treatment exhibits limited long-term gains that often disappear once treatment is discontinued (Chronis et al., 2006; Steinberg-Epstein et al., 2011). As a result, it is important to further investigate lifelong adjunctive or alternative potential treatment options that may benefit children with ADHD.

The executive function model (Pennington and Ozonoff, 1996) has been proposed to explain ADHD deficits (Sergeant et al., 2003). Emerging research consistently documents that ADHD is characterized by cognitive deficits, especially in executive function (Barkley, 1997; Barkley and Lombroso, 2000). Executive function (EF), or executive control, is an umbrella term that is used to describe “higher or meta-” cognitive function, including shifting, updating, inhibition (Miyake et al., 2000), switching, working memory, and sustained and selective attention (Alvarez and Emory, 2006). A previous meta-analytic study indicated that compared to healthy controls, children with ADHD consistently performed worse on EF tasks. The effect sizes of all measures fell within the medium range (0.46–0.69), but the strongest and most consistent effects were obtained for measures of response inhibition, vigilance, working memory, and planning (Willcutt et al., 2005). These studies point to a deficiency in multiple aspects of EF in ADHD.

Some studies have shown a positive effect of exercise or physical fitness on several aspects of EF in children with ADHD. For example, motor ability (Hung et al., 2013) and physical fitness (Tsai et al., 2016) were reported positively associated with better inhibitory function, a subcomponent of EF, in children with ADHD. Similarly, a long-term exercise program enhanced performance on inhibitory tasks (Chang et al., 2014) and idle state brain oscillations during a resting condition (Huang et al., 2014). Similarly, acute exercise benefits EF not only in healthy controls but also in children with ADHD. Medina et al. (2010) found that sustained attention was significantly improved following exercise. Furthermore, evidence indicated that acute exercise facilitated set shifting and inhibition performance as assessed by the Wisconsin Card Sorting Test and the Stroop Test, respectively (Chang et al., 2012). Similarly, Pontifex et al. (2012) found that single bouts

of moderately intensive aerobic exercise improved inhibitory control in children with ADHD. The beneficial effects of acute exercise on inhibition was corroborated by a recent finding that acute exercise significantly improved performance on all three conditions of the Stroop Task but not on the Tower of London or Trail Making Test, in children with and without ADHD (Piepmeier et al., 2015).

In addition to behavioral indicators, event-related potentials (ERPs) have been used to assess the neurophysiological processes involved in the performance of executive function tasks. P3, one of the most studied ERP components, is a positive-going deflection in the ERP waveform that occurs in response to a stimulus, with the amplitude of the component reflecting the allocation of neural resources toward the stimulus (Polich, 2007). Therefore, measurement of P3 can reveal detailed neurophysiological information elicited by the EF task. In the only study involving acute exercise and EF performance with ERP measurement in children with ADHD, Pontifex et al. (2012) found that both children with ADHD and healthy controls exhibited larger P3 amplitudes during a flanker task, an inhibition task, after exercise compared with after reading, indicating acute exercise facilitate neurophysiological processes enabling regulatory adjustments in behavior. Thus, more studies employing ERPs to reveal the underlying neurophysiological processes for the benefit of acute exercise on the EF function in children with ADHD, are warranted.

Most of the studies examining the effects of acute exercise on EF in children with ADHD have focused on sustained attention (Medina et al., 2010), inhibitory control (Chang et al., 2012; Pontifex et al., 2012; Piepmeier et al., 2015), and set shifting (Chang et al., 2012). Whether the beneficial effects of exercise can be extended to other subcomponents of EF, working memory in particular, remains unknown. Task switching paradigms are one of the tasks that have been used extensively to examine the working memory, inhibition, and mental flexibility aspects of executive function (Monsell, 2003). The advantage of this paradigm is that it enables the separation of different components of executive control, such as task-set selection and maintenance (working memory), task-set switching (mental flexibility), and interference control (inhibition; Cepeda et al., 2001). A typical task switching paradigm consists of two conditions, such as repeated task trials in task-homogenous blocks (e.g., AAAA, BBBB: pure condition), switching trials in task-heterogeneous blocks (e.g., AB or BA: switching mixed condition), and non-switching or repeated task trials in task-heterogeneous blocks (e.g., AA or BB: non-switching mixed condition). The global switch costs, a measure of working memory, refer to the difference in reaction time (RT) between mixed and pure conditions, as this difference reflects the efficiency in maintaining multiple task sets in working memory as well as the selection of the task to be performed next (Kray and Lindenberger, 2000). The local switch costs, a measure of inhibition and mental flexibility, refer to the differences in reaction time between non-switching and switching trials in mixed condition, reflecting the effectiveness of the executive control processes responsible for activating the currently relevant task set and deactivating

the task set that was relevant on the previous trial (Kray and Lindenberger, 2000).

Children with ADHD have shown impaired performance in task switching. Studies comparing task switching performance between children with and without ADHD have shown that children with ADHD demonstrate substantially larger switch costs than children without ADHD. However, when on medication, ADHD children's switch performances were equivalent to those of control children. In addition, medication was observed to reduce ADHD children's switch cost (Cepeda et al., 2000). Adults with ADHD also show generally slower and less accurate performances on task switching, suggesting deficits on several EF components in ADHD (King et al., 2007). Despite the lack of studies using ERPs to examine the differences on this topic, studies using functional magnetic resonance imaging (fMRI) have shown that ADHD adults do not display specific executive control problems at the behavioral level but do engage different brain areas during task switching than healthy controls; these differences include reduced activation in the bilateral inferior prefrontal cortex, caudate and thalamus during both Stop and Switching tasks, as well as in the left parietal lobe during the Switching task. The authors suggested that adults with childhood ADHD experienced reduced activation and inter-regional functional connectivity of fronto-striatal networks (Cubillo et al., 2010).

Based on the evidence mentioned above, acute exercise may be particularly beneficial for executive function. However, there are many different aspects of executive function. To our knowledge, no ERP studies to date have directly compared the differences between individuals with ADHD and healthy controls, and the effects of acute moderate exercise on the working memory, inhibition, and mental flexibility aspects of executive function in ADHD as measured by task switching have not yet been explored. Accordingly, we conducted two studies. In Study 1, we compared the task switching performance of children with and without ADHD to examine components of executive function deficiencies in task switching performance of children with ADHD. In Study 2, we further examined the effects of a single bout of acute aerobic exercise on components of executive function using the task switching performance of children with ADHD, particular regarding the aspects of working memory, inhibition, and mental flexibility. Based on previous findings, it was hypothesized that children with ADHD would show deficient task performance in task switching, along with reduced activation of the P3 component. Furthermore, acute exercise would improve performance during the task switching as well as demonstrate increased P3 amplitude and decreased P3 latency elicited by the task switching paradigm.

STUDY 1

Method

Participants

In total, 20 children with a clinical diagnosis of ADHD and 20 healthy match control children (all boys) were recruited through advertisements placed at a local elementary school and ADHD

association. To be included in this study, participants had to meet the following criteria: (1) aged between 8 and 12 years (2) lack of hearing or vision problems and free of brain injury and disease (i.e., epileptic seizure) and (3) lack of comorbid developmental disorders including learning difficulties, dyslexia, Tourette's syndrome, epilepsy and pervasive developmental disorder. All participants had not received medication for at least 24 h before the experiment. This study was conducted in accordance with the Declaration of Helsinki and approved by the National Taiwan University Institutional Review Board. Written assent was obtained from the children, and written informed consent was provided by their legal guardians. In addition to the formal diagnosis provide by a pediatrician or psychiatrist, their legal guardians confirmed the presence of the ADHD symptoms a priori by using the Chinese version of the ADHD test (Cheng, 2008) originally developed by Gilliam (1995), and the Chinese version of the Child Behavior Checklist (CBCL; Chen et al., 2006) originally developed by (Achenbach and Rescorla, 2001). All children were screened for IQ (Test of Non-verbal Intelligence) and were administered the Movement Assessment Battery for Children-2 (MABC-2).

Procedure

After receiving ethics approval, the participants visited the lab on two separate days. During the first visit, the experimental procedure was explained to participants and their legal guardians. Then a health history, demographics questionnaire, ADHD-T, CBCL, and an informed consent form were completed by their legal guardians. The participants were then requested to complete IQ and MABC-2. All of the participants' heights and weights were also measured to calculate their body mass indexes (BMI).

At the second visit, the task switching experiment was explained and carried out in a sound and magnetic shielded room with dimmed lights. Before the formal experiment, the participants were outfitted with an electrode cap and instructed on the task. Both the pure and mixed condition trials were practiced until a criterion of 80% correction rate was reached by participants.

Task Switching Paradigm

Cognitive performance was assessed using the task switching paradigm modified from Dai et al. (2013), presented on a computer monitor controlled via Neuroscan Stim software (ver. 2.0; Neuro Inc., El Paso, TX, USA). In the task, a white numeric digit (digits 1–9, excluding 5) was presented in the center of a computer screen on a black background. The task include pure and mixed task conditions. Pure conditions include two subtests; for the first subtest, participants had to identify whether the number, surrounded by a solid-line rectangle, was smaller (1, 2, 3, and 4) or bigger (6, 7, 8, and 9) than 5 (i.e., AAA. . .). On the second subtest, the participants had to identify whether the number, surrounded by a dashed square, was odd (1, 3, 7, and 9) or even (2, 4, 6, and 8) (i.e., BBB. . .). For the mixed task condition, the two subtests from the pure task condition were combined to form an alternating-runs paradigm (i.e., AABBA. . .) (Rogers and Monsell, 1995). The participants

were instructed to press with their thumb of each hand to make a corresponding response on the key pad as quickly and accurately as possible. All eight digits appeared with equal probability in a random order. The digits were presented for 400 ms, with a 3000 ms response-stimulus interval. If no response was made, the trial was terminated 3500 ms after the onset of stimuli. Participants completed 64 trials in each of the pure task condition and 128 trials (64 trials \times 2 blocks) in the mixed task condition. The first trial in each block for both pure task and mixed-task condition was discarded from the analyses. The viewing distance was approximately 60 cm and visual angles were 3.82°. The task performance measures for reaction time (RT), response accuracy, and global and local switch cost on RT were derived.

ERP Recording and Analysis

Event-related potential was measured with an electrode cap with electrodes placed at 30 sites using the 10–20 system; each electrode was referenced to an average of the mastoid electrodes, and the impedances were kept below 10 k Ω . Continuous data were digitized at a sampling rate of 500 Hz and amplified 500 times with a DC to a 70 Hz filter, and a 60-Hz notch filter was applied using a Neuroscan SynAmps2 amplifier. Only the ERP data recorded from the midline frontal (Fz), central (Cz), and parietal (Pz) locations were analyzed in this study (Dai et al., 2013). The offline data reduction included merging with the behavioral data. The ERP data were corrected for ocular artifacts. Epochs were defined as 100 ms pre-stimulus to 900 ms post-stimulus, and baseline corrections were performed using the 100-ms pre-stimulus interval. A low-pass filter with a 30 Hz cutoff (12 db/octave) was employed to further attenuate noise. ERP trials with amplitudes outside the range of ± 100 μ V were excluded from further analysis. The correct trials were separately averaged. To detect ERP components, P3 mean amplitudes were calculated for 300–700 ms time intervals within a 50-ms interval surrounding the largest positive going peak. Peak latencies were measured within the latency window.

Statistical Analysis

To ensure equivalence between the ADHD and control groups, a *t*-test was applied to compare the demographic data between the two groups. A 2 (Group: ADHD, Controls) \times 2 (Condition: Pure and mixed) and a 2 (Group: ADHD, Controls) \times 2 (Condition: non-switching and switching) mixed ANOVA were employed to analyze the RT and accuracy of the global switch and local switch, respectively. In addition, separate independent sample *t*-tests were used for global (the difference in RT and accuracy between the mixed and pure conditions) and local (the difference in RT and accuracy between non-switching and switching trials in the mixed condition) switch cost between the exercise and resting sessions. The P3 ERP component was assessed separately for amplitude and latency in 2 (Group: ADHD, Controls) \times 2 (Condition: Pure and mixed or non-switching and switching trials) \times 3 (site: Fz, Cz, Pz) mixed ANOVAs for global and local switch.

Results

Demographic Analyses

There were no significant differences between the groups in age [$t(38) = 0.09, p > 0.05$], BMI [$t(38) = 0.44, p > 0.05$], IQ [$t(38) = -1.88, p > 0.05$], MABC-2 score [$t(38) = -1.64, p > 0.05$], suggesting equivalence between the two groups. However, as expected, the results indicated that children with ADHD scored significantly higher than the healthy controls on the ADHD-Q [$t(38) = 7.41, p < 0.01$] and CBCL [$t(38) = 6.86, p < 0.01$]. The demographic characteristics of participants in both groups are summarized in **Table 1**.

Task Performance

Global switch

Table 2 presents the detailed behavioral data for the task switching indices for each group. For reaction time, a two-way ANOVA revealed main effects of Group [$F(1,38) = 10.97, p < 0.01, \eta_p^2 = 0.22$] and Condition [$F(1,38) = 140.74, p < 0.01, \eta_p^2 = 0.79$], with the results indicating that children with ADHD responded slower than the controls. In addition, all participants responded faster during the pure condition compared to the mixed condition (see **Table 2**). The interaction of Group \times Condition was not significant [$F(1,38) = 0.06, p > 0.05$].

For response accuracy, a two-way ANOVA revealed main effects of Group [$F(1,38) = 13.99, p < 0.001, \eta_p^2 = 0.27$] and Condition [$F(1,38) = 54.76, p < 0.001, \eta_p^2 = 0.59$], which were superseded by a Group \times Condition interaction [$F(1,38) = 12.80, p < 0.001, \eta_p^2 = 0.25$]. Because there was a significant interaction effect, a follow-up of simple main effects analysis was utilized to decompose the Group \times Condition interaction. A significant Condition effect was found for both groups, which revealed that ADHD [$F(1,19) = 39.20, p < 0.001, \eta_p^2 = 0.67$] and control [$F(1,19) = 15.78, p < 0.001, \eta_p^2 = 0.45$] groups showed higher

TABLE 1 | Participant demographic characteristics for Study 1.

Variable	ADHD (N = 20)	Control (N = 20)	P
Gender (M:F; M [SD])	20:0	20:0	
Age (years; M [SD])	10.24 \pm 1.78	10.20 \pm 1.09	0.93
BMI (kg/m ² ; M [SD])	17.22 \pm 4.14	16.77 \pm 1.66	0.66
Test of Non-verbal intelligence (score)	100.80 \pm 15.28	108.25 \pm 8.91	0.07
MABC-2 (score)	10.55 \pm 2.91	11.90 \pm 2.25	0.11
ADHD Q (score)	97.05 \pm 14.31	68.35 \pm 9.76	<0.001
CBCL (score)	66.30 \pm 7.39	46.25 \pm 10.77	<0.001
ADHD subtype (N [%])			
ADHD-I	4 (20)		
ADHD-HI	1 (5)		
DAHD-C	15 (75)		
Medicine Intake	9 (45)		

BMI, body mass index; ADHD Q, ADHD quotient, higher quotient indicates higher severity of the ADHD symptom; ADHD-I, predominantly inattentive subtype; ADHD-HI, predominantly hyperactive-impulsive subtype; ADHD-C, combined hyperactive-impulsive and inattentive subtype; N, number of participants; %, percentage in the group.

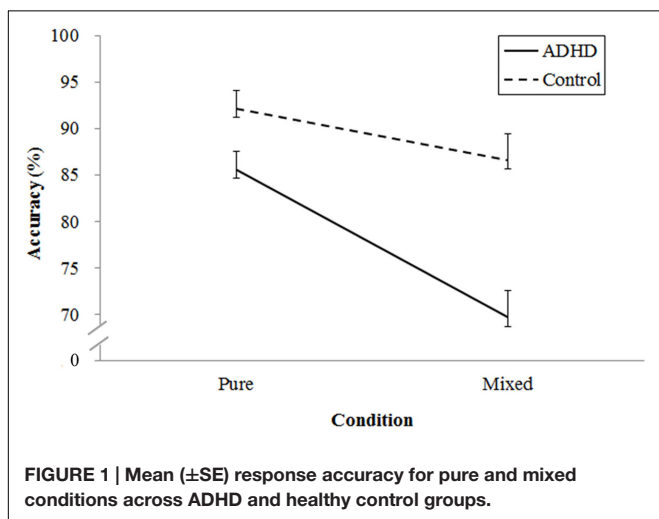
TABLE 2 | Means, and standard deviations for the task switching in Study 1.

Variable	ADHD (<i>n</i> = 20)	Control (<i>n</i> = 20)
	<i>M</i> (<i>SD</i>)	<i>M</i> (<i>SD</i>)
Global switch RT (ms)		
Pure trials	885.91 ± 139.13	691.42 ± 179.17
Mixed trials	1290.34 ± 247.89	1113.01 ± 248.23
Global switch cost	404.43 ± 261.15	421.59 ± 169.60
Local switch RT (ms)		
Non-switch trials	1255.39 ± 229.84	1066.36 ± 244.93
Switch trials	1324.98 ± 271.58	1163.96 ± 255.56
Local switch cost	69.59 ± 95.74	97.60 ± 74.76
Global switch accuracy (%)		
Pure trials	85.63 ± 10.05	92.14 ± 7.09
Mixed trials	69.67 ± 14.57	86.59 ± 10.65
Global switch cost	15.96 ± 11.11	5.56 ± 6.10
Local switch accuracy (%)		
Non-switch trials	70.24 ± 13.65	86.88 ± 10.72
Switch trials	69.12 ± 16.39	86.29 ± 11.56
Local switch cost	1.12 ± 7.76	0.59 ± 6.34

response accuracy in the pure condition than in the mixed condition. Additionally, the significant group effect revealed that the ADHD group had lower accuracy than that of the controls in the pure [$F(1,38) = 54.76, p < 0.001, \eta_p^2 = 0.59$], and mixed condition [$F(1,38) = 54.76, p < 0.001, \eta_p^2 = 0.59$] (see **Figure 1**).

Local switch

Regarding reaction time, a two-way ANOVA revealed main effects for Group [$F(1,38) = 5.01, p < 0.05, \eta_p^2 = 0.12$] and Condition [$F(1,38) = 37.89, p < 0.01, \eta_p^2 = 0.50$], with the results indicating that controls responded faster than participants with ADHD. In addition, all participants responded faster during the non-switch condition compared to the switch condition (**Table 2**). The interaction of Group \times Condition was not significant [$F(1,38) = 1.06, p > 0.05$].

**FIGURE 1 | Mean (±SE) response accuracy for pure and mixed conditions across ADHD and healthy control groups.**

For response accuracy, although no main effects were observed for Condition [$F(1,38) = 0.55, p > 0.05$] or the interaction of Group \times Condition [$F(1,38) = 0.05, p > 0.05$], a significant main effect of Group [$F(1,38) = 17.56, p < 0.01, \eta_p^2 = 0.32$] was shown, with higher accuracy in the control than in the ADHD group (**Table 2**).

Global and local switch costs

There were no significant differences between groups in the global switch costs [$t(38) = -0.25, p > 0.05$] or local switch costs [$t(38) = -1.03, p > 0.05$] for the reaction time. However, a global switch cost for accuracy was observed [$t(38) = 3.58, p < 0.001$]. Examination of the means indicated that the global switch cost in ADHD participants ($M = 15.9\%, SE = 2.55$) was significantly higher than that in controls ($M = 5.56, SE = 1.40$). There was no group difference in local switch cost for accuracy [$t(38) = 0.23, p > 0.05$].

ERP Data

Global switch: P3 amplitudes

The grand average ERP waveforms for task switching, group, and site are illustrated in **Figure 2**. A three-way ANOVA revealed main effects of Group [$F(1,38) = 13.99, p < 0.001, \eta_p^2 = 0.27$] and Condition [$F(1,38) = 54.76, p < 0.001, \eta_p^2 = 0.59$], which were superseded by a Group by Site interaction [$F(2,76) = 4.38, p < 0.05, \eta_p^2 = 0.10$], but no effects were observed for the three-way or other two-way interactions and main effects. Decomposition of the interaction between Group and Site revealed a significant Group effect in the Pz [$t(19) = -2.11, p < 0.05$], with a greater amplitude in controls ($M = 19.98 \mu V, SE = 1.28$) than in participants with ADHD ($M = 15.85 \mu V, SE = 1.48$). Furthermore, a significant Site effect in the ADHD group was revealed [$F(1,38) = 145.75, p < 0.01, \eta_p^2 = 0.88$], with greater amplitudes in the following order: Pz ($M = 15.85 \mu V, SE = 1.48$) > Cz ($M = 6.09 \mu V, SE = 1.60$) > Fz ($M = -2.21 \mu V, SE = 1.43$). For controls, the same effects were found [$F(1,38) = 144.06, p < 0.01, \eta_p^2 = 0.88$], with greater amplitudes in the following order: Pz ($M = 19.98 \mu V, SE = 1.28$) > Cz ($M = 9.49 \mu V, SE = 0.78$) > Fz ($M = -1.25 \mu V, SE = 0.87$).

Global switch: P3 latencies

A three-way ANOVA revealed an interaction of Group and Condition [$F(1,38) = 5.29, p < 0.05, \eta_p^2 = 0.12$], but no effects were observed for the three-way or other two-way interactions and main effects. Decomposition of the interaction between Group and Condition revealed a significant Group effect in pure trials [$t(38) = 3.21, p < 0.01$], with a shorter latency in controls ($M = 541.67, SE = 10.71$) than in ADHD participants ($M = 592.23, SE = 11.55$). However, there were no significant effects for mixed trials. Furthermore, a significant Condition effect in controls [$t(19) = -2.38, p < 0.05$] was observed, with a shorter latency in pure trials ($M = 541.67, SE = 10.71$) than in mixed trials ($M = 577.70, SE = 12.75$). No effects were found in the ADHD group.

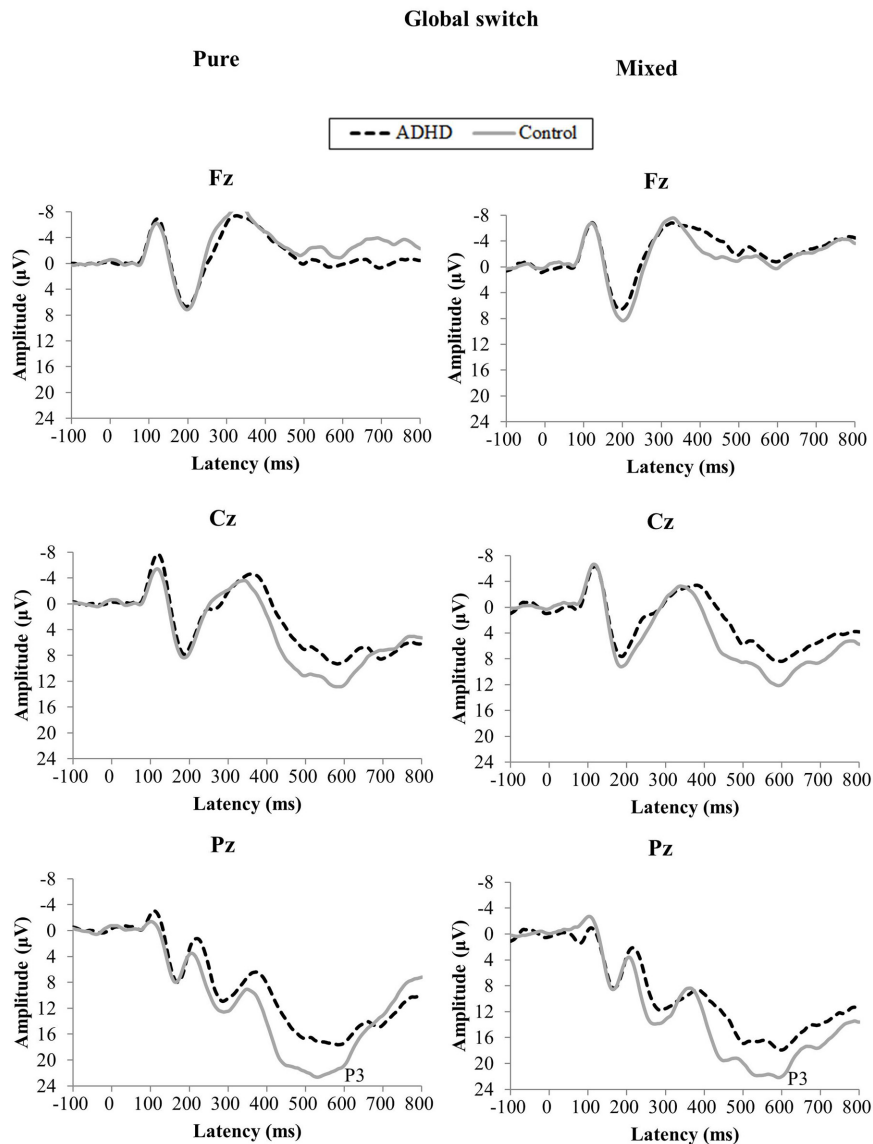


FIGURE 2 | Grand average event-related potentials (ERPs) for pure and mixed conditions of global switch at the Fz, Cz, and Pz sites stratified by group.

Local switch: P3 amplitudes and latency

No three- or two-way interactions or main effects were observed in P3 amplitudes and latency.

STUDY 2

Method

Participants

In this study, 36 children with a clinical diagnosis of ADHD were recruited through advertisements placed at a local elementary school. The inclusion criteria were the same as those of Study 1. Two participants were excluded from the ERP analysis due to contaminated (e.g., VEOG, HEOG, and electromyogram) ERP

data. All participants met the criteria assessed by the Physical Activity Readiness Questionnaire (PAR-Q) before performing the single bout of aerobic exercise to ensure no potential risk factors while performing a single bout of aerobic exercise.

Procedure

Day 1. In the first day, the participants and their parents completed all of the paperwork and tests as described in Study 1.

Days 2 and 3. During the second and third laboratory visits, participants visited the laboratory at the same time of day on two separate days within a given week. After arriving at the lab, the participants were connected to a HR monitor and had their HR recorded after 5 min of seated rest. The participants were counterbalanced into a resting and an exercise

session, to minimize the potential order effect. In the resting session, the participants watched a video for 30 min. In the exercise session, the participants were instructed to complete a 30 min treadmill exercise. Participants were equipped with a Neuroscan Quickcap and administered cognitive tasks following each resting/exercise session. The task switching paradigm and neuroelectrical measurements were the same as those in Study 1.

Acute Exercise Session Manipulation

In the moderate-intensity aerobic exercise session, the participants were instructed to complete 30 min of treadmill exercise, including 5 min of warming up, 20 min of main exercise, and 5 min of cooling down (Chang et al., 2012). The exercise intensity was measured by heart rate reserve (HRR), which was calculated by the formula of Karvonen et al. (1957) as maximal HR minus resting HR, and maximal HR was estimated with an indirect formula of “ $206.9 - (0.67 \times \text{age})$ ” (Gellish et al., 2007). The target HR was calculated by a formula as follows: Target HR = [(maximal HR – resting HR) \times percentage intensity desired + resting HR]. Moderate intensity was set at 50–70% HRR for each participant’s individual HRR. Heart rate (HR) was measured with a Polar heart monitor (Polar RS800CX; Polar Electro Oy, Kempele, Finland) throughout the test. Treadmill velocity was slightly modified based on HR. Three HR data points were recorded, including resting HR, post-exercise HR and mean HR. In addition, ratings of perceived exertion (RPE; Borg, 1998), which provided a subjective rating of each individual’s perceptions of effort during exercise, were assessed every 2 min. We utilized the modify Borg scale, which ranges from 0 to 10 scores.

Statistical Analysis

To assess the exercise intensity manipulation, a 2 (Session: exercise, resting) \times 3 (Time: pre-HR, avg-HR, and post-HR) repeated measures ANOVA for HR was performed.

A 2 (Session: post-exercise, post-resting) \times 2 (Condition: Pure and mixed) and a 2 (Session: post-exercise, post-resting) \times 2 (Condition: non-switching and switching trials in the mixed condition) repeated measures ANOVA were employed to analyze the RT and accuracy of the global switch and local switch, respectively. In addition, separate paired t-tests were used for global (the difference in RT and accuracy between mixed and pure conditions) and local (the difference in RT and accuracy between non-switching and switching trials in the mixed condition) switch cost between the exercise and resting sessions.

The P3 ERP component was assessed separately for amplitude and latency in 2 Session: post-exercise, post-resting) \times 2 (Condition: pure and mixed or non-switch and switch trials) \times 3 (site: Fz, Cz, Pz) analyses for global and local switch.

Results

Demographic Information

The demographic characteristics of participants in Study 2 are summarized in Table 3.

Exercise Manipulation Check

The descriptive data for HR and RPE are summarized in Table 4. The results of the 2 \times 3 repeated ANOVA for HR revealed a significant main effects of Session [$F(1,33) = 889.7, p < 0.001, \eta_p^2 = 0.96$] and Time [$F(2,66) = 802.66, p < 0.001, \eta_p^2 = 0.96$], which were superseded by a Session \times Time interaction [$F(2,66) = 988.88, p < 0.01, \eta_p^2 = 0.97$]. The follow-up simple main effects analysis revealed a significant session effect for avg-HR [$t(33) = 66.94, p < 0.01$] and post-HR [$t(33) = 8.75, p < 0.01$], but not pre-HR [$t(33) = 0.25, p > 0.05$]. Examination of the means showed that the avg-HR and post-HR were significantly higher for the exercise than for the resting session.

Task Performance

Global switch

Table 5 presents the detailed behavioral data for the task switching indices for each group. For reaction time, the results of the 2 \times 2 repeated ANOVA revealed no significant main effects of Session [$F(1,33) = 0.22, p > 0.05$], but significant main effects of Condition [$F(1,33) = 106.01, p < 0.001, \eta_p^2 = 0.76$], which were superseded by a significant Session \times Condition interaction [$F(1,38) = 12.80, p < 0.01, \eta_p^2 = 0.25$]. The follow-up simple main effects analysis revealed significant condition effects for

TABLE 3 | Participant demographic characteristics for Study 2.

Variable	ADHD (N = 34)
Gender (M:F; M [SD])	33:1
Age (years; M [SD])	10.16 \pm 1.74
BMI (kg/m ² ; M [SD])	17.81 \pm 3.80
Test of Non-verbal Intelligence (score)	104.91 \pm 16.89
MABC-2 (score)	10.68 \pm 2.90
ADHD Q (score)	98.66 \pm 14.65
CBCL (score)	67.53 \pm 7.55
ADHD subtype (N [%])	
ADHD-I	8 (23.5)
ADHD-HI	2 (5.9)
ADHD-C	24 (70.6)
Medicine Intake	14 (41.2)

ADHD-I, predominantly inattentive subtype; ADHD-HI, predominantly hyperactive-impulsive subtype; ADHD-C, combined hyperactive-impulsive and inattentive subtype; N, number of participants; %, percentage in the group.

TABLE 4 | Descriptive data for exercise manipulation check in Study 2.

Variable	Control	Exercise
	M (SD)	M (SD)
HR-pre (bpm)	83.88 \pm 8.30	83.62 \pm 7.26
HR-avg. (bpm)	155.11 \pm 3.41	81.5 \pm 6.81
HR-post (bpm)	97.82 \pm 11.34	80.91 \pm 7.03
RPE-avg.	–	5.08 \pm 1.58

bpm, beats per minute; -pre, variable assessed before each treatment interventions; -avg., Average variable assessed during the treatment stage; -post, variable assessed immediately before cognitive task.

TABLE 5 | Means and standard deviations for the task switching in Study 2.

Variable	Post-exercise	Post-resting
	<i>M (SD)</i>	<i>M (SD)</i>
Global switch RT (ms)		
Pure trials	951.96 ± 193.01	900.93 ± 172.54
Mixed trials	1244.95 ± 258.75	1274.02 ± 252.89
Global switch cost	292.99 ± 191.13	373.09 ± 233.71
Local switch RT (ms)		
Non-switch trials	1205.29 ± 254.45	1237.14 ± 245.97
Switch trials	1287.36 ± 280.19	1311.49 ± 269.15
Local switch cost	82.07 ± 132.36	74.34 ± 111.23
Global switch accuracy (%)		
Pure trials	85.24 ± 9.40	87.29 ± 8.86
Mixed trials	73.32 ± 13.49	73.06 ± 14.58
Global switch cost	11.92 ± 11.34	14.23 ± 10.68
Local switch accuracy (%)		
Non-switch trials	74.45 ± 13.05	73.21 ± 14.62
Switch trials	72.21 ± 14.72	72.92 ± 15.72
Local switch cost	2.24 ± 6.90	0.29 ± 8.52

both sessions. Participants responded faster in the pure condition than in the mixed condition both post-exercise [$t(33) = 8.94, p < 0.01$] and post-resting [$t(33) = -9.31, p < 0.01$]. However, no significant differences in pure [$t(33) = 1.85, p > 0.05$] or mixed [$t(33) = -0.95, p > 0.05$] conditions were observed between the two types of sessions.

For response accuracy, although no main effects were observed for Session [$F(1,33) = 0.41, p > 0.05$] or for the interaction of Session \times Condition [$F(1,33) = 1.36, p > 0.05$], a significant main effect of Condition [$F(1,33) = 65.98, p < 0.01, \eta_p^2 = 0.67$] was shown, with more accuracy in the pure ($M = 86.27\%, SE = 1.19$) than in the mixed ($M = 73.19\%, SE = 2.32$) condition.

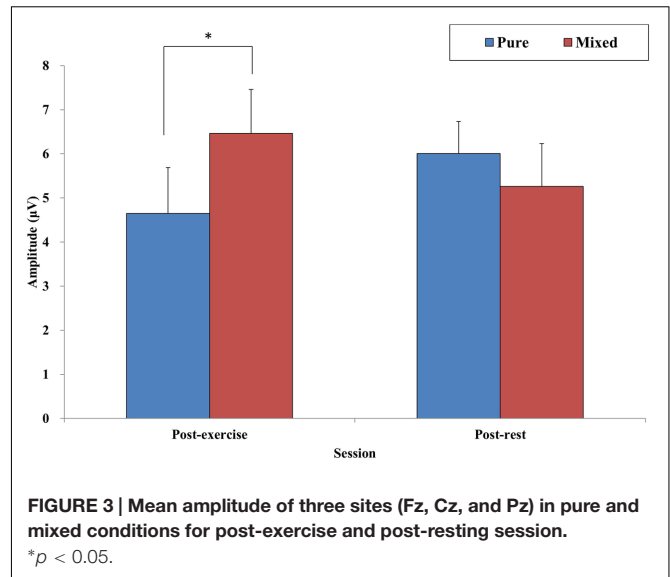
Local switch

For reaction time, a 2×2 mixed ANOVA revealed that there was a significant main effect of Condition [$F(1,33) = 19.79, p < 0.01, \eta_p^2 = 0.37$]; participants responded faster in the non-switching ($M = 1221.22$ ms, $SE = 40.41$) than in the switching ($M = 1299.42$ ms, $SE = 43.54$) trials. However, the main effects of Session [$F(1,33) = 0.84, p > 0.05$] and the interaction of Session \times Condition [$F(1,33) = 0.11, p > 0.05$] were not significant.

For response accuracy, no effects were observed for the main effect of Session [$F(1,33) = 0.04, p > 0.05$] or Condition [$F(1,33) = 2.04, p > 0.05$] or the interaction of Session \times Condition [$F(1,33) = 0.97, p > 0.05$].

Global and local switch costs

As for RT, there were significant differences in global switch costs [$t(33) = -2.34, p < 0.05$] between post-resting ($M = 373.09$ ms, $SE = 40.08$) and post-exercise sessions ($M = 292.99$ ms, $SE = 32.78$). However, no significant differences in local switch costs [$t(38) = -1.03, p > 0.05$] were observed.



With regards to response accuracy, no significant differences in global [$t(33) = -1.17, p > 0.05$] and local switch costs [$t(33) = 0.98, p > 0.05$] were observed.

ERP Data

Global switch: P3 amplitudes and latency

A three-way ANOVA revealed an interaction of Session and Condition [$F(1,33) = 8.67, p < 0.01, \eta_p^2 = 0.21$], but no effects were observed for the three-way or other two-way interactions or for the main effects. A decomposition of the interaction between Session and Condition revealed a significant Condition effect in the post-exercise session [$t(33) = -2.32, p < 0.05$], with a greater amplitude in mixed trials ($M = 6.46 \mu V, SE = 0.72$) than in pure trials ($M = 4.65 \mu V, SE = 1.04$). However, no effects were found in the post-resting session (see **Figure 3**). Regarding P3 latency, no three- or two-way ANOVA interactions or main effects were observed.

Local switch: P3 amplitudes and latency

No three- or two-way ANOVA interactions or main effects were observed in P3 amplitudes and latency.

DISCUSSION

The purposes of this two-part study were to examine the differences in task switching performance in children with and without ADHD and to examine the effects of acute exercise on the switching aspect of executive function, particularly regarding the aspects of working memory, inhibition, and mental flexibility in children with ADHD. In Study 1, the results indicated that children with ADHD demonstrated significantly longer RTs and less accuracy during global switch and local switch conditions than controls. Moreover, children with ADHD had higher global switch cost for accuracy. The ERPs of ADHD participants showed smaller amplitudes and longer latencies in P3 in global switch effects. As for Study 2, the results revealed that following a single

30-min bout of exercise, children with ADHD exhibited smaller global switch costs on RT compared with the costs after a control session. The P3 amplitude only increased following exercise in mixed conditions relative to the pure condition, whereas no effects were found in the control session.

The slower RT in both task switch condition and higher global switch cost for accuracy in children with ADHD are consistent with those of Wu et al. (2006), who found generally slower response times and less accurate responses in a task switching paradigm in children with ADHD. These deficiencies may persist into adulthood, as King et al. (2007) also observed a generally slower and less accurate performance in task switching in adults with ADHD. Global switch costs have been linked to the ability to maintain and schedule multiple task sets and working memory that responds to loading multiple tasks (Kray and Lindenberger, 2000). The finding of higher global switch cost in the present study suggests deficits in working memory in the children with ADHD. This deficiency has been observed in adults with ADHD as well (White and Shah, 2006). This suggests that both children and adults with ADHD may be specifically impaired in working memory, a result consistent with previous findings suggesting working memory is one of the EF components which showed stronger and consistent deficiency in ADHD participants (Willcutt et al., 2005).

The smaller P3 amplitudes in global switch effects in children with ADHD suggest deficiencies in neural resource allocation when challenged with demands on inhibition, mental flexibility, and working memory. Despite the limited research on task switching using ERPs in ADHD participants, the available studies on different executive function tasks also demonstrated smaller P3 amplitudes in ADHD participants. Specifically, during a continuous performance test, a task that demands sustained attention, children with ADHD-com showed smaller frontal N1 and N2 amplitudes and parietal P2 and P3 amplitudes to target stimuli, indicating diminished evaluative and processing capabilities (Lawrence et al., 2005). Similarly, children with ADHD-com exhibited smaller N2 and P3 amplitudes to incongruent flankers, suggesting problems in conflict/inhibition processing (Johnstone et al., 2009).

Additionally, compared to healthy controls, children with ADHD demonstrated a longer P3 latency in the pure condition. Furthermore, only healthy controls showed differences in P3 latency between pure and mixed conditions. This longer P3 latency during task switching in ADHD participants was also observed in other tasks involving executive function. For example, Fisher et al. (2011) found that ADHD participants not only made significantly more commission errors than controls on NoGo trials but also exhibited longer N2 and P3 latencies on a Go/Nogo task. They proposed that the core deficit of ADHD pertained to a regulation disorder that could manifest in a lack of inhibition (commissions) in some situations or over-inhibition in others (omissions), depending on the circumstances. Notably, our ADHD participants showed longer P3 latencies in the pure condition, a condition that requires less mental effort, suggesting an inefficient and possibly disordered regulation in this population. Taken together, our results suggest an impaired working memory at the behavioral level and a slower cognitive

processing and classification speed with regulation disorder in children with ADHD.

Although children with ADHD have shown deficiency in working memory as reflected in higher global switch cost, acute exercise can improve working memory. Specifically, compared with the control session, the acute exercise resulted in smaller global switch costs in RT in children with ADHD. The selective effect of acute exercise on global switch cost is consistent with findings regarding chronic exercise in elderly participants. Themanson et al. (2006) found a lower global switch cost in older adults with higher levels of physical activity than the lower levels of physical activity counterparts. Similarly, older participants who regularly engage in open-skill exercise demonstrated lower global switch costs than sedentary controls (Dai et al., 2013). These results suggest that physical activity may exhibit a greater influence on global switch costs due to the increased working memory load required in mixed trials compared to pure trials (Themanson et al., 2006). Regarding acute exercise and individuals with ADHD, previous studies have shown benefits of acute exercise on other aspects of executive function using the Stroop task (Chang et al., 2012; Piepmeier et al., 2015), Wisconsin Card Sorting Test (Chang et al., 2012), FLanker task (Pontifex et al., 2012), and Go/Nogo tasks (Chuang et al., 2015). Additionally, a meta-analytical review has shown that acute, intermediate-intensity exercise has a strong beneficial effect on response speed in working memory tasks (McMorris et al., 2011). This is the first study to demonstrate that the benefits of acute exercise can be extended to global switch cost in ADHD and suggests that working memory in children with ADHD is more susceptible to the effects of acute exercise. This finding may be particularly important because children with ADHD frequently exhibit impairments in working memory (Martinussen et al., 2005; Willcutt et al., 2005).

Enhanced P3 amplitudes in the mixed condition relative to the pure condition after acute exercise suggests enhancement of working memory processes and is consistent with the behavioral results. A previous study using a flanker task found that following acute exercise, both children with ADHD and healthy controls exhibited larger P3 amplitudes (Pontifex et al., 2012). Similarly, acute exercise also enhanced the preparatory process measured by contingent negative variation (CNV) in children with ADHD (Chuang et al., 2015). These findings of enhanced regulatory processes following acute exercise are particularly beneficial for children with ADHD, considering the fact that one of the core deficits of ADHD is disordered regulation (Fisher et al., 2011).

Although the mechanisms behind the beneficial effects of acute exercise on task switching in children with ADHD have yet to be fully understood, the facilitation effects of acute exercise may potentially be explained by changes in arousal. As the hypoarousal model of ADHD (Satterfield and Cantwell, 1974) suggests, children with ADHD are characterized by a generally lower arousal, and thus acute bouts of exercise that generally arouse the organism can lead to changes in cognitive function (Kamijo et al., 2004). The upregulation of neurochemicals such as neurotrophins (e.g., brain-derived neurotrophic factor, BDNF) and neurotransmitters (e.g., dopamine and norepinephrine) is another possible explanation. Dopamine would be the most

likely candidate for the beneficial effects of acute exercise on global switch cost because of its positive effects on processing speed in tasks involving working memory (McMorris et al., 2011). Using spontaneous eye blinks and the acoustic startle eye blink response as non-invasive measures of dopamine, Tantillo et al. (2002) showed the effectiveness of acute exercise on increasing dopamine levels in children with ADHD. This dopamine hypothesis is also consistent with stimulant treatment, such as with MPH, which increases dopamine levels by blocking dopamine transporters (Swanson and Volkow, 2003) and improves performance on aspects of executive function including spatial working memory, response inhibition, and set-shifting (Arnsten, 2006).

It is important to note that although this study provides evidence supporting the beneficial effects of acute exercise on task switching in children with ADHD using behavioral and neuroelectric measures, several issues should be considered for future research efforts. First, the severity of ADHD symptoms was not evaluated in this study. However, given that 41% of the children in the present study were not receiving medication, it is likely that the participants had mild to moderately severe ADHD symptoms. Therefore, the extent to which we can generalize our results to individuals with more severe ADHD symptoms is unknown. Thus, future research should characterize ADHD symptoms to better understand the utility of acute exercise in enhancing switching ability in these populations. Second, issues related to exercise intensity, types of exercise modality, dose–response relationships, potential moderators (i.e., physical fitness), and time delay effects (Barella et al., 2010), need to be considered in future efforts to design better exercise regimens for adjunctive treatment of children with ADHD.

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CONCLUSION

The present study extends current knowledge on the relationship between acute exercise and executive function in children with ADHD. This study showed that children with ADHD experience certain deficiencies in performing task switching compared to healthy controls. Additionally, the findings indicated that acute bouts of exercise can ameliorate these deficiencies, especially those regarding switching conditions that require greater working memory involvement.

AUTHOR CONTRIBUTIONS

CLH is responsible for the research idea, implementing the study and manuscript writing up. CJH is responsible for the statistical support and discussion commentary. YJT is responsible for assisting on the data collection and analysis. YKC is responsible for consulting the methodology and interpretation of the findings. TMH is responsible for the discussion of the research idea, supervision of the data collection, and comment on the manuscript writing up.

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Decreased Activation of Subcortical Brain Areas in the Motor Fatigue State: An fMRI Study

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One aspect of motor fatigue is the exercise-induced reduction of neural activity to voluntarily drive the muscle or muscle group. Functional magnetic resonance imaging provides access to investigate the neural activation on the whole brain level and studies observed changes of activation intensity after exercise-induced motor fatigue in the sensorimotor cortex. However, in human, little evidence exists to demonstrate the role of subcortical brain regions in motor fatigue, which is contradict to abundant researches in rodent indicating that during simple movement, the activity of the basal ganglia is modulated by the state of motor fatigue. Thus, in present study, we explored the effect of motor fatigue on subcortical areas in human. A series of fMRI data were collected from 11 healthy subjects while they were executing simple motor tasks in two conditions: before and under the motor fatigue state. The results showed that in both conditions, movements evoked activation volumes in the sensorimotor areas, SMA, cerebellum, thalamus, and basal ganglia. Of primary importance are the results that the intensity and size of activation volumes in the subcortical areas (i.e., thalamus and basal ganglia areas) are significantly decreased during the motor fatigue state, implying that motor fatigue disturbs the motor control processing in a way that both sensorimotor areas and subcortical brain areas are less active. Further study is needed to clarify how subcortical areas contribute to the overall decreased activity of CNS during motor fatigue state.

Keywords: motor fatigue, sensorimotor areas, thalamus, basal ganglia, fMRI

INTRODUCTION

The neural mechanism of motor fatigue is still unclear although this phenomenon has been studied for almost half a century (Bigland-Ritchie and Lippold, 1979). In the early works, most studies focused on the physiological changes of the muscles (Bigland-Ritchie et al., 1986; Gandevia, 1988; Pagala et al., 1991), such as run-down of the energy reserves, accumulation of blood lactate, unbalance of H⁺, as well as increased free radical content (St Clair Gibson et al., 2005; Reid, 2008). Therefore, motor fatigue was primarily referred to as muscular fatigue. However, accumulating evidences reveal that the supraspinal structures also exhibit some changes of activity under motor fatigue state, especially the motor cortex of the central nervous system (CNS; Taylor et al., 1996, 2006; Gandevia, 2001), whose output is not sufficient to drive the muscle maximally under motor fatigue state (Jones and Hunter, 1983; Enoka, 1995; Taylor et al., 2000; Amann and Dempsey, 2008).

Cortical sensorimotor areas play a dominant role in human motor control. Recent studies found that there was a linear relationship between neuronal activity in several brain areas (i.e., the primary motor cortex and the cerebellum) and the sum of EMG activity of various hand muscles in monkeys (Townsend et al., 2006; Bourguignon et al., 2013). Our previous human study also shows that the change of activation level in cortical brain areas is related to motor fatigue (Hou et al., 2012). Also, there is evidence showing that CNS has to increase its drive to relevant motor neuron pools or increase the firing frequency of the already active units to increase muscle force production, as is demonstrated by an increase in the electromyography (EMG) activity (Jones and Hunter, 1983).

The subcortical brain regions, such as the thalamus and the basal ganglia, play critical roles in the regulation of motor function (Buot and Yelnik, 2012). By using the electrophysiological recording technique, we observed the variation of neuron activity in striatum and found that the percentage of high frequency neurons is significantly increased, and the amounts of bursting style neurons are increased in striatum during motor fatigue (Qiao et al., 2010).

Inconsistent findings were reported regards change of brain activation in response to motor fatigue. Dettmers et al. (1996) show that motor fatigue does not induce changes in the activation intensity of the primary sensorimotor areas, SMA, or basal ganglia, while the only changed activation was observed in a small cluster in the dorsolateral prefrontal cortex. In contrary, other studies found increase in the number of activation voxels in the sensorimotor cortex, SMA, cerebellum, and dorsolateral prefrontal cortex in motor fatigue condition (Liu et al., 2003).

Brain image studies observed associations between motor fatigue and altered neural activation on the cortical level, indicating involvement of the CNS in motor fatigue (Gandevia, 2001; Taylor et al., 2006; Hou et al., 2012). To further our understanding of how the CNS acts in response to motor fatigue, the present study aims to examine how the activation of the sub-cortical areas changes when people experience exercise-induced motor fatigue. To this end, we recruited a group of healthy subjects to measure (1) the changes of activation in sub-cortical areas, i.e., thalamus and basal ganglia, when performing simple motor tasks before and under exhaustive exercise, which induces motor fatigue state with well-control procedures, and (2) the changes of activation in the sensorimotor areas, in which inconsistency observation of activation changes have been made.

MATERIALS AND METHODS

Subjects

Eleven healthy right-handed subjects participated in this study after signing informed consents (seven male, four female; age 23.2 ± 3.5 years). All subjects underwent the comprehensive verbal screening procedure to ensure that they do not violate any of the exclusive criteria for functional magnetic resonance imaging (fMRI) experiment: (1) history of neurological or

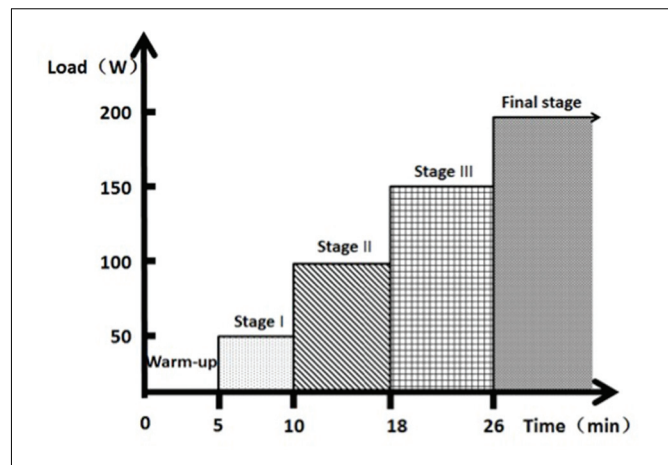
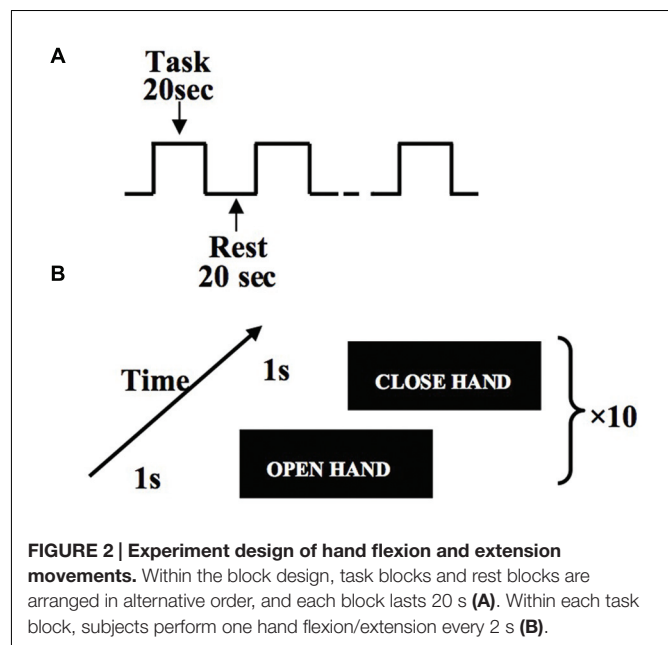


FIGURE 1 | Protocol of exhaustive exercise. The process begins from a 5 min warm-up, 5 min stage I, 8 min stage II, 8 min Stage III, and a certain duration of final stage according to the physical ability of the participants.



cardiovascular disease; (2) medications; (3) cochlear implants or any metal objects in the body; (4) cardiac or neural pacemakers; and (5) history of musculoskeletal injury in both lower limbs. The Ethics Committee of Beijing Normal University approves all experimental procedures.

Exhaustive Exercise Protocol

In order to reach the motor fatigue state, participants performed the exhaustive exercise protocols on a cycle ergometer (Monark 834 E, Sweden). Heart rate was measured every 5 s using short-range radio telemetry (S725XPolar, Finland). A portable lactate analyzer (YLS9-Lactate scout, German) was used to analyze all blood samples extracted from the finger sticks. Participants completed a 5-min warm-up with no resistance at a cadence

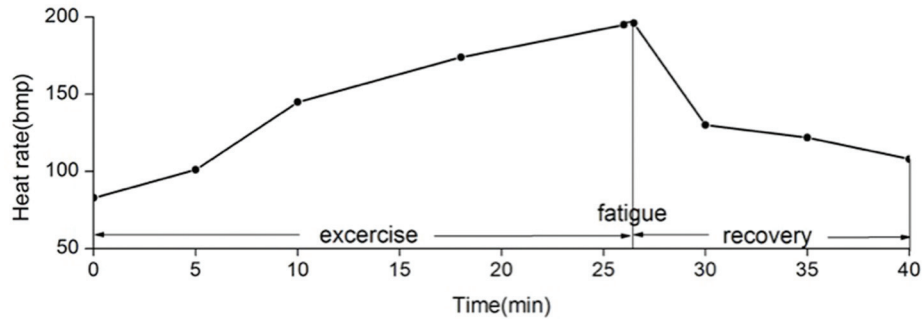


FIGURE 3 | Mean heart rate changes during and after exhaustive exercise. We define that the state of motor fatigue is the time point when the participant cannot maintain the cadence, and when heart rate arrives 90% of the maximum heart rate. In this experiment, participants reach the fatigue state after 26 min of exhaustive exercise in average.

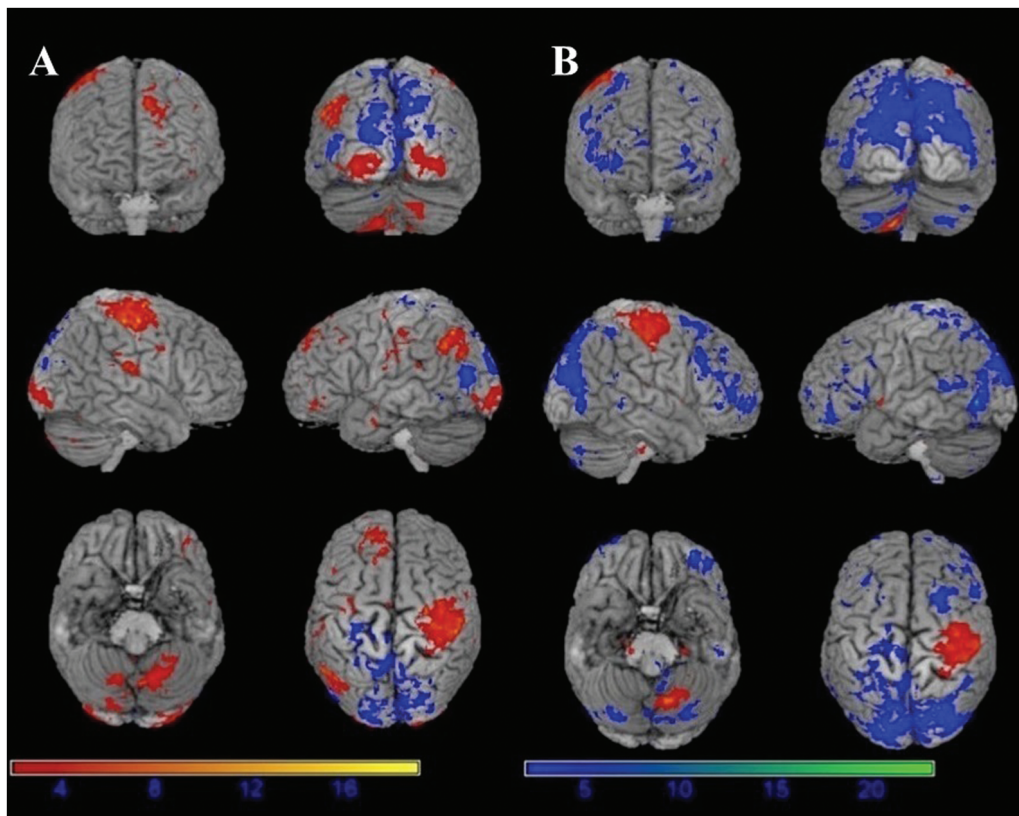


FIGURE 4 | Significant activation (red) and deactivation (blue) volumes when subjects are performing hand movement before (A) and during (B) motor fatigue. Color bar: *t*-values.

of 50 rev min⁻¹. At the conclusion of the warm-up, 1 kg of resistance is added, yielding a power output of 50 W. Power output increases to 100 W at the second stage, to 150 W for the third stage and 200 W for the final stage. Participants pedaled at a constant cadence of 50 rev min⁻¹ until reaching exhaustive state (Figure 1). The state of motor fatigue is defined as the time point when the participant cannot maintain the cadence and the heart rate arrives 90% of the maximum heart rate.

Experiment Design

After subjects reached the motor fatigue state, they were guided to the MRI scanner. Subjects performed hand flexion and extension movements according to the instructions reflected in the mirror from a computer screen. For every subject, we controlled the delay to be approximately equal between the exhaustive exercise and the MRI acquisition. Subjects were guided to the scanner right after the physical measurement. Each

TABLE 1 | Significant[†] activation volumes before and during motor fatigue state.

Region		Before			t	BA*	NbVx**		During			t	BA	NbVx
		x	y	z					x	y	z			
Sensorimotor	R	45	-15	57	17.65	4	72	R	42	-18	51	12.10	4	66
	R	51	-21	57	13.43	3,2,1	154	R	42	-24	57	9.17	3,2,1	136
SMA	R	6	-3	60	6.16	6	108	R	9	9	63	3.52	6	56
	L	-6	-3	60	7.47	6	77	L	-9	-3	63	4.03	6	42
PM	R	24	-12	69	16.19	6	260	R	36	-12	57	18.70	6	101
	L	-42	0	54	4.43	6	40							
Cerebelum_6	R	27	-57	-24	16.82	37	174	R	27	-63	-18	6.01	19	76
	L	-30	-51	-24	14.05	37	210	L	-9	-60	-15	10.27	18	171
Thalamus	R	18	-15	3	17.65	NA	171	R	6	-27	3	7.04	NA	82
	L	-9	-9	9	5.19	NA	72							
Putamen	R					NA	144	R					NA	85
	L					NA	72							
Caudate	R					NA	40	R					NA	9
	L					NA	84							
Pallidum	R					NA	25	R					NA	11
	L					NA	18							
Insula	R	44	5	6	3.39	48	65	R	45	3	6	9.53	48	61
	L	-44	-3	9	4.15	48	31	L	-42	-3	3	4.99	48	31
postcentral	L	-45	-21	33	7.61	3	107							
parietal_inf	L	-51	-21	42	6.99	40	23							
Rolandic_Oper								R	45	-3	9	2.93	48	56
								L	-45	-6	3	12.10	48	39
supraMarginal	L					40	68	R	48	-33	28	5.13	48	47
temporal_sup	L	-51	-30	21	7.34	48	39	R	57	-18	12	5.70	48	26
frontal_sup	L	-18	54	30		9	185							
frontal_sup_medial	L	-9	39	57		9/8/10	43							
Frontal_Mid_Orb	L	-39	48	-9		10	29							
Frontal_Inf_Orb	L					11	52							
Frontal_Inf_Tri	L	-45	39	6		46/47	25							
Temporal_Mid	L	-57	-3	-18		21	78							

[†]P < 0.05, cluster size > 54. *BA, Brodmann area, **Nb Vx, number of voxels. Abbreviations: SMA, supplementary motor area; PM, premotor area; inf, inferior; Oper, Operculum; sup, superior; Mid, middle; Orb, orbital; Tri, triangularis.

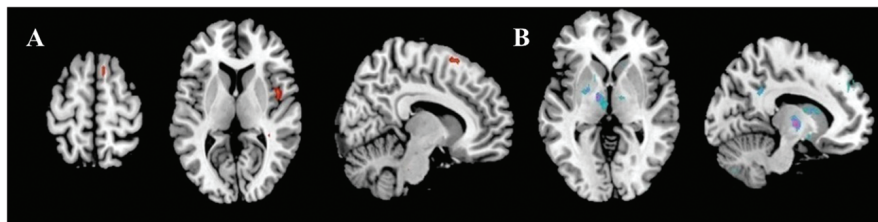


FIGURE 5 | Activation volumes show significant differences before and after exhaustive exercise. (A) After > before; (B) Before > after.

run of MRI scanning includes 10 trials with each trial lasting 20 s (Figure 2A). Within each task trial, there are 10 hand open and 10 hand close movements, which are guided by visual signals (Figure 2B).

MRI Data Acquisition

The MR protocol was performed using 3 Tesla whole-body system (Siemens, Erlangen, Germany) using blood oxygen

level-dependent (BOLD) fMRI. The head of the subject was immobilized using foam cushions and tape, with their ears plugged. The protocol included: (i) one sagittal T1-weighted image to localize functional and anatomical axial slices; (ii) 33 axial gradient echo-planar images (EPI; 3.8 mm, gap 0.7 mm, TR = 2 s, TE = 30 ms, bandwidth = 2520 Hz/pixel, Flip angle = 90°, FOV = 218 mm × 218 mm, in-plane resolution = 64 mm × 64 mm). The whole protocol lasts 40 min.

TABLE 2 | Brain volumes showing significantly[†] different level of activation before and under fatigue state.

	Region	Hemisphere	x	y	z	t
Before > after	Thalamus	L	-12	-9	0	4.66
	Striatum	L				
After > before	SMA	R	9	15	63	3.14
	Insula	R	45	3	6	4.17
	Hippocampus	R	33	-30	-6	3.29
	Cerebellum	R	39	-42	-33	3.48

[†]*P* < 0.05, cluster size > 54.

Functional MRI Data Analysis

Functional images were preprocessed using SPM5 software (Wellcome Department of Cognitive Neurology, London, UK) and Matlab. Before the preprocessing, the first five time points were discarded to avoid the disequilibrium in the magnetic field in the beginning of the scan. Images of each subject were realigned by using the first slice as the reference, and then normalized into the MNI space (Montreal Neurological Institute) using the template provided by SPM. At last, the 3 mm × 3 mm × 3 mm functional images were spatially smoothed with a Gaussian filter (4 mm × 4 mm × 4 mm full-width at half-maximum).

After the above preprocesses, functional images were first analyzed on the individual level to determine the activation volumes in response to hand movement. According to our block design, the boxcar waveform of the sustained activity of simple hand movement was convolved with the theoretical

hemodynamic response, for both before and after exhaustive exercise conditions. Then, on the group level, 2-tailed one sample *t*-test analysis was performed based on the outcome of each subjects' statistical parameter maps from the individual level analysis. Those volumes, which pass the significance threshold (*p* < 0.05) and had cluster size larger than 54 voxels, were reported as activation volumes. In the end, we compared the activation differences before and under motor fatigue state. Only the voxels that are activated in both before and under motor fatigue state conditions were included in this contrast. To do that, a union mask was generated from the significant positive activation maps of the two groups.

RESULTS

All subjects reached motor fatigue state after the exhaustive exercise (Figure 3).

Brain Activation Maps before and during Motor Fatigue State

The motor task evoked significant activations in the following brain areas in both before and under motor fatigue state conditions: the bilateral sensorimotor area, the premotor area, the supplementary motor area, the cerebellum, and the basal ganglia (Figure 4).

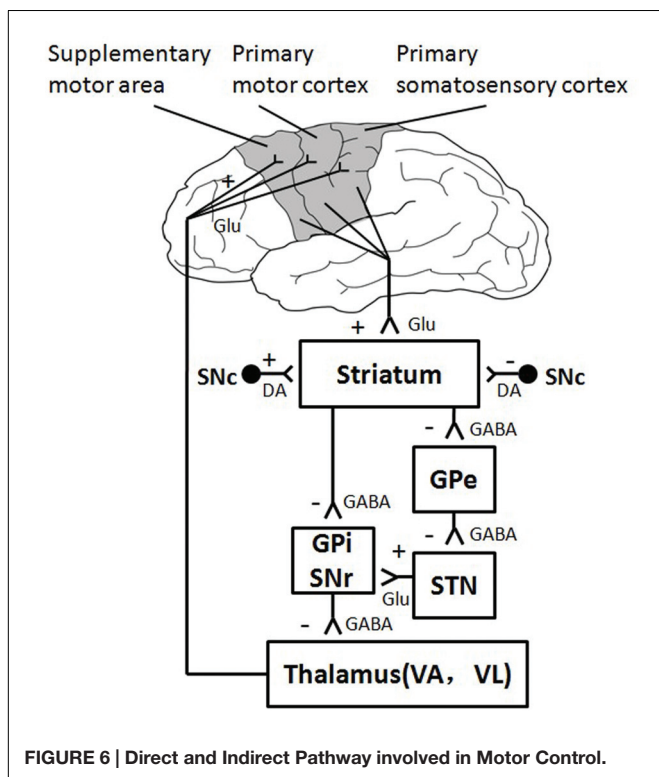
Differences on Activation Volumes before and under Motor Fatigue State

The results showed the intensity and size of activation volumes of ipsilateral thalamus and striatum significantly decreased during the motor fatigue state as compared to before the motor fatigue state. Moreover, the size of activation volumes in contralateral sensorimotor area was smaller (see in Table 1, sensorimotor area, Nb Vx), but there was no significant difference in the strength of activation (Figure 5; Table 2). It is worth noting that there was also difference between the extending of two activation maps. When the subjects perform the simple motor task under motor fatigue, the ipsilateral sensorimotor area was not as active as it was before, indicating the CNS tends to recruit less cortical and subcortical volumes when performing movements under motor fatigue.

DISCUSSION

From this experiment, we found that motor fatigue affects not only the neural excitability on the cortical level (i.e., sensorimotor cortex and the SMA), it affects also the subcortical brain regions (i.e., thalamus and striatum). This finding has not been reported systematically in previous studies.

Previous neurophysiological studies found that, when under motor fatigue, the output of cortical sensorimotor areas may not be adequate to drive the motor neurons to produce maximal movement force (Jones and Hunter, 1983; Enoka, 1995). This finding is supported by recent studies using transcranial magnetic stimulation (TMS), which find that some force loss in fatigue may



be due to inadequate descending drive from the motor cortex (Taylor and Gandevia, 2001). The present study found that neural activation in the sensorimotor cortex and the SMA declined when people under motor fatigue. This finding provides neural image supports for the notion that the cortical areas play role in motor fatigue.

The thalamus and the striatum are two important subcortical brain areas, which participate in human motor control by interacting with the cerebral cortex through parallel neural circuits (Alexander et al., 1986; Baston and Ursino, 2015). Recent diffusion weighted imaging and tractography study in human find a regional network among SMA, subthalamic nucleus (STN), and the inferior frontal cortex (IFC; Aron et al., 2007). Striatum, more specifically the STN, is the input station of the basal ganglia that receive excitatory afferent input from the cortical motor areas, such as the sensorimotor cortices and SMA (Prodoehl et al., 2009). The thalamus receives inhibitory signals from the output station (i.e., Gpi and SNr) of the basal ganglia, and it sends excitatory output to cerebral cortex in turn (Figure 6). The present study found decreased neural activation in the subcortical structures of thalamus and striatum, indicating that the subcortical areas involve in motor fatigue as well. In other words, motor fatigue seems to be associated with declined neural activation on both cortical and subcortical levels. Although the excitatory and inhibitory circuits between striatum and thalamus is beyond the exploration of this experiment such that we are not able to infer the temporary changes of activation from cortical region to thalamus, from above mentioned knowledge, we can still make the preliminary conclusion that motor fatigue state influences the neural substrates of simple grasp movement. This effect exhibits as an overall decrease of activation of the corticostriatal and cortico-thalamic pathways. It is highly possible that the decreased activation of sensorimotor areas may account for the decreased activation of striatum, and the decreased activation of thalamus may contribute to the loss of activation of sensorimotor areas.

We want to clarify that the present study did not monitor the force and movement frequency produced by finger movements due to technical limitation associated with MRI scanning. As a consequence, we cannot ascertain if the decreased cortical and subcortical activation observed would attribute to the possible reduction of motor performance. In other words, the present study found an association between motor fatigue and the CNS, while future studies are needed to further examine the proposed causal relationship between the less activated cortical and subcortical areas and the declined motor performance under fatigue state. We also want to point out that subjects were asked to evaluate their own performance in the study. In general, subjects reported to be able to follow the pace and range of the movement as required in both conditions. However, the self-report alone is not sufficient to present the real change of movement performance due to fatigue. Moreover, studies have

shown that there could be a motor fatigue associated over-estimation of force of muscle contraction (Jones and Hunter, 1983). Therefore, we still cannot rule out the possibility that the reduced activity on the cortical and subcortical level is partially responsible for reduced muscular contraction under fatigue.

Recent study had found that striatum is highly relevant with motor and cognitive function. Our knowledge of striatum's role in motor control mainly came from some common neurodegenerative disease, such as Parkinson disease (PD), Huntington disease (HD), Tourette syndrome (TS), etc. Patients with these diseases exhibit motor deficits and always show some structural abnormality of striatum. The disturbance of striatum activation during motor fatigue state might also interrupt with the motor and cognitive function; further experiment is needed to examine how motor fatigue affects motor and cognitive functions.

CONCLUSION

We used exhaustive physical exercise to induce motor fatigue state; then, under the effect of motor fatigue, the neural response of simple movements exhibiting dramatic differences compared with before the exhaustive physical exercise. We observed an overall decrease of activation in sensorimotor areas, SMA, as well as in the thalamus and basal ganglia.

It is well known that the cortical motor areas have anatomical connection with the subcortical brain regions, and these connections form complex excitatory and inhibitory circuits (Alexander et al., 1986). The interference of these neural circuits leads to various pathological conditions (Shepherd, 2013). The result from this experiment revealed for the first time the overall changes of these circuits in response to motor fatigue; however, the contribution of each brain region is still not clear. We propose that further study should be deployed in two directions, the role of basal ganglia in central motor fatigue and the effect of motor fatigue on patients with various movement disorders.

AUTHOR CONTRIBUTIONS

LH and JW conceived and designed the study. YY and JC performed the experiments. ZS and LH analyzed the data. ZS and ZP wrote the paper. LH, ZS, and ZP reviewed and edited the manuscript. All authors read and approved the manuscript.

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EEG Brain Activity in Dynamic Health Qigong Training: Same Effects for Mental Practice and Physical Training?

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In recent years, there has been significant uptake of meditation and related relaxation techniques, as a means of alleviating stress and fostering an attentive mind. Several electroencephalogram (EEG) studies have reported changes in spectral band frequencies during Qigong meditation indicating a relaxed state. Much less is reported on effects of brain activation patterns induced by Qigong techniques involving bodily movement. In this study, we tested whether (1) physical Qigong training alters EEG theta and alpha activation, and (2) mental practice induces the same effect as a physical Qigong training. Subjects performed the dynamic Health Qigong technique *Wu Qin Xi* (five animals) physically and by mental practice in a within-subjects design. Experimental conditions were randomized. Two 2-min (eyes-open, eyes-closed) EEG sequences under resting conditions were recorded before and immediately after each 15-min exercise. Analyses of variance were performed for spectral power density data. Increased alpha power was found in posterior regions in mental practice and physical training for eyes-open and eyes-closed conditions. Theta power was increased after mental practice in central areas in eyes-open conditions, decreased in fronto-central areas in eyes-closed conditions. Results suggest that mental, as well as physical Qigong training, increases alpha activity and therefore induces a relaxed state of mind. The observed differences in theta activity indicate different attentional processes in physical and mental Qigong training. No difference in theta activity was obtained in physical and mental Qigong training for eyes-open and eyes-closed resting state. In contrast, mental practice of Qigong entails a high degree of internalized attention that correlates with theta activity, and that is dependent on eyes-open and eyes-closed resting state.

Keywords: Health Qigong, dynamic Qigong, mental practice, relaxation, EEG

INTRODUCTION

Eastern meditation techniques are a common integral part in everyday life as a means to alleviate working stress, and fostering an attentive mind. One main aim is to enable meditation practitioners to lead a more conscientious style of life. Besides Hindu meditation techniques as different styles of Yoga, Buddhist and Taoist meditation such as Qigong experiences an increasing number of practitioners in the Western hemisphere. Qigong comprises several techniques applied

in Traditional Chinese Medicine (TCM) to strengthen physical and mental health. Qigong is commonly divided into static and dynamic forms. Static forms contain meditational techniques whereas dynamic forms afford bodily movements as a tool to direct practitioners' attention (Tsang et al., 2002; Shinnick, 2006). Research on meditative Qigong practice demonstrates beneficial effects on health (for an overview see Ng and Tsang, 2009). A beneficial influence of Qigong practice on the cardiovascular system, i.e., on blood pressure (Xu, 1994; Lee et al., 2003; Cheung et al., 2005), and electrocardiographic parameters (Lee et al., 2000, 2003) was investigated. Furthermore, Qigong meditation lead to changes in breathing frequency (Sun, 1988). Positive effects of Qigong practice on mental health could be demonstrated in major depression (Tsang et al., 2003; Wang C.W. et al., 2013; Wang F. et al., 2013; Yeung et al., 2013; Yin and Dishman, 2014; Liu et al., 2015; Martinez et al., 2015), anxiety disorders (Lee et al., 2004a,b; Abbott and Lavretsky, 2013; Chan et al., 2013), post-traumatic disorders (Grodin et al., 2008; Kim et al., 2013), in the burnout syndrome (Stenlund et al., 2009, 2012), and in tinnitus (Biesinger et al., 2010). In several studies, a stress alleviating and relaxing effect in healthy subjects (Lee et al., 2005; Posadzki et al., 2010; Terjestam et al., 2010; Gleib et al., 2012; Sousa et al., 2012; Hwang et al., 2013; Shim, 2014; Wang et al., 2014) was shown.

An essential research question is how the beneficial effects of Qigong meditation on physical and mental health are mediated by neurophysiological processes. Several studies applying electroencephalography (EEG) and fMRI demonstrated changes in brain activity induced by Qigong meditation. Most studies report increases in theta and alpha activity after Qigong meditation. The first studies on the effect of Qigong meditation on electrical brain activity reported alpha activity predominantly in the anterior brain regions (Wallace, 1970). More differentiated results of effects of meditational Qigong techniques on EEG activity are shown dependent on expertise level. Shifts in alpha activation were observed from posterior to anterior regions during Qigong meditation (Zhang et al., 1988a,b; Jang et al., 2004; Qin et al., 2009). Yang et al. (1994) obtained effects of Zhanzhuang Qigong on brain activity. After 1 year of practice, the alpha activity of the right frontal and right temporal regions increased significantly. The beta index of the right frontal and right temporal regions decreased significantly. A synchronization of brain activity was shown. The effects did not occur after half a year of regular Qigong meditation practice. Therefore, it was assumed to be a gradually adjusting process.

Psychophysiological states of wakefulness and arousal as measured in terms of activation of particular EEG frequency bands are commonly correlated with distinct self-reported experiences of the Qigong state in regular meditators. Mostly, an increase of alpha activity is related to an experience of relaxation and increased well-being. An increase of EEG frontal theta activity is correlated to a self-report of mindfulness, an attentive state which is one of the main aims to reach in Buddhist meditation techniques (for an overview see Tomasino et al., 2014). How are changes in brain activity induced by Qigong meditation correlated to the reported psychophysiological states of relaxation, attentiveness, and attentional processing? In a recent study, EEG alpha-2 activity in posterior right parietal

Brodmann areas 5, 7, 31, and 40 during Qigong meditation was demonstrated (Faber et al., 2012). The authors argue that the found patterns of brain activation reflect self-reference, attention and input-centered processing in Qigong meditation. Pan et al. (1994) identified frontal mid-line theta rhythm during the concentrative Qigong state compared to the state of mind reached by non-concentrative Qigong engagement. Shim (2012) found theta activity centering around the frontal lobe parts in Qigong masters and decreased alpha activity compared to beginners. The authors argue that Qigong experts maintained more deeply internalized and relaxed theta activity in the frontal lobe, which reflects an attentive mind. Qigong masters show efficiency in keeping a relaxed and attentive mind around central midline. Lee et al. (1997) investigated effects of ChunDoSunBup Qi-training on brain activity. The Qi-training consisted of acoustic exercises, bodily motion, and meditation. Increases in alpha activity in ChunDoSunBup Qi-training were observed in the occipital regions in eyes-open conditions. The increase in occipital alpha activity was correlated with less self-reported state anxiety. The authors argue that in ChunDoSunBup Qi-training activity of the occipital cortex is reduced and the thalamus is influenced.

On a more structural level, Lehmann et al. (2012) showed reduced functional connectivity between cortical sources in Qigong meditation and reduced functional interdependence between brain regions. These results were interpreted to be a correlate of the reported subjective experience of non-involvement, detachment and letting go, as well as of all-oneness and dissolution of ego borders during Qigong meditation. Cheng et al. (2010) showed an effect of Qigong meditation on prefrontal activity. Practitioners showed in comparison to non-practitioners a significant decrease in deoxyhemoglobin levels suggesting an increase in prefrontal activation during Qigong meditation. Two fMRI studies report changes in brain activity under the state of Qigong during pain exposure in Qigong masters correlating with reduced pain sensation (Chan et al., 2006; Yu et al., 2007). Functional activation in the SII-insula region and other brain areas was reported, whereas a functional suppression under the state of Qigong meditation was observed. Thus, the found functional suppression in brain regions may be responsible for the reduced pain sensation in Qigong masters under the Qigong state.

In conclusion, systematical effects of Qigong meditation on EEG brain activity can be stated with most studies reporting increases in frontal theta and posterior alpha activity as a neurophysiological correlate for a relaxed and attentive mind.

To our knowledge, there are no systematical studies reported in the Western hemisphere on effects on brain activity of dynamic Qigong techniques that afford bodily movement. For instance, the Health Qigong technique *Wu Qin Xi* comprises a consecutive sequence of complex movement configurations. These configurations are symbolic exposures of five animals (tiger, deer, bear, monkey, bird) with each movement sequence performed for several minutes. Practitioners are requested to focus on breathing when performing the movement sequences. According to theoretical assumptions of TCM, *Wu Qin Xi* is an intervention to strengthen especially physical health in general

(for an overview see Yang and Wu, 2011). Only a few studies have been reported in the Western hemisphere on the dynamic Qigong technique *Wu Qin Xi* on physical health. Positive effects of *Wu Qin Xi* training are reported on lumbar spinal disease (Yeom et al., 2013; You et al., 2013, 2014; Zhang et al., 2014), blood lipid levels and the antioxidant enzyme activities (Chen, 2011). To date, no studies have been reported in the Western hemisphere on effects of *Wu Qin Xi* on neurophysiological parameters.

In previous experimental studies conducted in our working group, increases in midline fronto-central theta and shifts in alpha activity from posterior to anterior regions over the whole scalp after physical training of the dynamic Qigong technique *Wu Qin Xi* were observed (Henz et al., 2013, 2014, 2015; Henz and Schöllhorn, 2015). From a qualitative point of view, the found brain activation patterns were in line with findings of studies conducted with meditational (static) Qigong. As in studies on meditational Qigong, increases in fronto-central theta and posterior alpha activity were obtained. Thus, a relaxing effect in sense of an evidence-based approach can be stated for the dynamic Qigong technique *Wu Qin Xi*.

As dynamic Qigong affords series of complex bodily movements, practitioners have to invest high effort and attention to learn the new movements. One benefit of that is that directing attention toward the movement execution and kinaesthetic sensations is intended to result in a centered state of internalized concentration. This mechanism plays a key role in mind-body therapies such as dynamic Qigong to draw the attention away from the everyday mind flow to reach an attentive state (for an overview, see Schmalzl et al., 2014).

In the present study, we tested whether the beneficial effects of physical practice of dynamic Qigong on EEG brain activity can be reached by practicing the dynamic Qigong technique *Wu Qin Xi* mentally. From a theoretical point of view, this question is relevant because a large number of brain mapping studies have shown that the same neural areas are activated during either physical or mental simulation of motor actions. The rationale behind is that mental practice with motor content engages areas of the brain that govern movement execution (for an overview see Cicinelli et al., 2006; Sharma and Baron, 2013). This was not only demonstrated for cortical areas such as the supplementary area, the premotor cortex, and the primary motor cortex, but also for subcortical areas such as the basal ganglia and the cerebellum (Lotze et al., 1999; Jeannerod, 2001; Lafleur et al., 2002; Munzert et al., 2008). From a practical point of view, mental Qigong practice becomes relevant for the everyday practitioner in situations when the physical practice of Qigong is impracticable. I.e., when waiting at the train station surrounded by many other passengers it is not possible to practice dynamical Qigong physically due to a limited physical space. Further, this research question is relevant for designs of Qigong courses for elderly persons or patients with bodily impairments. Especially at the beginners' stage, the movement sequences are practiced many times with several hours of practicing physically leading to increased physical tiredness. Here, the research question arises whether intervals of mental practice sessions would have the same effect on EEG brain activity as a merely physical Qigong practice. Finally, mental Qigong practice becomes relevant especially in

persons who do not have the possibility to engage in Qigong training physically, either on the short-term, or on the long-term perspective. For instance, in stroke patients who have a low capability to move or in patients with chronically relapsing diseases of the musculoskeletal system mental practice of *Wu Qin Xi* would be a suitable therapeutic intervention. In achievement sports, when athletes experience phases of immobility due to sports concussions, mental practice of *Wu Qin Xi* Qigong would be an appropriate alternative to physical training.

Recent research has shown that mental practice in the form of motor imagery causes comparable patterns of brain activation as physical training of the same movement. From this, we suppose that mental practice of the dynamic Qigong technique *Wu Qin Xi* leads to comparable effects in EEG brain activation as in physical Qigong training. More precisely, in line with the findings of previous studies on Qigong meditation, and on the dynamic Qigong technique *Wu Qin Xi* on EEG brain activity we hypothesize an increase in EEG midline fronto-central theta and posterior alpha activity when practicing the dynamic Qigong technique *Wu Qin Xi* mentally.

The aims of the present study are as follows:

- (1) The analysis of the spontaneous eyes-closed and eyes-open EEG spontaneous activity after training of the dynamic Qigong technique *Wu Qin Xi*.
- (2) Comparison of effects of mental practice and physical training of the dynamic Qigong technique *Wu Qin Xi* on the spontaneous eyes-closed and eyes-open EEG brain activity.

MATERIALS AND METHODS

Participants

Twenty-five subjects (mean age 27.9 years, $SD = 2.91$; age range: 19–47; 12 males, 13 females) volunteered in this study. Subjects were recruited from the Qigong workshops at the Institute of Sports Science of the University of Mainz and from sports science courses. Inclusion criteria for the study were participation in a Qigong workshop (30 h of lessons) and at least 1 h practice per week for 1 year. Regular Qigong practice was assessed prior to the experiment by a questionnaire. The subjects were all healthy, and had no current diseases or a history of neurological impairments or intake of medication that may have affected EEG recordings. All subjects were naïve as to the purpose of the current study. All subjects gave written informed consent. The experimental procedures were approved by the local ethics committee at the Johannes Gutenberg University of Mainz, Germany. All experimental procedures were carried out in accordance with the Declaration of Helsinki.

Experimental Procedure

The subjects were sat comfortably in a dimly-lit isolated room. At each measurement time point, participants began with a resting condition. Spontaneous EEG of the subject was recorded for 2 min for eyes-open, and 2 min for eyes-closed conditions. Then, they were required to perform a 30-min training session.

The experiment contained three training conditions: Participants were required to perform the dynamic Qigong technique *Wu Qin Xi* (five animals) physically, and mentally in a within-subjects design. In the mental training condition, subjects were asked to perform the movement sequence mentally from the ego-perspective with imagination of kinaesthetic and visual cues. Further, they were required to apply the same breathing technique in the mental practice condition as in physical training. Additionally, a control condition was tested where subjects were presented a video showing practitioners performing the Qigong exercise *Wu Qin Xi*. All participants were familiar with mental practice of the Qigong technique *Wu Qin Xi*. Experimental conditions were randomized. All training sessions, and the control session were performed with eyes-open. EEG data were obtained during the four resting conditions: (1) pretraining rest, (2) post-Qigong training rest, (3) post-mental Qigong training rest, (4) post-video control rest, which were then used for subsequent analyses.

EEG Data Acquisition and Analysis

Electroencephalography was recorded through the Micromed Brain quick amplifier and Micromed Brainspy software (Micromed, Venice, Italy). Recordings were taken from nineteen electrodes (Fp1, Fp2, F3, F7, Fz, F4, F8, C3, Cz, C4, T3, T4, P3, P7, Pz, P4, P8, O1, O2) placed according to the Int. Ten to twenty systems with reference to the nose. All electrode impedances were kept at 10 k Ω or below. The EEG signals were continuously recorded and digitized at a sampling rate of 256 Hz. The EEG signal was amplified with a fixed time constant of 0.3 s with a Butterworth second order high-pass filter at 0.5 Hz, and a low-pass filter at 120 Hz (frequency range: 0.5–120 Hz). Electrooculography (EOG) was monitored placed at the medial upper and lateral orbital rim of the right eye (time constant: 0.3 s; high-pass filter: 0.1 Hz; low-pass filter: 120 Hz; frequency range: 0.5–120 Hz).

The spontaneous EEG was recorded for 2 min with eyes-closed, and 2 min eyes-open conditions. Subsequent analyses were performed separately for eyes-closed, and eyes-open conditions. The EEG and EOG signals were visually scored and portions of the data that contained aberrant eye movements, muscle movements of artifacts were removed. The EEG was analyzed and Discrete Fast Fourier Transform was used to obtain the mean power amplitudes in theta (4–7.5 Hz), low-frequency alpha-1 (8–10 Hz), high-frequency alpha-2 (10–12.5 Hz), beta (13–30 Hz), and gamma (30–40 Hz) bands. The ranges of high- and low-frequency alpha bands were defined according to previous studies by Aeschbach et al. (1999) and Cantero et al. (2002).

Statistical Analyses

A statistical comparison of power of theta, alpha-1, alpha-2, beta, and gamma bands was calculated by repeated-measure analyses of variance (ANOVA) including the within-subject factors as training condition (physical Qigong training, mental Qigong training, video control, baseline rest), condition (eyes-open, eyes-closed), and location (Frontal, Central, Temporal, Parietal, Occipital), followed by Bonferroni corrected *post hoc* tests for

further comparisons. η_p^2 was calculated to obtain effect sizes. Effects were considered to be statistically significant when the *p*-values were less or equal than 0.05. All data are expressed as the mean \pm S.E.

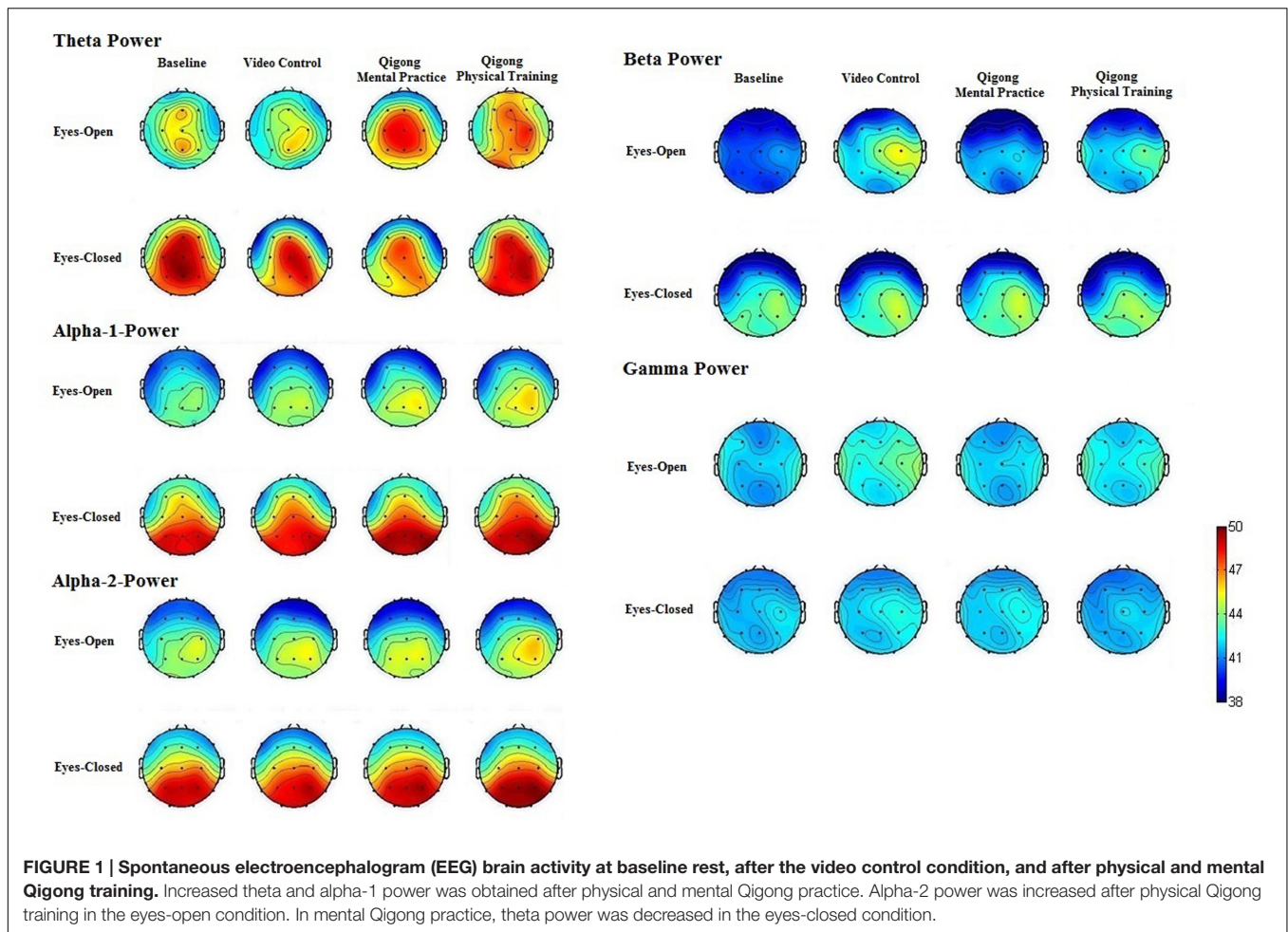
RESULTS

Statistical Description: Spontaneous EEG

Figure 1 shows the mean power spectra for the theta, alpha-1, alpha-2, beta, and gamma band. The ANOVA of theta responses revealed significant differences for training, $F(3,72) = 3.77$, $p = 0.014$, $\eta_p^2 = 0.13$. *Post hoc* comparisons showed that the spontaneous EEG theta power was significantly increased after physical and mental Qigong training compared to the video control condition, $p = 0.03$, and baseline rest, $p = 0.01$. No significant difference was obtained between physical and mental Qigong practice. The ANOVA of theta responses revealed highly significant differences between conditions (eyes-open, eyes-closed), $F(1,24) = 8.92$, $p = 0.006$, $\eta_p^2 = 0.27$. The ANOVA of theta responses revealed significant results for training \times condition, $F(3,72) = 3.95$, $p = 0.012$, $\eta_p^2 = 0.14$. *Post hoc* comparisons showed that in mental practice theta activity was decreased in the eyes-closed condition compared to physical training, $p = 0.007$, video control, $p = 0.021$, and baseline rest, $p = 0.015$. The ANOVA of theta responses revealed significant differences between locations, $F(4,96) = 3.41$, $p = 0.015$, $\eta_p^2 = 0.12$. *Post hoc* comparisons showed that spontaneous EEG theta power was increased at frontal, $p = 0.017$, central, $p = 0.009$, parietal electrodes, $p = 0.021$, and occipital electrodes, $p = 0.007$.

The ANOVA of alpha-1 responses revealed highly significant differences for training, $F(3,72) = 4.34$, $p = 0.007$, $\eta_p^2 = 0.15$. *Post hoc* comparisons showed that the spontaneous EEG alpha-1 power was significantly increased after physical and mental Qigong training compared to the control condition, $p < 0.01$ each, and baseline rest, $p < 0.01$ each. No significant difference was obtained between physical and mental Qigong practice. The ANOVA of alpha-1 responses revealed highly significant differences between experimental conditions (eyes-open, eyes-closed), $F(1,24) = 14.088$, $p = 0.001$, $\eta_p^2 = 0.370$. The ANOVA of alpha-1 responses revealed no significant results for the factor training \times condition. The ANOVA of alpha-1 responses revealed significant differences between locations, $F(4,96) = 4.437$, $p = 0.002$, $\eta_p^2 = 0.156$. *Post hoc* comparisons showed that spontaneous EEG alpha-1 power was higher at central, and parietal electrodes than that of frontal, $p < 0.05$ each, temporal, $p < 0.05$ each, and occipital electrodes, $p < 0.05$ each.

The ANOVA of alpha-2 responses revealed significant differences for training, $F(3,72) = 3.30$, $p = 0.025$, $\eta_p^2 = 0.12$. *Post hoc* comparisons showed that the spontaneous EEG alpha-2 power was significantly higher after physical Qigong training compared to mental practice, $p = 0.03$, video control,



$p = 0.02$, and baseline rest, $p = 0.01$. No difference was obtained between mental practice, video control, and baseline rest. The ANOVA of alpha-2 responses revealed highly significant differences between experimental conditions (eyes-open, eyes-closed), $F(1,24) = 13.38$, $p = 0.012$, $\eta_p^2 = 0.36$. The ANOVA of alpha-2 responses revealed no significant results for training \times condition. The ANOVA of alpha-2 responses revealed significant differences between locations, $F(4,96) = 3.93$, $p = 0.005$, $\eta_p^2 = 0.14$. *Post hoc* comparisons showed that the spontaneous EEG alpha-2 power at central, and parietal electrodes was higher than that of frontal, $p < 0.05$ each, temporal, $p < 0.05$ each, and occipital electrodes, $p < 0.05$ each.

The ANOVA of beta responses revealed significant differences for training, $F(3,72) = 3.02$, $p = 0.033$, $\eta_p^2 = 0.11$. *Post hoc* comparisons showed that the spontaneous EEG beta power was increased in the video control condition, compared to mental practice, $p = 0.04$, physical training, $p = 0.02$, and resting baseline, $p = 0.03$. Significant differences were found between experimental conditions (eyes-open, eyes-closed), $F(1,24) = 4.66$, $p = 0.041$, $\eta_p^2 = 0.16$. The ANOVA of beta responses revealed significant results for training \times condition, $F(3,72) = 3.17$, $p = 0.030$, $\eta_p^2 = 0.12$. The ANOVA of beta responses

revealed significant differences between locations, $F(4,96) = 2.74$, $p = 0.033$, $\eta_p^2 = 0.10$. *Post hoc* comparisons showed that the spontaneous EEG beta power at central electrodes was higher than that of frontal, $p = 0.03$, temporal, $p = 0.04$, parietal, $p = 0.04$, and occipital electrodes, $p = 0.03$.

The ANOVA of gamma responses revealed significant differences for training, $F(3,72) = 3.43$, $p = 0.022$, $\eta_p^2 = 0.13$. *Post hoc* comparisons showed that the spontaneous EEG gamma power was increased in the video control condition, than in mental, $p = 0.03$, and physical Qigong practice, $p = 0.02$, and at baseline rest, $p = 0.04$. Significant differences were found for experimental conditions (eyes-open, eyes-closed), $F(1,24) = 5.45$, $p = 0.028$, $\eta_p^2 = 0.19$. The ANOVA of gamma responses revealed significant results for training \times condition, $F(3,72) = 3.04$, $p = 0.034$, $\eta_p^2 = 0.11$. *Post hoc* comparisons showed that gamma activity was increased in the video control condition in eyes-open compared to eyes-closed condition, $p = 0.031$. The ANOVA of gamma responses revealed significant differences between locations, $F(4,96) = 2.83$, $p = 0.029$, $\eta_p^2 = 0.11$. *Post hoc* comparisons showed that spontaneous EEG gamma power at temporal electrodes was increased compared to frontal, $p = 0.02$, central, $p = 0.03$, parietal, $p = 0.03$, and occipital electrodes, $p = 0.03$.

DISCUSSION

The literature includes several previous investigations on effects of Qigong meditation on EEG brain activity. Most studies report an increase in EEG frontal theta and shift of alpha activity from posterior to anterior regions during and after Qigong meditation. In the present study, we demonstrate an increase of midline fronto-central theta and posterior alpha-1 and alpha-2 activity after practice of the dynamic Qigong technique *Wu Qin Xi*. The finding of increases in midline fronto-central theta and shifts in alpha-1 and alpha-2 activity from posterior to anterior regions after physical training of the dynamic Qigong technique *Wu Qin Xi* was replicated (Henz et al., 2013, 2014, 2015; Henz and Schöllhorn, 2015). Further, our results mirror the findings of previous studies of effects on EEG brain activity after Qigong meditation demonstrating a shift of EEG activity from posterior to anterior regions (Zhang et al., 1988a,b; Yang et al., 1994; Litscher et al., 2001; Jang et al., 2004; Tei et al., 2006a,b, 2009; Qin et al., 2009; Faber et al., 2012). Therefore, a comparable effect for dynamic Qigong on EEG brain activity as found in studies on meditational Qigong can be stated. Training of the dynamic Qigong technique *Wu Qin Xi* induces a relaxed and attentive mind as indicated by an increase in midline fronto-central theta and shifts in alpha activity from posterior to anterior regions. In conclusion, a relaxing effect of the dynamic Qigong technique *Wu Qin Xi* in sense of an evidence-based approach can be stated. Our results indicate that the dynamic Qigong technique *Wu Qin Xi* induces a centered state of mind that has to be distinguished from mind-wandering. Empirical evidence is shown that frontal EEG theta activity is activated in attentional processes and correlates negatively with the default mode network in resting state (Scheeringa et al., 2008).

The highlighted finding of this study is that mental practice of the dynamic Qigong technique *Wu Qin Xi* causes significant modulations of EEG brain activity. Practicing the dynamic Qigong technique *Wu Qin Xi* mentally results in increased fronto-central midline theta activity and increases in alpha-1 power in the same intensity as in physical training in eyes open-conditions. Therefore, mental practice of the dynamic Qigong technique *Wu Qin Xi* has the same effect on EEG brain activity as physical training considering the eyes-open condition.

In the present study, the training by condition interaction with respect to changes in the theta band was replicated. In a previous study, it was shown that after training of the dynamic Qigong technique *Wu Qin Xi* theta activity was increased in eyes-open conditions whereas in the eyes-closed condition theta activity was diminished (Henz et al., 2013). In the current study, the same pattern of results with a training \times condition interaction was demonstrated. Our results are in line with a study conducted by Aftanas and Golocheikine (2001) who examined EEG brain activity in meditation in eyes-open and eyes-closed conditions. Depending on eyes-open and eyes-closed conditions, different patterns of anterior and midline theta activity occurred. The authors argue that the obtained theta activity reflects internalized attentional processes during meditation that are dependent on eyes-open and eyes-closed states.

The found patterns of brain activations are partially in line with our hypotheses. Our speculation was that based on findings from EEG studies on motor imagery (i.e., Jeannerod, 2006) nearly same effects on EEG brain activity would occur in mental as well as in physical training of the Qigong technique *Wu Qin Xi*. Having a closer look at theta activity, a decrease in mental practice in the eyes-closed condition compared to physical training, and the remaining conditions was observed. Therefore, our results indicate different underlying cognitive and neurophysiological processes in mental and physical *Wu Qin Xi* Qigong training. We hypothesized that different attentional processes during mental and physical *Wu Qin Xi* Qigong training play an essential part that lead to the obtained brain activation patterns. In previous studies on Qigong meditation, frontal and central midline theta activity was associated with internalized attentional processes. For instance, frontal mid-line theta rhythm during the concentrative Qigong state compared to the state of mind reached by non-concentrative Qigong meditation was shown by Pan et al. (1994). In the same manner, Shim (2012) demonstrated frontal theta activity after Qigong meditation in experienced practitioners. In previous studies on Qigong meditation, spontaneous resting EEG was measured with eyes-open. Therefore, no comparison between eyes-open and eyes-closed resting state EEG after Qigong meditation was done although this might allow insights into the underlying attentional processes. For instance, recent studies revealed an association between theta power and switching of involuntary attention from internally directed attention specific to the eyes-closed state to externally directed attention specific to the eyes-open state (Boytsova and Danko, 2009). Specific for meditational states, Aftanas and Golocheikine (2001) demonstrated effects of eyes-open resting state, and eyes-closed resting state on EEG theta activity as a correlate for internalized attentional processes.

One line of argumentation to explain the obtained results for modulations of theta activity dependent on eyes-open and eyes-closed state could be that practicing Qigong physically requires the subjects to strongly direct their attention to the performance of the complex movement sequences and the resulting kinaesthetic sensations as it is conceptualized in mind-body therapies such as Qigong. One main aim is to draw the attention away from the everyday mind flow to reach an attentive state (for an overview, see Schmalzl et al., 2014). As a consequence, a state of deep relaxation is reached in physical training mirroring in increased theta activity in eyes-open as well as eyes-closed conditions.

In physical *Wu Qin Xi* training, many details of the complex movement sequences have to be considered during movement performance, which might lead to a strong internalized attentional processing. From studies on the role of attentional focus during movement performance it is known that an external focus of attention alleviates movement performance, and therefore requires less effort, whereas an internal focus of attention requires more attentional demands. For instance, it has been shown that movement performance benefits from an external focus of attention in gymnastics (Abdollahipour et al., 2015). Several recent studies have provided evidence that movement efficiency, or the physical effort exerted to produce

a given performance level or outcome, is also enhanced by an external focus (Zachry et al., 2005; Marchant et al., 2009). Benefits of directing external focus have been found to result in more effective motor performance than those inducing an internal focus by directing attention to the body movements themselves (Totsika and Wulf, 2003; Wulf et al., 2003, 2009; Wulf, 2007). It is argued that focusing on the intended movement effect facilitates the utilization of unconscious or automatic processes, resulting in greater movement ease or fluidity (Wulf et al., 2001; Wulf and Lewthwaite, 2010). Conversely, focusing on one's own movements leads to a more conscious type of control, thereby constraining the motor system and disrupting automatic control processes (Wulf et al., 2001). It has been shown that relative to an internal focus, an external focus reduces attentional demands and results in the utilization of fast reflexive (automatic) feedback loops (Wulf et al., 2001). Transferring these findings on the physical performance of dynamic Qigong technique *Wu Qin Xi*, induction of internalized attention might be best reached with an attentional demanding complex motor task as in the movement sequences of *Wu Qin Xi*.

From this point of view, an important question arises: does an internal focus of attention during Qigong practice lead to a more demanding type of movement control, and therefore binds more attention which finally results in increased EEG theta and alpha activity? Especially in non-expert practitioners, demanding monitoring processes of movement performance during Qigong practice could result in enhanced stress reduction mirrored by increased EEG theta and alpha activity. We argue that one underlying cognitive mechanism is a working memory load which results from increased motor affordances. From a neurophysiological point of view, frontal theta power has been found to increase with working memory load (Gevins et al., 1997; Krause et al., 2000; Jensen and Tesche, 2002; Onton et al., 2005). Challenging working memory finally results in a loss of a merely executive action control due to limited capacity. Subsequently, practitioners' attention is drawn away from cognitive engagement in everyday thoughts by a demanding monitoring process. Additionally, a loss of cognitive action control toward a state of non-focusing and non-involvement on the everyday mind flow is one of the main aims in Eastern meditation techniques. Especially in Buddhism-related meditation traditions a mindfulness state is reached by sustained attention on the body. Activations in midline fronto-central lobe structures associated with attentional processes possibly confirm the fundamental role of mindfulness shared by many Buddhist meditations (for an overview see Tomasino et al., 2014). The finding of increased theta activity after physical Qigong training in eyes-closed conditions in our study indicates that internalized attention might be reached more easily when attention and breathing behavior is guided by movements in Qigong training.

A second line of argumentation is that breathing behavior in physical practice underlies a tighter regulation and a stronger forcing due to a coupling to the movement sequences of *Wu Qin Xi* than in mental practice. The breathing technique is strongly determined in the movement sequences of *Wu Qin Xi*, which might lead to the observed increase in low frequencies in the EEG in physical training due to a strong reinforcement

by the movement sequences. Recent EEG studies have shown that abdominal breathing techniques lead to increased frontal theta activity (e.g., Yu et al., 2011; Chervin et al., 2012; Park and Park, 2012). Considering breathing as a meditation technique, it was observed that Shaolin Dan Tian Breathing increases EEG frontal theta activity (Chan et al., 2011). The authors argue that the observed increase in frontal theta activity in Shaolin Dan Tian Breathing is a correlate for an attentive mind. Further, several studies have shown that abdominal breathing enhances EEG alpha activity. For instance, Arambula et al. (2001) demonstrated increases in EEG alpha activity in abdominal breathing techniques. Increased alpha band activity with decreased theta band activity was achieved by abdominal breathing during Zen practice (Arita, 2012). Comparing alpha-1 and alpha-2 activity Fumoto et al. (2004) showed increases in alpha-1 activity with disappearance of alpha-2 activity in voluntary abdominal breathing. Therefore, a stronger reinforcement of breathing behavior by movement performance in physical training would be a suitable interpretation for the obtained pattern of EEG brain activity. This might explain a diminished theta activity in mental practice in the eyes-closed condition compared to physical training.

A third line of argumentation considers the role of visual processing during mental practice. Dimitriadis et al. (2015) showed modulations in theta activity as a correlate for mental workload in visual processing. Transferring these findings on mental practice of *Wu Qin Xi*, the decreased theta activity in eyes-closed conditions might reflect attentional demands of visual processing of the movement sequence.

To our knowledge, the present study is the first one that compares effects of mental and physical dynamic Qigong training on EEG brain activity in eyes-open and eyes-closed conditions. From this, we supposed that mental practice of the dynamic Qigong technique *Wu Qin Xi* leads to comparable effects in EEG brain activation than in physical Qigong training. Having a closer look at theta activity in mental practice, a centering around the central areas compared to activation of a broader range of locations after physical training in eyes-open conditions was obtained. From this, we conclude different attentional processes in mental and physical *Wu Qin Xi* Qigong training.

The results of our study have important implications for the design of interventions applying the dynamic Qigong technique *Wu Qin Xi*. Especially in clinical populations who display reduced spontaneous alpha activity as in stress mediated diseases like burnout (see van Luitjelaar et al., 2010), but as well as in anxiety, depression, and bipolar disorders a strong induction of alpha activity by Qigong practice is essential for the therapeutic success of the intervention. With the results of the current study we showed that physical as well as mental training of the Qigong technique *Wu Qin Xi* lead to significant increases in low frequencies in spontaneous EEG. Therefore, mental practice of *Wu Qin Xi* is a suitable alternative therapeutic as to physical dynamic Qigong training.

In the present study, we tested whether the beneficial effects of physical practice of the dynamic Qigong technique on EEG brain activity can be reached by practicing *Wu Qin Xi* mentally. This research question becomes relevant for the everyday

practitioner in situations when the physical practice of Qigong is impracticable. Further, this research question is relevant for designs of Qigong courses for elderly persons or patients with bodily impairments. Especially at the beginners' stage, the movement sequences are practiced many times with several hours of practicing physically leading to increased physical tiredness. Here, the research question arises whether intervals of mental practice sessions would have the same effect on EEG brain activity as a merely physical Qigong practice.

Secondly, mental Qigong practice becomes relevant especially in persons who do not have the possibility to engage in Qigong training physically, either on the short-term, or on the long-term perspective. For instance, in stroke patients who have a low capability to move, mental training of the Qigong technique *Wu Qin Xi* would be an appropriate intervention to induce low frequencies in EEG brain activity and especially stimulate the Mu wave activity in the motor and premotor areas. Recent research has shown that mental practice in the form of motor imagery causes comparable patterns of brain activation as physical training of the movement. The rationale behind is that mental practice with motor content engages areas of the brain that govern movement execution (for an overview see Cicinelli et al., 2006; Sharma and Baron, 2013). Increases in alpha power are reported during mental practice of swimming movements (Beyer et al., 1990). In the same line, changes in EEG alpha oscillations in mental practice of volleyball serves were observed (Stecklow et al., 2010). Transient activations of the M1 area during mental practice are reported (Jeannerod, 1994, 2006; Pfurtscheller et al., 1997; Romero et al., 2000; Munzert et al., 2009). More precisely, inhibition of a movement leads to synchronization in alpha activity whereas preparation, execution and imagery lead to a de-synchronization in sensorimotor areas in the alpha and beta bands (Neuper et al., 2005; Neuper and Pfurtscheller, 2010). In a recent study, it was shown that these specific neuronal circuits are built with increasing experience with a motor task (Nakata et al., 2010). Reiterated engagement of motor areas as in mental *Wu Qin Xi* training is intended to influence brain plasticity phenomena, improving functional outcomes (Cramer et al., 2011; Dimyan and Cohen, 2011; Moissello et al., 2013). Recently, the

rehabilitative potential of motor imagery was shown contributing to significantly better motor functional outcomes in sub-acute stroke patients with severe motor impairments (Pichiorri et al., 2015). Further, in patients with chronically relapsing diseases of the musculoskeletal system mental practice of *Wu Qin Xi* would be a suitable therapeutic intervention. Finally, in achievement sports, when athletes experience phases of immobility due to sports concussions, mental practice of *Wu Qin Xi* Qigong would be an appropriate alternative to physical training.

Further research is needed to clarify whether regular mental practice enhances neuroplasticity as shown for physical training of the dynamic Qigong technique *Wu Qin Xi* (Henz et al., 2013) or in meditational Qigong. From studies on the role of expertise level in Qigong practice it was demonstrated that development of a frequency-specific brain excitability is a long-term process. For instance, Shim (2012) demonstrated theta activity centering around the frontal lobe parts (Fp1, Fp2, Fz, F4) in Qigong masters and decreased alpha activity compared to beginners. The authors argue that Qigong experts maintained more deeply internalized and relaxed theta activity in the frontal lobe which reflects an attentive mind. Therefore it is argued that Qigong masters show efficiency in keeping a relaxed and attentive mind around central midline. One further interesting question is whether the same effects would be expected for mental training of the Qigong technique *Wu Qin Xi* in clinical populations with reduced alpha oscillations at resting baseline (see Basar et al., 2012).

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Neural Basis of Working Memory Enhancement after Acute Aerobic Exercise: fMRI Study of Preadolescent Children

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Working memory lies at the core of cognitive function and plays a crucial role in children's learning, reasoning, problem solving, and intellectual activity. Behavioral findings have suggested that acute aerobic exercise improves children's working memory; however, there is still very little knowledge about whether a single session of aerobic exercise can alter working memory's brain activation patterns, as assessed by functional magnetic resonance imaging (fMRI). Therefore, we investigated the effect of acute moderate-intensity aerobic exercise on working memory and its brain activation patterns in preadolescent children, and further explored the neural basis of acute aerobic exercise on working memory in these children. We used a within-subjects design with a counterbalanced order. Nine healthy, right-handed children were scanned with a Siemens MAGNETOM Trio 3.0 Tesla magnetic resonance imaging scanner while they performed a working memory task (N-back task), following a baseline session and a 30-min, moderate-intensity exercise session. Compared with the baseline session, acute moderate-intensity aerobic exercise benefitted performance in the N-back task, increasing brain activities of bilateral parietal cortices, left hippocampus, and the bilateral cerebellum. These data extend the current knowledge by indicating that acute aerobic exercise enhances children's working memory, and the neural basis may be related to changes in the working memory's brain activation patterns elicited by acute aerobic exercise.

Keywords: acute aerobic exercise, working memory, N-back, brain activation patterns, fMRI, preadolescent children

INTRODUCTION

Working memory involves temporary storage and manipulation of information assumed necessary for a wide range of complex cognitive activities (Baddeley, 1992, 2003). It is an essential element for learning, memory, decision-making, cognitive control, other high-level cognitive activities, and brain development (Baddeley, 1992, 2003; Ericsson and Kintsch, 1995; Bechara et al., 1998; de Jong, 1998; Passolunghi and Siegel, 2001; Swanson and Sachse-Lee, 2001; Bull et al., 2008). Deficits in working memory will seriously harm the development of children's physical, mental, and social achievements; conversely, individuals, local communities, and society will benefit from well-developed working memory (Siegel and Ryan, 1989; Ericsson and Kintsch, 1995; Bechara et al., 1998; de Jong, 1998; Passolunghi and Siegel, 2001; Bull et al., 2008). Although working memory has a neural

anatomical basis, it is flexible and plastic, and thus, can be improved through training, especially in high correlation with children's cognitive development (Olesen et al., 2004; Westerberg and Klingberg, 2007; Takeuchi et al., 2010; Kamijo et al., 2011; Mrazek et al., 2013). Therefore, working memory has attracted increasing research attention in various fields and has become a frontier in interdisciplinary research. Identification of effective methods to develop children's working memory is a focus of the current research.

A burgeoning body of literature has emerged on exercise's positive effects on the brain and cognition. Aerobic exercise as an effective method for improving children's brain and cognitive function has been gradually recognized and practiced (Davis et al., 2007, 2011; Erickson and Kramer, 2009; Diamond and Lee, 2011; Erickson et al., 2011; Chang et al., 2012, 2013; Chapman et al., 2013; Hötting and Röder, 2013; Verburch et al., 2013; Chen et al., 2014). Recent findings have also suggested that acute aerobic exercise enhances working memory (Pontifex et al., 2009; McMorris et al., 2011; Li et al., 2014). Nevertheless, it is not clear whether the neural basis of improvement in children's working memory is elicited by acute aerobic exercise.

Functional magnetic resonance imaging (fMRI) non-invasively and safely measures and maps brain activity (Fox and Raichle, 2007). With fMRI, brain activation can be evaluated by measuring the blood oxygenation level-dependent (BOLD) contrast signal, which reflects a change in the ratio of oxygenated to deoxygenated hemoglobin that occurs with brain activation and increases in local blood volume. In a growing number of studies, fMRI is applied to directly understand how brain function changes with aerobic exercise or training (Kramer and Erickson, 2007; Chaddock et al., 2010; Davis et al., 2011; Erickson et al., 2011; Li et al., 2014). Through fMRI, working memory's neural basis has been found to have a specific pattern of brain activation, which, in working memory, mainly includes frontal and parietal cortices (Cohen et al., 1997; Klingberg et al., 2002; Baddeley, 2003; Klingberg, 2006; Darki and Klingberg, 2014; Ester et al., 2015; Harding et al., 2015). These brain regions' functional specialization and cooperation are the operating basis of working memory (Diwadkar et al., 2000; Baddeley, 2003). Moreover, studies from cognitive psychology and neuroscience have revealed that working memory training increases and decreases in task-related BOLD activity in different regions associated with increases in working memory capacity (Constantinidis and Klingberg, 2016). That is, both increases and decreases in the BOLD signal can be informative about the stimulus maintained in working memory, reflecting excitatory and suppressive responses to stimuli's orientation and motion. Additionally, a decrease in the BOLD signal in a certain area is often interpreted as an increase in the area's "efficiency" in performing its function. In short, improvement in working memory relates to its activation pattern changes (Klingberg et al., 2002; Olesen et al., 2004; Westerberg and Klingberg, 2007; Takeuchi et al., 2010; Kamijo et al., 2011; Mrazek et al., 2013). Therefore, the key to clarifying the neural basis of working memory enhancement caused by children's acute aerobic exercise is to reveal changes in working memory's brain activation patterns elicited by acute aerobic exercise.

In summary, the present study explores the effect of acute moderate-intensity aerobic exercise on working memory and its brain activation patterns in preadolescent children, and further explored the neural basis of acute aerobic exercise on working memory in these children. We hypothesize that our study would both replicate previous studies, demonstrating that acute aerobic exercise improves working memory, and extend our current understanding of this process to discover that acute aerobic exercise can better optimize working memory's brain activation pattern.

MATERIALS AND METHODS

Ethic Statement

The study protocol was approved by the Institutional Review Board of Beijing Normal University. All participants and their guardians provided written consent, and the protocol was approved by the institutional review board of Beijing Normal University.

Participants

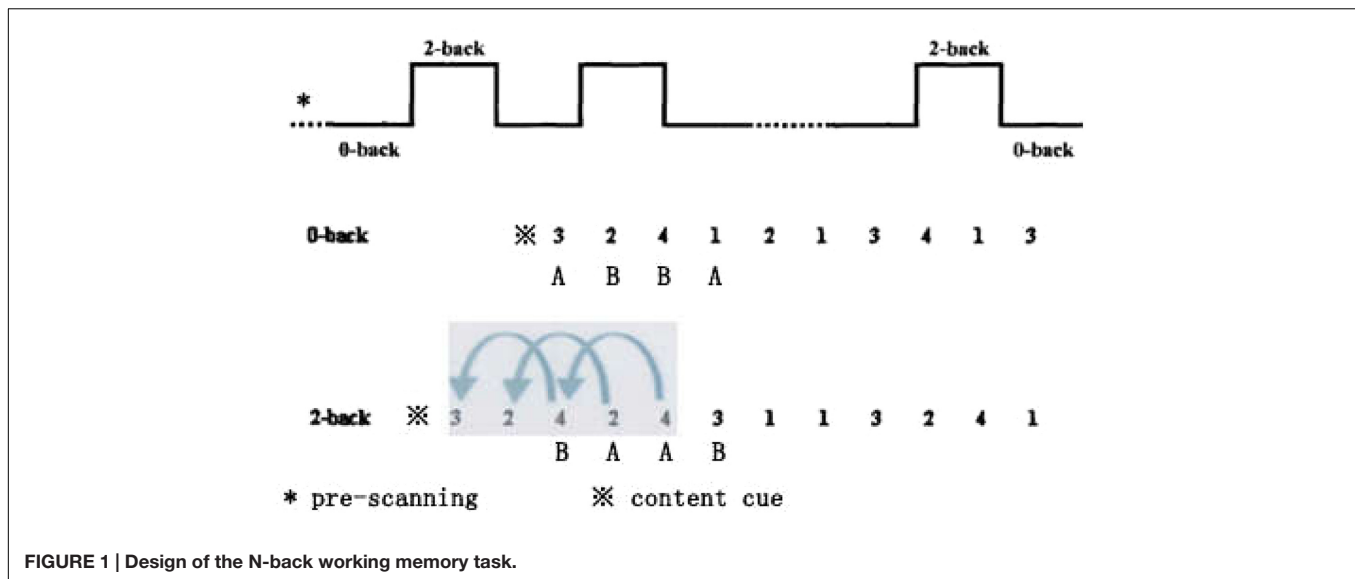
Nine healthy children in fifth grade (10 years old; five males, four females) were recruited through primary school advertising. They had normal or corrected-to-normal vision and were right-handed, as assessed by the Edinburgh Test (Oldfield, 1971). They also completed a set of questions about history of drug abuse or inherited disease and general intelligence (Wechsler Intelligence Scale for Children-IV-Chinese Version, WISC-IV-C) (Zhang, 2009). Exclusions included any medical condition that would limit physical activity or affect study results (including neurological or psychiatric disorders). The study was conducted in accordance with the Declaration of Helsinki. The study protocol was approved by the Institutional Review Board of the Brain Imaging Center of the State Key Laboratory of Cognitive Neuroscience and Learning, Beijing Normal University. Written informed consent was obtained from all participants and their legal guardians after experimental procedures were fully explained.

Acute Aerobic Exercise Protocol

Exercise was performed on a bicycle ergometer (MONARK 834, Sweden) with moderate intensity, which has been shown to benefit children's cognition (Hillman et al., 2009; Chen et al., 2014). This study used 60–69% of the predicted maximal heart rate, defined as 220 minus age in years, to determine the exercise intensity target heart rate (American College of Sports Medicine, 2006). Participants spent 2 min cycling to warm up, 30 min exercising at moderate intensity, and finally, 3 min cooling down at a self-determined pace. Heart rate was monitored in real time using a Polar heart rate monitor (RS800XSD, Finland) throughout the acute aerobic exercise protocol, which was led by the same instructor, one-on-one.

Working Memory Task

An N-back task (Smith and Jonides, 1997; Owen et al., 2005) was programmed using E-Prime to assess working memory.



The task used group block design (Figure 1), contained 0- and 2-back conditions, and was performed alternately. The 2-back condition was a working memory task, while the 0-back condition was a control task that did not need to manipulate working memory. Thus, the contrast between 0-back and 2-back conditions reflects, to a large degree, time-related aspects of processing stimuli in working memory. The N-back task had five 2-back conditions and six 0-back conditions. The task consisted of a series of changing number stimuli displayed at the center of a computer screen (i.e., 1, 2, 3, 4). The first block of one to four phases was adaptive, and the screen center was “ready,” prompting the participant to prepare to respond. Each stimulus was displayed on the screen for 2,000 ms and each phase’s duration was 2,000 ms. The presentation time for each number stimulus was 1,500 ms, and the reaction time was 500 ms. Working memory’s behavioral performance consisted of mean reaction time (RT) and mean accuracy (Acc); consequently, the shorter the RT, and/or the higher the Acc, the better the participant’s working memory.

Each 0-back condition included 11 phases in random order. The first phase was an experimental content cue: *single* or *double*; when the numeral 1 or 3 appeared, pressing button A; when the numeral 2 or 4 appeared, pressing button B. These four numbers were displayed randomly at equal probability.

Each 2-back condition included 13 phases in random order. The first phase was an experimental content cue: *Remember and go back 2*; the participant needed to remember the second and third stimuli in order of appearance. When the fourth stimulus appeared, they had to respond whether it was the same as the second; if it was, they pressed A, and otherwise, pressed B on the keyboard with both hands.

Experimental Procedure

This experiment had a completely within-subjects design. It was conducted in the Imaging Center for Brain Research at Beijing Normal University. During the first visit, participants and

their legal guardians completed all paperwork, including written informed assent/consent, as previously described. Following paperwork completion, an N-back task practice was administered to each participant, and the experimenter checked their performances to ensure that the participants understood the task. If a participant’s task performance was below 80% (Acc), the same practice was re-administered. In the formal experiment, all participants attended two sessions (i.e., baseline and exercise), with the order counterbalanced across participants at the same time on two separate days (a 7-day interval) in which they had not participated in physical education or other structured physical activity. So half participants received the baseline session on the first day and the exercise session on the second day. The other half received the exercise session on the first day and the baseline session on the second day. The baseline session consisted of 30 min of seated rest, during which all participants were fitted with a heart rate monitor and their resting heart rates were recorded. Following the seated rest period, participants completed an N-back task during MRI scanning. The exercise session consisted of a 30-min rest, with the resting heart rate recorded, and an acute aerobic exercise protocol during which HR was recorded in real time. Following the completion of the acute aerobic exercise protocol, once participants’ HRs returned to within 10% of their resting heart rate levels, the N-back task was performed during MRI scanning. Upon completion of both sessions, participants and their legal guardians received fair remuneration for their involvement in the experiment.

Functional MRI Data Acquisition and Image Processing

Participants underwent one scan for high-resolution structural images of the whole brain on a 3T Siemens Magnetom Trio system (Siemens) with total imaging matrix in the Imaging Center for Brain Research, Beijing Normal University. Functional images were obtained using an echo-planar imaging (EPI) sequence, with the following scan parameters: TR = 2,000 ms,

TE = 30 ms, gap = 1 mm, flip angle (FA) = 90°, slice thickness = 3.0 mm, field of view (FOV) = 200 × 200 mm, and inplane resolution = 64 × 64. Resulting data included 148 brain volumes with 33 axial slices. During the fMRI scans, all participants were instructed to stay relaxed and move as little as possible. High-resolution structural images were acquired using a magnetization-prepared rapid gradient echo, three-dimensional T1-weighted sequence (TR = 2000 ms, TE = 3.39 ms, T1 = 1100 ms, FA = 7°, thickness = 1.33 mm, FOV = 200 × 200 mm, acquisition matrix = 256 × 256).

Functional image preprocessing and statistical analyses were conducted with DPABI¹, based on SPM8². The first three volumes of functional images were discarded for signal equilibrium and participants' adaptation to scanning noise. Subsequent functional images underwent the following preprocessing steps: slice-timing correction, realignment, co-registration, and New Segment + Diffeomorphic Anatomical Registration Through Exponentiated Liealgebra (DARTEL) with high-resolution structural scans (Ashburner and Friston, 2005). The DARTEL tool (Ashburner, 2007) was used to compute transformations from individual native space to Montreal Neurological Institute (MNI) coordinate space. Then, the segmented BOLD volumes were normalized into standardized MNI space using the DARTEL template and resampled to 3 mm × 3 mm × 3 mm isotropic voxels. Finally, normalized images were smoothed with an 8 mm × 8 mm × 8 mm full width at half maximum Gaussian kernel.

Statistical Analysis

First, descriptive data were evaluated to determine the appropriateness of exercise intensity manipulation. Second, behavioral improvements in RT and Acc for working memory across the two sessions were analyzed by paired *t*-test with SPSS. Third, functional changes were analyzed with two procedures, individual and group analysis. Individual analysis: The 0-back condition was considered a control task in the present study. The 2-back condition required maintenance and permanent update of relevant pieces of information in working memory. Then, statistical parametric maps (SPMs) were computed for individual participants by using the general linear model (GLM) with separate hemodynamic basis response function modeling MR signal responses for each task period. Contrast images (2-back vs. 0-back) on estimates of interest were obtained for each participant. Group analysis: There were two steps for group analysis based on the contrast images (2-back vs. 0-back). The first was to identify whether the N-back task is successfully inducing the common brain activation patterns of working memory for each session (baseline and exercise), we conducted a one-sample *t*-test on the contrast images (2-back vs. 0-back). The second was to examine neural activation differences between two sessions (baseline and exercise); we conducted a paired *t*-test on the contrast images (2-back vs. 0-back) to detect acute aerobic exercise gains.

¹<http://rfmri.org/dpabi>

²<http://www.fil.ion.ucl.ac.uk/spm/software/spm8/>

RESULTS

Exercise Intensity Manipulation

The heart rates for the baseline and exercise sessions were 41.80 and 64.52%, respectively, of the predicted maximal heart rate [$t(8) = 23.70, p < 0.001$]. The two sessions' differing heart rates and percentages of the predicted maximal heart rate suggest that the selected moderate-intensity exercise was appropriate.

Behavioral Performances

Table 1 presents detailed behavioral measures based on the two sessions and the N-back conditions. To explore behavioral performance differences between the baseline and exercise sessions, a paired *t*-test was used on RT and accuracy. There were no significant differences between the baseline and exercise sessions on 0-back RT [$t(8) = 1.37, p > 0.05$ and Acc $t(8) = -1.03, p > 0.05$; 2-back Acc $t(8) = -0.82, p > 0.05$]. The 2-back RT of the exercise session showed significant improvement after acute exercise $t(9) = 2.79, p < 0.05, r^2_{pb} = 0.49$; that is, shorter RT demonstrated better working memory.

Brain Activation Differences

The first analytical goal was to examine working memory activation in two scanning sessions (baseline and exercise) with contrast images (2-back vs. 0-back), calculated as an assessment of the dynamic range of neural differences activation between a control task and a working memory task. The statistical threshold was set at $p < 0.001$ with a cluster size threshold of 75 voxels, which is equivalent to cluster-level $p < 0.05$, AlphaSim corrected. Specifically, in the baseline session, regions of significant BOLD activation were the left Superior Frontal Gyrus (SFG), bilateral Middle Frontal Gyrus (MFG), right Inferior Frontal Gyrus (IFG), bilateral Parahippocampal gyrus (PHP), right Middle Occipital Gyrus (MOG), left Superior Temporal Gyrus (STG), and bilateral Cerebellum Posterior Lobe; in the exercise session, right Medial Frontal Gyrus (MEDFG), MFG, left Superior Parietal Lobule (SPL), right Inferior Parietal Lobule (IPL), right Superior Occipital Gyrus (SOG), left Anterior Cingulate Cortex (ACC), right Posterior Cingulate Cortex (PCC), and bilateral Cerebellum Posterior Lobe were activated (**Table 2; Figure 2**). This analysis of activated brain regions revealed the common patterns of working memory.

In further comparisons of brain regions' activated changes between the baseline and exercise sessions, a paired *t*-test revealed significant difference. The statistical threshold was set at $p < 0.025$ with a cluster size threshold of 100 voxels, which is equivalent to cluster-level $p < 0.05$,

TABLE 1 | Children's behavioral performances on N-back task in baseline and exercise sessions.

Condition	Baseline session		Exercise session	
	ACC (%)	RT(ms)	ACC (%)	RT(ms)
0-back	0.95 ± 0.03	682.80 ± 52.68	0.96 ± 0.03	643.92 ± 73.35
2-back	0.81 ± 0.03	963.13 ± 125.24	0.82 ± 0.03	877.57 ± 53.74

TABLE 2 | Children's brain activation patterns during working memory in baseline and exercise scanning sessions.

Primary regions	Brodmann area	Size (voxel)	Max T-statistic	MNI coordinates		
				x	y	z
Baseline session						
B_Cerebellum Posterior Lobe	–	103	21.84	–36	–60	–45
L_Parahippocampa Gyrus	30	235	–13.23	–24	–21	–18
R_Parahippocampa Gyrus	34	4249	–21.29	24	–3	–15
L_Medial Frontal Gyrus	10	506	–8.62	–3	54	–6
R_Middle Occipital Gyrus	18	1352	–19.21	21	–99	3
L_Superior Temporal Gyrus	48	289	–13.35	–39	–15	15
R_Inferior Frontal Gyrus	47/48	158	17.06	30	18	–9
B_Middle Frontal Gyrus	32/24	844	17.54	0	18	42
L_Superior Frontal Gyrus	9	108	–9.41	–9	48	42
Exercise session						
L_Cerebellum Posterior Lobe	–	111	18.25	–33	–66	–48
B_Cerebellum Posterior Lobe	–	80	9.59	–6	–81	–30
L_Middle Frontal Gyrus	47	84	–9.48	–36	39	–18
L_Anterior Cingulate Cortex	25	131	–9.72	–6	24	–6
R_Medial Frontal Gyrus	10	371	–15.92	6	63	–3
R_Middle Frontal Gyrus	46	246	11.51	45	48	15
R_Superior Occipital Gyrus	19	160	–9.06	33	–84	24
R_Posterior Cingulate Cortex	30	442	–9.49	6	–51	21
L_Middle Frontal Gyrus	32	1015	34.98	–6	21	39
L_Superior Parietal Lobule	7	320	11.94	–6	–72	57
R_Inferior Parietal Lobule	40	215	14.84	48	–42	39

The statistical threshold was set at $p < 0.001$, with a cluster size threshold of 75 voxels, which is equivalent to cluster-level $p < 0.05$, AphaSim corrected. All coordinates are reported in the MNI format.

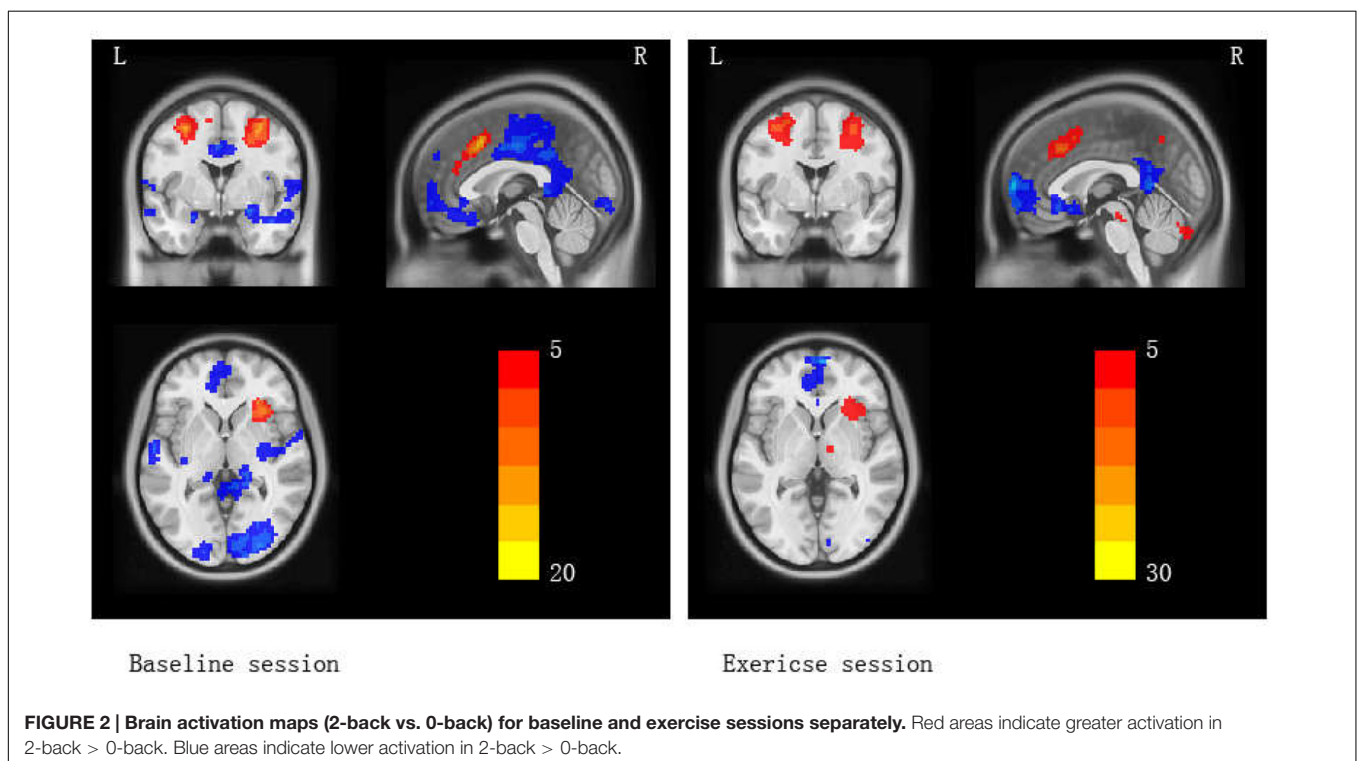


TABLE 3 | Significant brain activation of paired t-test between children's baseline and exercise MRI scanning.

Primary regions	Brodmann area	Size (voxel)	Max T- statistic	MNI coordinates		
				x	y	z
L_Superior/Inferior Parietal Lobule	7/40	497	8.64	-15	-57	45
R_Superior Parietal Lobule	5/7	191	6.57	12	-60	54
L_Hippocampus	20	208	8.23	-30	-30	-12
L_Cerebellum	-	347	7.18	-24	-60	-33
R_Cerebellum	-	108	6.47	36	-63	-27

The statistical threshold was set at $p < 0.025$, with a cluster size threshold of 100 voxels, which is equivalent to cluster-level $p < 0.05$, AphaSim corrected. All coordinates are reported in the MNI format.

AphaSim corrected. Specifically, the exercise session resulted in greater activation of SPL, IPL, left Hippocampus (HIP), and bilateral Cerebellum (**Table 3**; **Figure 3**). This analysis indicated that acute aerobic exercise significantly increased some regions of brain activities for working memory.

DISCUSSION

This study investigated behavioral and neural effects of children's acute aerobic exercise on a working memory task. In both baseline and exercise sessions, participants were tested with a working memory task while being scanned. The exercise session, an acute aerobic exercise protocol, was compared to a baseline session without exercise intervention. Consequently, reliable acute aerobic exercise gains emerged, allowing us to test for acute aerobic exercise effects on the two sessions.

Behavioral Performances

A rapidly growing body of literature indicates that, from both behavioral and neuroelectric perspectives, acute aerobic exercise improves working memory (Lardon and Polich, 1996; Hillman et al., 2008; Erickson and Kramer, 2009; Chang et al., 2012, 2013; Chen et al., 2014; Drollette et al., 2014). As observed here, children's working memory performance in the exercise session was better than in the baseline session—in agreement with previous studies. Accordingly, the present behavior results have been again verified: acute aerobic exercise beneficially impacts children's working memory.

Brain Activation Patterns of Working Memory

To our knowledge, many previous studies have explored the macroscale of working memory's neural system. First, in an N-back task, the present study successfully induced working memory's common brain activation patterns. However, how

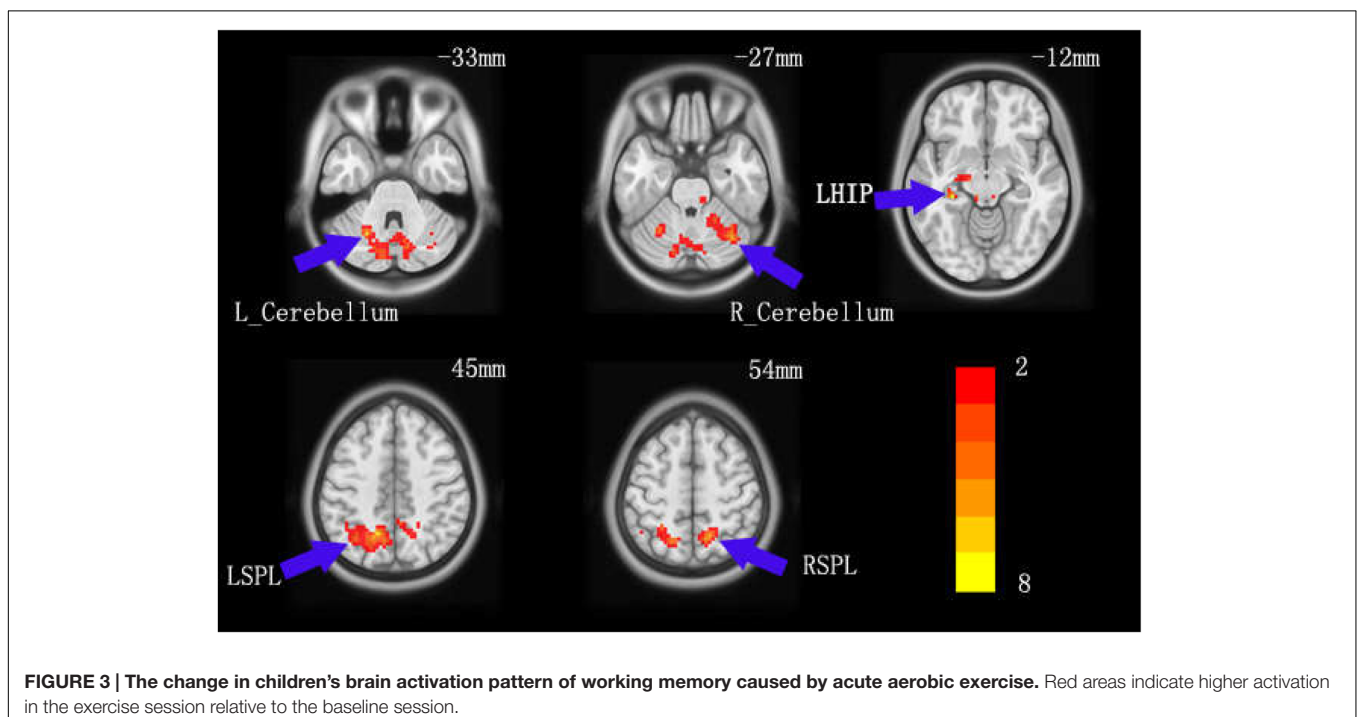


FIGURE 3 | The change in children's brain activation pattern of working memory caused by acute aerobic exercise. Red areas indicate higher activation in the exercise session relative to the baseline session.

do working memory's brain activation patterns run? Cognitive neuropsychology observations concluded that working memory's neural basis is quite widespread, including the cerebral cortex (e.g., frontal, temporal, parietal, and occipital lobes) and subcortical areas (e.g., thalamus, amygdala, hippocampus, and cerebellum) (LaBar et al., 1999; Smith and Jonides, 1999; Diwadkar et al., 2000; Baddeley, 2003; Osaka et al., 2004; Owen et al., 2005). Brain areas in our results matched these and indicated that the N-back task successfully induced working memory activation patterns.

Second, we compared the exercise session's brain activation maps to those of the baseline session. Regarding acute aerobic exercise-related neural effects, the main findings were that acute aerobic exercise raised working memory's brain activation patterns in bilateral SPL, left IPL, left HIP, and bilateral cerebellum. These results are fully consistent with our prediction that acute exercise significantly influenced working memory's brain activities. These results may be of great importance for understanding the neural basis of acute aerobic exercise's effect on working memory. Indeed, prior studies have demonstrated that improved cognitive function results from brain plasticity changes caused by aerobic exercise (Kramer and Erickson, 2007; Erickson and Kramer, 2009; Voss et al., 2013). One finding reported that high-fitness children showed greater bilateral hippocampal volumes, positively associated with performance on memory task (Chaddock et al., 2010); this study first suggested that aerobic fitness can impact the structure and function of the developing human brain. Similarly, a recent study suggested that significant changes after acute aerobic exercise with brain activation reflected improved working memory in young female college students (Li et al., 2014). Thus, acute aerobic exercise could benefit children's working memory at a macro-neural level.

What is the meaning of children's greater brain activation caused by acute aerobic exercise? Several previous studies have addressed this question, assisting us to understand these increases in SPL, IPL, and bilateral cerebellum and hippocampus. Development of functionality in these areas plays an important role in cognitive development during childhood, and parietal cortices are known to be involved in working memory (Jonides et al., 1998; Culham and Kanwisher, 2001; Koenigs et al., 2009). Working memory capacity significantly correlated with brain activities in the parietal gyrus (Vogel and Machizawa, 2004; Todd and Marois, 2005). Similarly, another study reported that higher parietal activity was associated with higher working memory capacity in children (Klingberg et al., 2002). A meta-analysis concluded that the functional topography of the cerebellum is particularly involved in sensorimotor, language, spatial, and working memory (Stoodley and Schmahmann, 2009). Prior evidence indicated that cognitive training could increase neural activity in cerebellar circuits in children with ADHD (Hoekzema et al., 2010). Recent studies of the cerebellum have revealed connections between the cerebellum and parietal gyrus that

might support cerebellar contributions to working memory (Allen et al., 2005). The hippocampus plays an important role in the formation of new memories about experienced events (Vinogradova, 2001). Moreover, previous observations found that the parietal lobule is the target of disynaptic output from two subcortical sites, in which the parietal lobule receives a strong disynaptic input from the hippocampus and the cerebellum contains a distinct output channel that targets a portion of the parietal lobule (Clower et al., 2001). At this point, we speculate on the neural basis of improved working memory induced by acute aerobic exercise: Greater activation of the cerebellum might contribute to the parietal cortex's better functioning and strengthened correlations between the hippocampus and parietal cortex, consequently enhancing children's working memory.

This study is not without its limitations. First, a completely within-subjects design was used in current study in which all subjects are exposed to every experimental session. The drawback of this design is the absence of control group. However, this design ensures that every subject acts at their own control, so there are few problems with matching age, gender, and lifestyle, reducing the chances of confounding factors. Second, generalization of our results is limited by small sample size. Future studies with a larger sample may be more amenable to investigate the effect of acute moderate-intensity aerobic exercise on working memory and its brain activation patterns in preadolescent children.

CONCLUSION

Here, we suggest that acute aerobic exercise results in children's greater cognitive gains. These data extended the current knowledge base by indicating that acute aerobic exercise enhances children's working memory, in which the neural basis may be related to changes in working memory's brain activation patterns elicited by the exercise.

AUTHOR CONTRIBUTIONS

A-GC and H-CY designed the study and oversaw the data collection. L-NZ and A-GC analyzed the data and wrote the initial manuscript. JY and H-CY assisted with data analysis and organized the manuscript. All authors played a part in the manuscript's preparation at each stage of its development. All authors have read and approved the manuscript's final version.

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Mind-Body Practice Changes Fractional Amplitude of Low Frequency Fluctuations in Intrinsic Control Networks

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Cognitive control impairment is a typical symptom largely reported in populations with neurological disorders. Previous studies have provided evidence about the changes in cognitive control induced by mind-body training. However, the neural correlates underlying the effect of extensive mind-body practice on cognitive control remain largely unknown. Using resting-state functional magnetic resonance imaging, we characterized dynamic fluctuations in large-scale intrinsic connectivity networks associated with mind-body practice, and examined their differences between healthy controls and Tai Chi Chuan (TCC) practitioners. Compared with a control group, the TCC group revealed significantly decreased fractional Amplitude of Low Frequency Fluctuations (fALFF) in the bilateral frontoparietal network, default mode network and dorsal prefrontal-angular gyri network. Furthermore, we detected a significant association between mind-body practice experience and fALFF in the default mode network, as well as an association between cognitive control performance and fALFF of the frontoparietal network. This provides the first evidence of large-scale functional connectivity in brain networks associated with mind-body practice, shedding light on the neural network changes that accompany intensive mind-body training. It also highlights the functionally plastic role of the frontoparietal network in the context of the “immune system” of mental health recently developed in relation to flexible hub theory.

Keywords: mind-body practice, frontoparietal network, fMRI, plasticity, amplitude, Tai Chi Chuan

INTRODUCTION

Various neurological diseases associated with significant deficits in cognitive control have been widely reported in the literature (Verma and Howard, 2012; Robbins and Cools, 2014). Notably, cognitive control declines with the development of the disease and is not reversible. Therefore, the prevention of neurological diseases associated with declining cognitive control appears to be important and should become one of the best long-term strategies to solve this public health problem.

Currently, physiotherapeutic or exercise-related interventions are regarded as promising non-pharmaceutical tools to help improve cognition, especially in older at-risk individuals (Aas-Hansen et al., 2000). A recent meta-analysis concluded that exercise, particularly when meeting physical activity guidelines, can improve clinical symptoms in adults with neurologic disorders by enhancing cognitive control (Adamson et al., 2015). Based on the findings of several studies investigating the involvement of fronto-limbic regions after mindfulness meditation practice, it has been proposed that mindfulness works by strengthening prefrontal cognitive control mechanisms and thus down-regulates activity in regions relevant to affective processing (Tang et al., 2015). Therefore, integrative mind-body practice, consisting of both physical exercise and a meditation component, could have broader implications for the cognitive treatment of psychiatric and neurologic disorders. It is worth mentioning that Marciniak et al. (2014) proposed a theoretical framework on the effects of mindfulness-based practice in the context of aging and neurodegenerative diseases, which addressed the pivotal role of cognitive control among various effects, including physiological aspects (cholesterol) and the neuroprotection profiles of emotions.

Recent advances of resting-state MRI in functional and structural connectomics have revealed a complex interplay of different brain areas (i.e., instead of a set of brain areas with highly specialized functions) (Power et al., 2011; Bullmore and Sporns, 2012). These advances also highlight the role of macro-scale networks in the study of resting human brain function and its association with the mind, behavior, and disease. Several new findings related to training for physical exercise and mind have emphasized the importance of crucial functional networks such as default network, frontoparietal network, and salience network (Brewer et al., 2011; Krafft et al., 2014). Notably, recent findings suggest the existence of a frontoparietal control system, consisting of flexible hubs that regulate distributed systems throughout the brain, which is closely associated with mental diseases (Cole et al., 2014). It is also suggested that extensive training could reduce activity of the frontoparietal control system (Chiesa et al., 2013). Alternatively, with the help of similar mental or physical training, the frontoparietal control system could be responsible for training other systems to automatically facilitate an optimal state; a state incompatible with a variety of harmful dysfunctions. Accordingly, it is of great importance to investigate whether mind-body practice could optimize the activity pattern of the frontoparietal control system.

Currently, low frequency oscillations (typically defined as frequencies < 0.1 Hz) in resting-state have gained increased attention based on observations using fMRI approaches (Fox and Raichle, 2007). The fractional Amplitude of Low Frequency Fluctuations (fALFF) is defined as the total power within the frequency range between 0.01 Hz and 0.1 Hz divided by the total power in the entire detectable frequency range, which is determined by sampling rate and duration (Zou et al., 2008). As a normalized index of ALFF, fALFF appears to be a biologically significant parameter for assessing regional brain function and can provide a more specific measure of low frequency oscillatory

phenomena (Zuo et al., 2010), which has recently been widely employed in mental diseases studies.

Tai Chi Chuan (TCC) originated in China as an integrative form of aerobic exercise and meditation and is regarded as a typical mind-body practice (Mansky et al., 2006; Sharma and Haider, 2015). Used as a clinical treatment, TCC has demonstrated potential benefits for people with chronic neurological diseases such as PD (Allen et al., 2011), AD (Klein, 2008), multiple sclerosis (MS), and mood disorders (Payne and Crane-Godreau, 2013; Wang et al., 2014). On a behavioral level, several cross-sectional and longitudinal studies have confirmed the positive effect of mind-body practice, mainly on cognitive control. A preliminary study with a pre-to-post test design observed TCC practitioners had significantly improved performance on cognitive executive control after a 10-week TCC program (Matthews and Williams, 2008). In more recent work, we have demonstrated that a TCC group showed shorter trend than a control group in reaction time (RT) in flanker test and this was significantly associated with TCC experience among aging TCC practitioners (Wei et al., 2013). This evidence provides a theoretical rationale for exploring the relationship between optimized macro-level brain networks and cognitive control following extensive TCC practice.

Hence, examining the low frequency activity pattern in resting-state is a potential way to explore the optimized large-scale brain networks induced by TCC, especially the frontoparietal control system. This research strategy will allow us to deepen our understanding of TCC's role in cognitive control and how that can be applied to clinical prevention and treatment of populations with neurological disorders. We hypothesize that frontoparietal control systems might be an important locus associated with TCC practice. Specifically, we predict that (1) we will find significant differences in fALFF in brain networks including frontoparietal network in experienced TCC practitioners compared with their matched controls; (2) these changes of fALFF in the frontoparietal network are behaviorally relevant to cognitive control in experienced TCC practitioners; (3) there is significant association between TCC experience and fALFF changes in practitioners' brain networks.

MATERIALS AND METHODS

Participants

We recruited TCC participants from a local community in Beijing. This group included 22 experienced TCC practitioners with a mean age of 52.4 ± 6.8 years old and a mean education level of 12.2 ± 2.9 years. On average, TCC participants had 14.6 ± 8.6 years of TCC experience and practiced 11.9 ± 5.1 h per week. One participant deviated from the group in terms of the intensity of TCC practice (30 h each week), and thus constituted an outlier. After removing this participant, the average years and intensity of TCC experience remained unchanged, but the estimation of practice hours (years \times practice hours per day \times days) changed from 9156 to 8775 h. This participant was kept in the following statistical analyses, including brain networks analyses and the correlation computation between

cognitive performance and brain networks, to avoid losing any statistical power by decreasing the sample size. According to a demographic survey, participants practiced different styles of TCC including Yang, Sun, Wu, and Chen with varied overt movements. However, these styles still share essential components of TCC. The control group was matched with the TCC cohort by age, gender, and education (age: 54.8 ± 6.8 years old, years of education: 11.8 ± 2.9 years). None of controls had experience in any type of regular TCC, yoga, meditation or exercise practices.

The Institutional Review Board of the Institute of Psychology, Chinese Academy of Sciences approved this study. This study was performed in accordance with the Declaration of Helsinki and its later amendments. The procedure of the study was fully explained to the participants and informed written consent was obtained from each of them before the study. None of the participants had any history of neurological or psychiatric illnesses, injury, seizures, metal implants, head trauma with loss of consciousness and were not on any chronic medications that could affect the experiment.

Cognitive Control Task

Ten participants separately from TCC group and control group completed the computerized Attention Network Test (ANT) (Fan et al., 2002) before fMRI scanning. In this study, we only report conflict effect (reaction time in inconsistent condition minus reaction time in consistent condition) during ANT that is target-related and reflects the performance of executive function. The experimental procedures have been previously described by Fan et al. (2002) and are only briefly introduced below. Each participant viewed a computer screen from a distance of around 50 cm after correcting for visual acuity. Eprime 2.0 professional (Psychology Software Tools, Pittsburgh, PA, United States) was used to program and present the task, which automatically collected all responses including reaction time and accuracy as soon as the participant completed the whole test. During the test, participants were instructed to respond as fast and accurately as possible via two input keys on a keyboard. The target stimulus was an arrow, indicating either the left or the right direction, presented in the center of a horizontal row of five arrows. The four surrounding flanker stimuli were all arrows pointing in the same direction as the central arrow (congruent condition) or the opposite direction of the target stimulus (incongruent condition) or were just neutral stripes (neutral condition). The target stimulus and the flanker stimuli were presented at a visual angle of 1.1 above or below a fixation cross presented in the middle of the screen. Preceding the presentation of the target, one of four cue conditions was provided: no cue; center cue; double cue; or spatial cue. These four conditions interact with the flanker task to influence RT. In brief, each trial consisted of the following structure: (1) a fixation cross was presented in the middle of the screen during a variable interval, ranging from 400 to 1600 ms; (2) a 100 ms cue was presented; (3) a 400 ms central fixation was presented; (4) the target stimulus was presented for 1700 ms, or shorter if a response was given within 1700 ms; (5) a fixation cross was presented during a variable

delay, which of this delay was determined by subtracting the RT plus 400 ms from the total trial duration being kept constant at 3500 ms.

The experiment lasted approximately 25 min and involved 288 trials, which were divided into three blocks of 96 trials each with a short break between blocks. Before the main task, all participants completed a training block of 24 randomly selected trials, which provided feedback for accuracy and RT at the end of each trial. This ensured that participants completely understood the task instructions.

Scanning Protocols

Brain imaging was performed on a 3T Trio system (Siemens, Erlangen, Germany) with a 12-channel head matrix coil. Two hundreds and forty-three volumes were obtained by using an echo planar imaging (EPI) sequence with the following scan parameters: repetition time (TR) = 2000 ms, echo time (TE) = 30 ms, flip angle (FA) = 90° , slice thickness = 3.0 mm, field of view (FOV) = $200 \text{ mm}^2 \times 200 \text{ mm}^2$, voxel-size = $3.4 \text{ mm} \times 3.4 \text{ mm} \times 4.0 \text{ mm}$, resulting in 243 brain volumes of 30 axial slices. During the resting scans, all participants were instructed to keep their eyes closed, relax, and move as little as possible. We also acquired a three dimensional magnetization prepared rapid gradient echo (3D MPRAGE) sequence for anatomical information with the resolution of $1.3 \text{ mm} \times 1.0 \text{ mm} \times 1.3 \text{ mm}$ (TR = 2530 ms, TE = 3.39 ms, FA = 7° , slice thickness = 1.33 mm) for better registration and overlay of brain activity.

Image Preprocessing and Quality Control Preprocessing Steps and QC Procedures

Preprocessing of the structural and functional images was implemented with the Connectome Computation System¹ (CCS) (Zuo et al., 2013; Xu et al., 2015), which is a computational platform for brain connectome analysis with integrating FreeSurfer (Dale et al., 1999; Fischl et al., 1999), FSL and AFNI to provide a pipeline system for multimodal image analysis. The data preprocessing was composed of steps for both anatomical and functional processing.

The structural image processing included the following steps of brain cortical surface reconstruction (Dale et al., 1999; Fischl et al., 1999; Segonne et al., 2004, 2007). Firstly, MR image noise was removed by using a spatially adaptive non-local means filter and MR intensity heterogeneity correction (Xing et al., 2011; Zuo and Xing, 2011). Brain was extracted with a hybrid watershed/surface deformation procedure and was segmented into different tissues such as the cerebrospinal fluid (CSF), white matter (WM), and deep gray matter (GM) volumetric structures. A triangular mesh tessellation was estimated over the GM-WM boundary and the mesh was deformed to produce a smooth representation of the GM-WM interface (white surface) and the GM-CSF interface (pial surface) spatial normalization from individual native space to *fsaverage* stereotaxic space. Correcting topological defect on the surface and inflated individual surface mesh into a sphere. Finally, making estimation

¹<https://github.com/zuoxinian/CCS>

of the deformation between the resulting spherical mesh and a common spherical coordinate system.

The functional image preprocessing discarded the first five EPI volumes from each scan to allow for signal equilibration, removed and interpolated temporal spikes, corrected acquisition timing among image slices and head motion among image volumes, normalized the 4D global mean intensity to 10,000 to allow inter-subject comparisons and regressed out the WM/CSF mean time series and the Friston-24 motion time series to reduce the effects of these confounding factors (Yan et al., 2013; Zuo et al., 2013). Finally, the residual time series with a band-pass (0.01–0.1 Hz) were filtered to extract the low frequency fluctuations. Both linear trends and quadratic trends were removed and individual motion corrected functional images were aligned to the individual anatomical image using the GM-WM boundary-based registration (BBR) algorithm (Greve and Fischl, 2009).

By combining BBR deformation and spherical surface normalization, the individually preprocessed 4D rfMRI time series were projected onto the *fsaverage* standard cortical surface with 163,842 vertices per hemisphere, with an average distance of 1 mm for neighboring pairs of vertices. Then, the data were down-sampled onto the *fsaverage5* standard cortical surface (3.8-mm neighboring-vertex distance), which contained 10,242 vertices per hemisphere (Yeo et al., 2011).

Quality Control Procedure

After preprocessing individual images, quality control procedure (QCP) was conducted by using CCS. Specifically, this procedure produces basic information concerning preprocessed images, including screenshots for visual inspection of: (1) skull stripping, (2) segmentation of brain tissue, (3) reconstruction of pial and white surface, (4) registration of BBR-based functional image, and (5) head motion processing during rfMRI. Several quantities are also produced, including the following: (1) the maximum distance of translational head movement (maxTran), (2) the maximum degree of rotational head movement (maxRot), (3) the mean frame-wise displacement (mean FD) (Power et al., 2012; Patriat et al., 2013), and (4) the minimal cost of the BBR co-registration (mcBBR). Any participants with bad brain extraction, tissue segmentation, and bad surface construction will be excluded from the subsequent analysis. Moreover, all datasets in the subsequent analysis must meet some criteria, which is described in detail of the website².

Computation of Network-Level fALFF

A set of spatial templates of 12 common intrinsic connectivity networks (ICNs) was generated from an independent sample, the NKI-rockland sample ($N = 126$), using an exploratory group-level intrinsic network discovery tool, gRAICAR (Yang et al., 2014; Figure 1). Data from the NKI-rockland sample were preprocessed using the same CCS pipeline. The preprocessed functional images were processed using gRAICAR (Yang et al., 2008, 2012) to characterize the consistency of the ICNs across all of the subjects. Spatial independent components (ICs) were derived from each subject using the MELODIC module of FSL,

where the number of independent components was automatically determined. All of the ICs from all the subjects were pooled in gRAICAR, and normalized mutual information between every pair of ICs was computed to yield a full similarity matrix. The full similarity matrix was then searched to match ICs across different subjects, forming group-level aligned components (ACs). Each AC was formed by a set of matched ICs containing one IC from each subject. For each of the ACs, a similarity matrix was computed to reflect the similarity between its comprising ICs, each representing a subject. In the inter-subject similarity matrix, the centrality of a subject's IC was computed by summing up the similarity metrics between that subject's IC and all other ICs in that AC. The significance of the centrality of each subject was examined using a permutation test. The centrality measures were then used as weights to average the spatial maps of the constituent ICs into a group-level spatial map for that AC. In summary, the spatial maps of the ACs represent ICNs or artifacts in the resting-state data, and the similarity matrices reveal inter-subject consistency of the ACs. According to the permutation test of inter-subject consistency, 12 ICN maps were (significantly) consistent across >60% of the subjects, and therefore these ICNs were used as ICN templates for further study.

Representative time series of these ICNs were obtained in both the original data and the band-pass (0.01–0.1Hz) filtered data by using spatial regression. The power of the representative time series from band-pass filtered data was divided by that of the original data, yielding fALFF of each ICN, a measure of the network dynamics or variability.

The fALFF metrics, demographic, and behavioral statistics were compared between TCC experts and control subjects using two sample *t*-tests in SPSS 20. Using SPSS, we conducted partial correlation analyses controlling for age, sex, and education between demographic data, behavioral data, and brain networks with significant differences. Only 21 TCC participants were involved in the correlation between TCC practice and brain networks due to the exclusion of an outlier based on practice hours each week.

RESULTS

Participant Demographics

Participant demographic data are provided in Table 1. The results showed that there was no significant difference in all relevant variables (i.e., gender, age, and education) between the TCC group and the control group. We also computed intracranial volume, global fALFF, and the root mean square of frame-wise displacement parameter (indicating head motion), and found no significant difference between the two groups.

Cognitive Control Performance

Response speed and accuracy are two important factors in assessing ANT performance. We computed the RT and accuracy in the cognitive control to examine the differences of executive function for the TCC group and the control group (see Figure 2). The two-sample *t*-tests revealed that the TCC group trended toward shorter RTs of cognitive control in ANT

²<http://fcd.psych.ac.cn/ccs/QC.html>

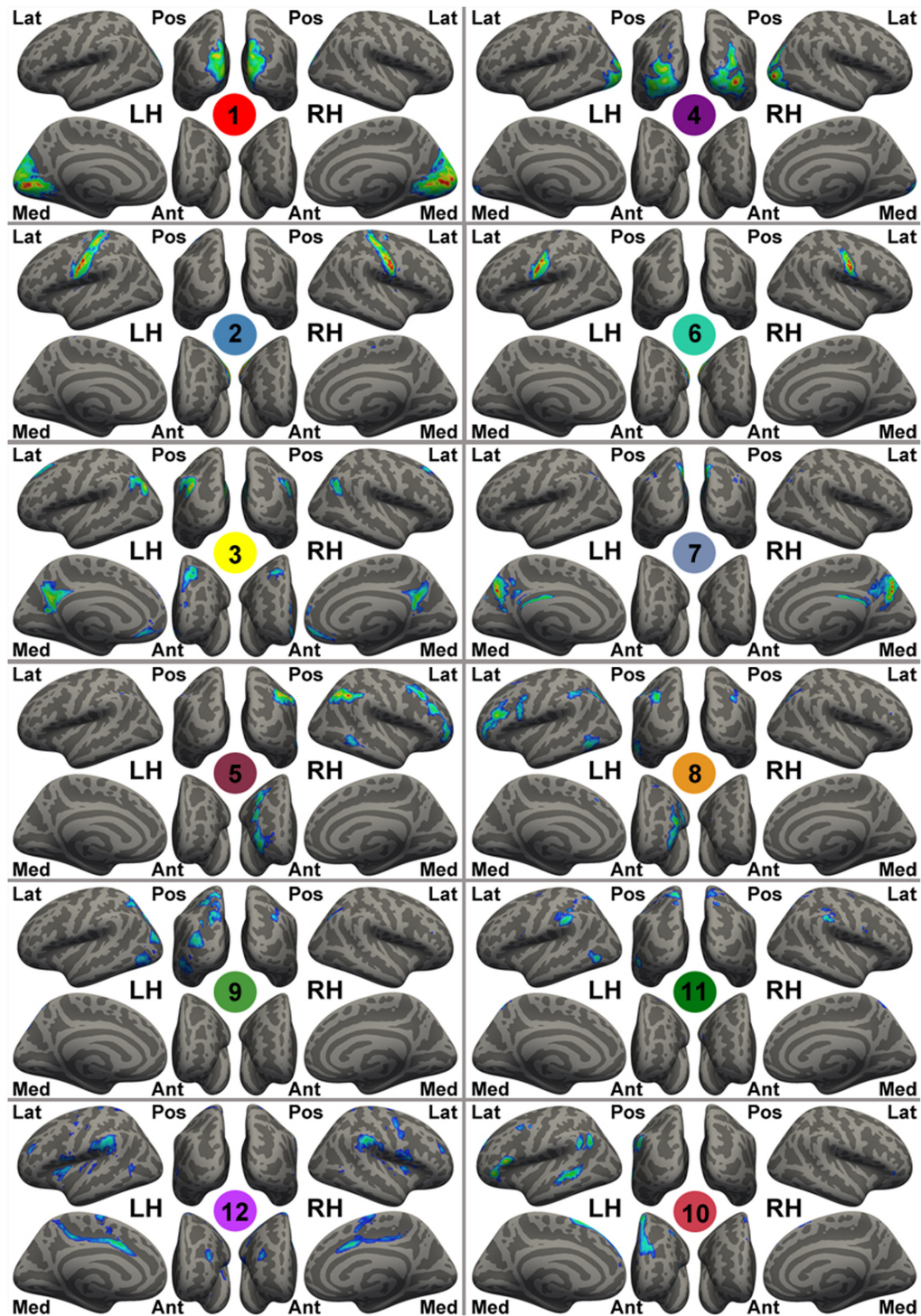


FIGURE 1 | Template intrinsic connectivity network (ICN) maps generated by gRAICAR using the NKI-RS sample. For the purpose of visualization, these template maps are rendered onto fsaverage standard surfaces from lateral (Lat), medial (Med), posterior (Pos), anterior (Ant) views in Freesurfer for both left (LH) and right (RH) hemispheres and thresholded at $|Z| > 1$. The Z-value here is the original intensity of the maps from gRAICAR (i.e., weighted z-scores given by MELODIC), instead of the value after spatial standardization. These ICN templates are labeled with different colorful circles and numbers at the center of the circles, representing: the primary visual cortex (medial occipital lobe, ICN01), bilateral primary motor network (ICN02), the default mode network (ICN03), the lateral posterior occipital cortex (ICN04), the right-lateralized frontal-parietal network (ICN05), bilateral ends of the central sulcus (ICN06), the precuneus-dorsal posterior cingulate network (ICN07), the left-lateralized frontal-parietal network (ICN08), the dorsal precuneus-bilateral angular gyri network (ICN09), the anterior cingulate-dorsal prefrontal-angular gyri network (ICN10), the dorsal precuneus-bilateral temporal network (ICN11), and the bilateral superior temporal-inferior frontal network (ICN12).

TABLE 1 | Participant characteristic.

	TCC Experts (N = 22)	Healthy Controls (N = 18)	p
Age (Years)	52.4 ± 6.8	54.8 ± 6.8	0.258
Gender (Males/Females)	7/15	8/10	0.425
Education (Years)	12.2 ± 2.9	11.8 ± 2.9	0.666
TCC Duration (Years)	14.6 ± 8.6	NA	NA
TCC Intensity (Hours/Week)	11.9 ± 5.1	NA	NA
ICV ¹ (Liter)	1.11 ± 0.17	1.12 ± 0.22	0.42
Global fALFF	0.45 ± 0.03	0.44 ± 0.03	0.38
rmsFD ³ (mm)	0.16 ± 0.09	0.12 ± 0.07	0.16

¹ICV, the intracranial volume; ²Global ReHo, Global mean regional functional homogeneity; ³rmsFD, root mean square of frame-wise displacements.

performance than the control group, although this difference was not significant. No significant group difference in accuracy of cognitive performance was detected. To examine the association between TCC practice and cognitive control, we also computed the correlation between these two factors. This showed that RT of cognitive control was negatively correlated with TCC experience ($r = -0.659$; $p = 0.038$).

Group Differences in fALFF among Brain Networks

To test the hypothesis that TCC practitioners might show differential changes in cognitive control-related brain networks relative to controls, we performed a MANOVA analysis controlling for gender, age, and education (see **Table 2**). It was observed that the default network (ICN03) significantly decreased in fALFF for the TCC group compared with the control group ($F = 5.344$, $p = 0.027$). The bilateral frontoparietal network [right FPN (ICN05): $F = 5.491$, $p = 0.025$; left FPN (ICN08): $F = 12.963$, $p = 0.001$] revealed significantly decreased fALFF in the TCC group compared with controls. The anterior cingulate-dorsal prefrontal-angular gyri network (ICN10) in the TCC group also showed a trend of significantly decreased fALFF relative to the control group ($F = 4.108$; $p = 0.05$).

Association between Behavioral Performance and Brain Networks

Correlational analyses were also conducted to examine whether the group differences among those brain networks were also related to the performance of cognitive control in the ANT task in the group of TCC practitioners. Partial r correlation coefficients were computed between the RT and the three fALFF values of the brain networks which were significantly different in the between-group comparisons, to avoid the occurrence of false positive results. We also performed partial correlation analyses between accuracy of the ANT task and the fALFF values in the three brain networks. Logarithmic transformation was also conducted for RT and accuracy of the ANT task since the values of these two groups were distributed non-normally. As **Figure 3** indicates, the results demonstrated that ICN08 was significantly correlated with RT of the ANT task ($r = 0.851$; $p = 0.015$) while no significant correlation was observed in the association between accuracy of the ANT task and brain networks.

Association between TCC Practice and Brain Networks

In our previous structural study on TCC practitioners, we found that intensity of practice is a valid and sensitive indicator of TCC experience (Wei et al., 2013). Hence, intensity of practice was also adopted in the present study to test the different fALFF among the brain networks in the TCC group that might be associated with TCC experience. We performed a partial correlation analysis between TCC experience and fALFF of brain networks controlling for age, sex, and education. The values for intensity of TCC practice were log-transformed for marginally non-normally distributed trend (Shapiro–Wilk Test, $p = 0.056$). It is likely that non-linear relationship may exist in the effect of TCC practice on brain networks. The normality of intensity scores of TCC practice was improved with this transformation (Shapiro–Wilk test, $p = 0.119$). Moreover, one participant had practiced TCC for at least 30 h each week, which is an outlier based on a distributed scatter plot of the descriptive data. We removed this participant from the analysis for calculating the correlation between the remaining 21 practitioners' fALFF in brain networks

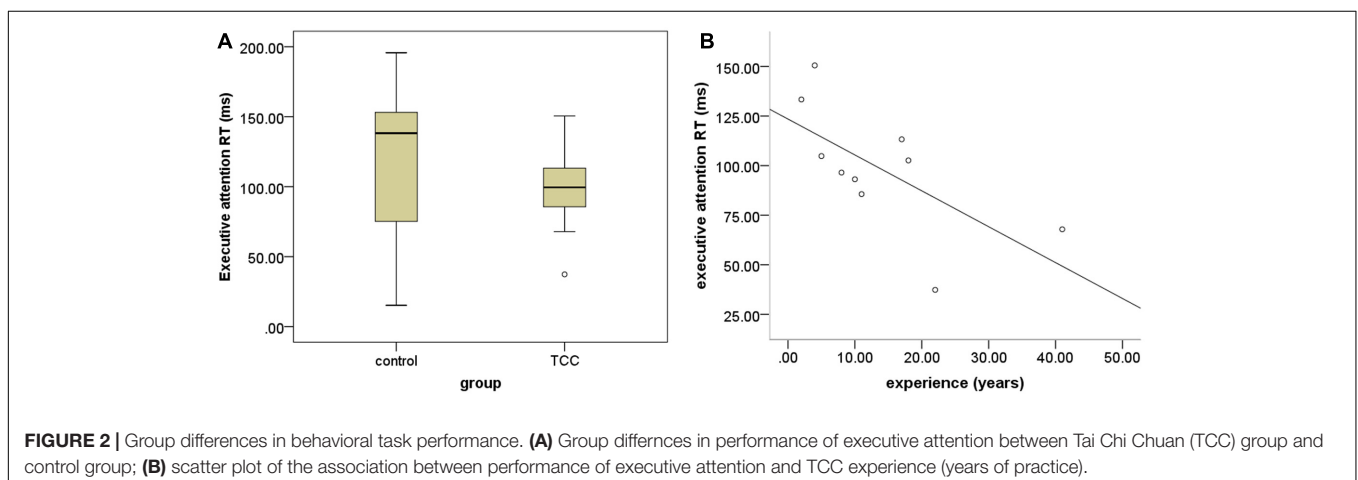
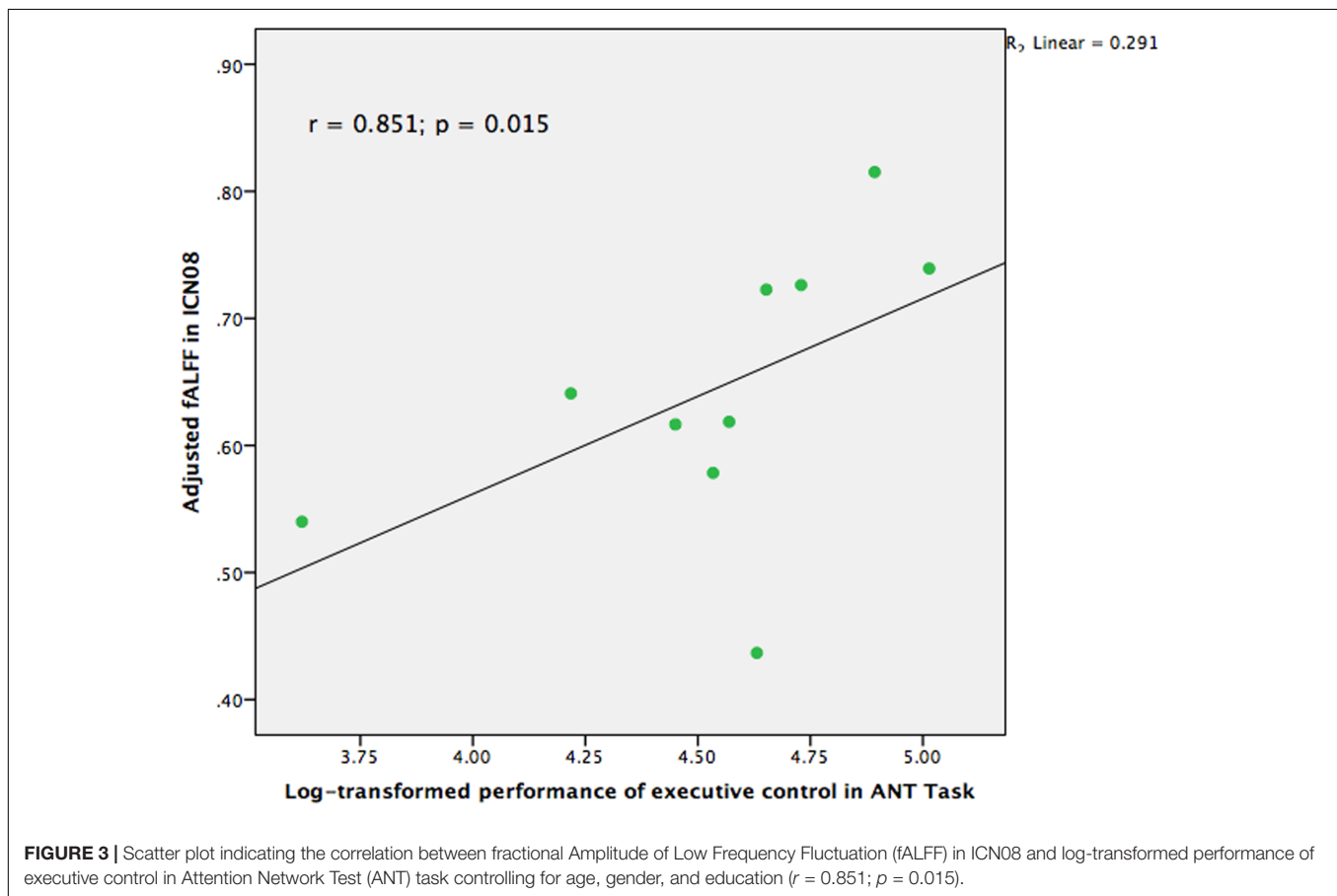


TABLE 2 | Group difference of fractional Amplitude of Low Frequency Fluctuation (fALFF) after controlling gender, age, and education as covariates.

	CTR (<i>n</i> = 18)		TCC (<i>n</i> = 22)		<i>t</i>	<i>p</i>
	<i>M</i>	<i>SD</i>	<i>M</i>	<i>SD</i>		
ICN01	0.738	0.125	0.724	0.145	0.117	0.907
ICN02	0.655	0.117	0.611	0.173	0.793	0.433
ICN03	0.831	0.075	0.751	0.124	2.312	0.027*
ICN04	0.692	0.117	0.699	0.151	-0.396	0.695
ICN05	0.685	0.087	0.614	0.123	2.343	0.025*
ICN06	0.602	0.111	0.545	0.168	1.084	0.286
ICN07	0.738	0.084	0.700	0.095	1.170	0.250
ICN08	0.765	0.079	0.672	0.095	3.600	0.001**
ICN09	0.690	0.103	0.679	0.138	-0.049	0.961
ICN10	0.677	0.084	0.599	0.146	2.027	0.050
ICN11	0.662	0.114	0.660	0.136	0.232	0.818
ICN12	0.718	0.075	0.670	0.122	1.285	0.207

*Indicated $p < 0.05$.**Indicated $p < 0.01$.

and intensity of TCC practice. As **Figure 4** indicates, we observed that the fALFF of ICN03 significantly correlated with the log-transformed intensity of practice ($r = 0.473$, $p = 0.047$). No other significant correlations based upon the transformed intensity in the significantly different brain networks between groups were detected.

DISCUSSION

To our knowledge, this is the first study to specifically examine the association between mind-body practice and large-scale brain networks. In this study, to investigate practice-induced resting low frequency activity in large-scale brain

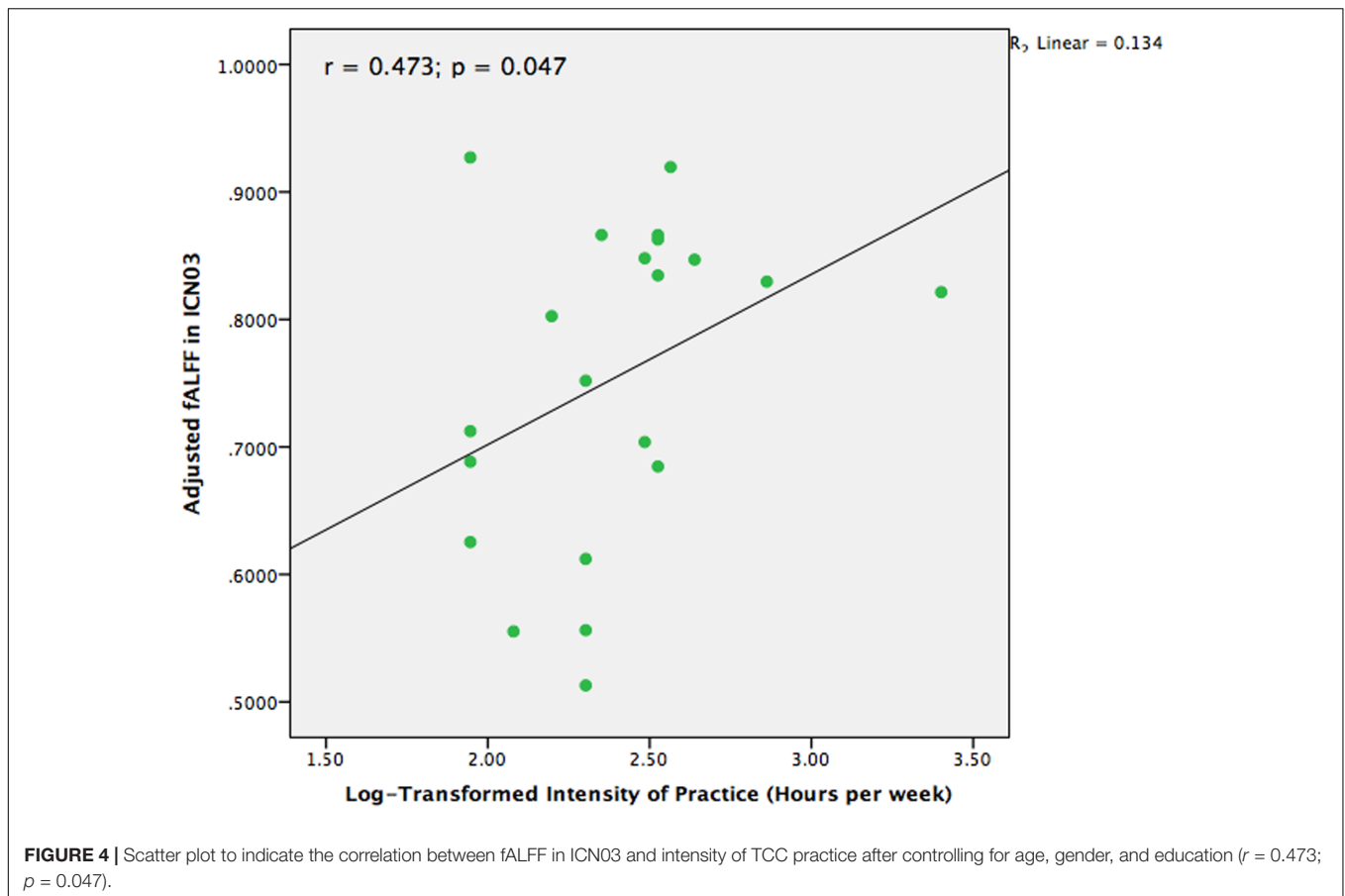


FIGURE 4 | Scatter plot to indicate the correlation between fALFF in ICN03 and intensity of TCC practice after controlling for age, gender, and education ($r = 0.473$; $p = 0.047$).

networks, we used a recently developed measure of inter-subject reproducibility-based algorithms for detection of functional brain networks – gRAICAR. Firstly, we compared the fALFF of brain networks between groups, which showed significant decreases of fALFF in the bilateral frontoparietal network (ICN08 and ICN05) in the TCC group compared to the control group. Additionally, fALFF values in the default mode network (ICN03) and the dorsal prefrontal-angular gyri network (ICN10) were also greatly decreased in practitioners relative to controls. Secondly, we aimed to explore whether extensive TCC practice induced the changed fALFF in brain networks. The results revealed associations between the left lateral FPN and performance of cognitive control, as well as an association between the default mode network and practice experience. This indicates the positive impact of extensive TCC practice on cognitive control-related brain systems.

Functional Plasticity Associated with Mind-Body Practice

This investigation exploring the altered intrinsic cortical network associated with mind-body practice showed that the effects of extensive mind-body training on brain networks were rather selective, being largely located in the frontoparietal, default mode, and dorsal prefrontal-angular networks. The alteration

on the amplitude of low frequency fluctuation in several brain networks, including frontoparietal network, strongly supports our hypothesis. It is suggested that the difference of fALFF in brain networks between the TCC group and the control group possibly reflect experience-dependent neural plasticity. Neural plasticity is the ability to make adaptive changes related to structure and function of the nervous system (Zilles, 1992). Numerous studies on animal models and human species have suggested that training is a key environmental factor to induce morphological alterations in brain areas, changes in neuron morphology, network alterations (including changes in neuronal connectivity), the generation of new neurons, and neurochemical changes (Fuchs and Flugge, 2014). Consistent with this result, previous studies have reported the selective effects of training on brain networks or regions.

Mind-Body Practice and Frontoparietal Network

Evidence from young adults has revealed that intensive reasoning training was associated with increased frontoparietal connectivity (Mackey et al., 2013). Moreover, 2 weeks working memory training was reported to alter the activity pattern of frontoparietal network (Schneiders et al., 2012). Although there are relatively few studies directly focused on the change of FPN associated with mind-body practice, several studies consistently observed such an effect on cognitive control-related brain activity involving prefrontal and parietal cortex.

A cross-sectional study on yoga practitioners observed less reactivity in right dorsolateral prefrontal cortex (involving attention and exerting cognitive control-related function) to negative images compared to neutral images (Froeliger et al., 2012), which indicated the top-down modulation of PFC on emotional regulation by mind-body practice. Recent neuroimaging evidence also found that mindfulness practitioners revealed decreased frontal activation during processing of emotionally aversive experiences (Gard et al., 2012). These findings likely reflected the attitude of acceptance and non-judgment without effortful cognitive control during emotional processing developed after extensive practice. Additionally, mind-body intervention studies using EEG indicated the critical role of prefrontal cortex in cognitive improvement among people with autism and choric epilepsy (Chan et al., 2009, 2011). Similarly, another randomized controlled study showed that Chinese chan-based mind-body intervention significantly improved frontal alpha asymmetry and intra- and inter-hemispheric theta coherence in frontoposterior and posterior brain regions among patients with major depressive disorder (Chan et al., 2013).

Notably, it is well established that the change in functional activity at the cortical level following intensive physical or mental training largely involves prefrontal and parietal regions (based on brain “location specific” approach). The functional plasticity of prefrontal and parietal cortex has been well documented in mindfulness and physical exercise studies, respectively. For instance, following 6 weeks of mindfulness training, dorsolateral PFC responses were increased during executive processing in an emotional Stroop task in healthy human participants (Allen et al., 2012), which indicated such training benefits the recruitment of a top-down mechanism to resolve cognitive conflict. A study showed that greater activation in parietal regions were also found after meditation training in groups with social anxiety (Goldin and Gross, 2010). By contrast, meditation experts were characterized by decreased activation in dorso- and ventrolateral PFC regions compared with controls in cross-sectional studies (Grant et al., 2011; Gard et al., 2012). These findings could be explained by the different demands of cognitive control between the beginning and expertised practice stages. Moreover, short-term mindfulness training studies also observed increased EEG power in the theta frequency at frontal midline electrodes (Tang et al., 2009). Regarding the alteration of brain structures induced by meditation a recent meta-analysis of morphometric neuroimaging in meditation examining approximately 300 meditation practitioners demonstrated that prefrontal cortex showed the most consistent differences between meditators and controls (Fox et al., 2014).

Additionally, convincing evidence from physical exercise studies has demonstrated the effect of physical activity on cognitive control, as well as on prefrontal and parietal cortex (Loprinzi et al., 2013). EEG studies have shown increased neural activity within the prefrontal cortex and improved executive functioning performance following acute physical activity (Hillman et al., 2003). This finding was also confirmed

with other cognitive control paradigms including a switching task and an attentional network test, which showed either reduced latency of neural activity or larger P3 amplitude in the prefrontal and parietal cortices. These results suggest the role of physical activity in improving cognitive performance through mechanisms related to cognitive control (Kamijo and Takeda, 2010; Chang et al., 2015). A recent review has also pointed out the involvement of FPN in initiation and flexible adjustments in cognitive control during engaging physical activity behavior (Buckley et al., 2014).

Mind-Body Practice Experience and Default Mode Network

To understand the question of whether mind-body practice resulted in the change of DMN, we compared the difference of fALFF in DMN between two groups and assessed the relationship between DMN changes and practice experience. The results showed that the fALFF in DMN was significantly different in TCC practitioners relative to controls and were positively correlated with practice intensity (practice hours/week). The association between experience and changed fALFF in DMN generally reflected experience-dependent functional plasticity, which confirmed the effect of mind-body practice on the functional pattern of DMN. Currently, an increasing amount of evidence consistently demonstrates a pattern of deactivation during a task-invoked state as well as activation across a network of brain regions during resting-state, including medial, lateral, and inferior parietal cortex, precuneus/posterior cingulate cortex (PCC) and medial prefrontal cortex, and (Raichle et al., 2001). This network is defined as the default mode network and is characterized by coherent low frequency neuronal oscillations, which reflect the associated psychological functions of introspection and self-referential thought (Broyd et al., 2009). An extensive body of research defines the DMN to be one of the critical networks of the human brain. It can also be altered by various practices. Consistent with this study, prior investigations of the link between large-scale brain network pattern and physical or mental training have also pointed to the importance of the default mode network. Voss et al. (2010a) adopted a seed-based functional connectivity analysis examining the association between aerobic fitness, cognitive performance and functional connectivity in the default mode network, which concluded that both specific and global default mode networks mediated the relationship between aerobic fitness and cognition. Furthermore, another 12-month randomized interventional study also observed that exercise training increased functional connectivity between some brain regions within the default mode network in elderly adults. This provided the first evidence for exercise-induced functional plasticity in large-scale brain systems in the aging brain (Voss et al., 2010b). In parallel to this, the evidence from mindfulness studies has consistently detected training-induced functional and structural changes in DMN, which suggests DMN plays a pivotal role in processes of internal mentation. For example, a cross-sectional study among meditators with extensive training demonstrated that meditation training can lead to functional connectivity changes between core DMN regions, possibly reflecting strengthened present-moment

awareness (Taylor et al., 2013). Meditation training has also been observed to increase the functional connectivity within DMN in elderly adults with mild cognitive impairments (Wells et al., 2013), which could be interpreted as a role of DMN of attenuating cognitive aging. A recent review on meditation suggested DMN is a biomarker for monitoring the therapeutic effects of meditation in mental disorders (Simon and Engstrom, 2015).

Tai Chi Chuan is a typical mind-body practice that is performed using a series of graceful concentric and eccentric movements that are linked together in a continuous sequence in semisquat positions. During the performance of TCC, deep breathing, slow movements, and mental concentration are required to achieve harmony between body and mind. Thus, TCC practice combines key components of various practices such as aerobic exercise and meditation (Yeh et al., 2014). Integrating these training factors may produce multiple effects. Thus, the findings of FPN and DMN changes in our study contribute to the multiple outcomes of long-term TCC practice, combining components of aerobic exercise and mindfulness.

FPN, Possible Neural Correlate Underlying the Effect of Mind-Body Practice on Cognitive Control

In this study, we observed that the TCC group had decreased fALFF in bilateral FPN as well as an association between cognitive control performance and fALFF in FPN. Alternatively, TCC practice optimizes the spontaneous activity of FPN, coupling with enhanced cognitive behavior performance. It is plausible that an optimized pattern of FPN might play a role in the effect of mind-body practice on cognitive control. Although further mediation analysis didn't show any significance to confirm fALFF in FPN mediated the effect of TCC practice on cognitive control, it is reasonable to infer that low frequency oscillation of regional brain function in FPN might be neural correlate underlying the effect of TCC practice on behavioral performance since FPN has highly flexible and variable connectivity throughout the brain, the functional connectivity of FPN with other brain systems and the global brain system. Given that a system refers to a set of widely distributed brain regions that exhibit consistently correlated spontaneous activity fluctuations, and characteristically respond toward a specific task, the brain is organized into multiple systems that have distinct and potentially competing functional roles. Recently, several big sample studies, across multiple datasets that aimed to explore the organization of human cerebral cortex estimated by intrinsic functional connectivity, have identified the frontoparietal control system as supporting cognitive control, including middle and superior prefrontal cortex (BA6, BA8, BA46, BA47, and BA10), superior parietal lobule (BA47), and inferior parietal lobule (BA 40) (Vincent et al., 2008; Yeo et al., 2011). Regions within the frontoparietal network showed similar functional connectivity fingerprints even when distributed across the cortex. Previous task-fMRI studies also indicated that many regions in the frontoparietal control system are activated during tasks requiring cognitive control or executive function (Hon, 2007). Furthermore, it is pointed out that this

control system is also involved in integrating information coming from the other systems and to adjudicate between potentially competing inner- versus outer-directed processes (Vincent et al., 2008). Convincing evidence suggests that the human ability to adaptively implement a wide variety of tasks is primarily a result of the operation of the frontoparietal brain network. More recently, Cole et al. (2013) found that FPN's brain-wide functional connectivity pattern shifted more than those of other networks across a variety of task states and that these connectivity patterns could be used to identify the current task. It was further confirmed that the frontoparietal network implements domain-general functions (e.g., the cognitive control system) made by flexible hubs (Cole et al., 2013). In view of the flexible hubs largely existent in FPN, we infer that extensive mind-body training exerts a preferential influence on general cognitive control among other multiple specific effects including self-awareness, self-regulation, proprioception, and goal planning. Notably, it has been emphasized that cognitive control showed the largest benefit of improved fitness among the different process-task types induced by physical exercise (Colcombe and Kramer, 2003; Chang et al., 2010). On a behavioral level, previous studies have observed such an effect of cognitive control following short-period TCC practice (Matthews and Williams, 2008). Intriguingly, our study also revealed that the TCC group showed marginally significant better performance than the control group. Moreover, the performance of cognitive control in the TCC group was correlated with TCC experience. These results further demonstrate the critical role of cognitive control during TCC practice. Hence, FPN, being responsible for general cognitive control, showed greater difference of regional brain function induced by mind-body practice relative to other brain systems.

Enhanced Cognitive Control Capacity via Multiple Feedbacks As Outcomes of TCC Practice

As stated above, based on the evidence from neuroimaging approaches, a wide variety of mental disorders involve impaired cognitive control abilities and altered function in control system. Cole et al. (2014) suggested the mechanism of mental health using the framework of brain feedback control: a control system consisting of flexible hubs that use feedback control to regulate symptoms and so promote mental health. Hence, an effective control system would be protective against a variety of mental diseases. In view of the findings of our study, we propose that TCC practice is an efficient means to reach the goal of successful feedback control via the flexible hubs of the frontoparietal network. Then how does this mind-body practice execute this function among multiple and complex brain systems? More generally, cognitive control capacity can vary substantially both within and across individuals. The reduction of the control system might be influenced by excessive stress, cognitive load, and negative affect. By contrast, motivation, effective strategies for the goal, and adaptive habits could increase cognitive control capacity (Cole et al., 2014). It is likely that TCC could enhance the cognitive control system

to perform indirect feedback control via multiple strategies such as deep breathing, mindfulness, and attention control, which is supported by relevant TCC studies on mental diseases. Previous short-period intervention studies suggested that TCC could be regarded as a treatment strategy to promote cognitive function in adults with cognitive impairment (Li et al., 2014) and cerebral vascular disorder (Wang et al., 2010), as well as depression, anxiety, and sleep disturbance (Field et al., 2013). We speculate that some health-related key components contained in TCC practice represent multiple channels of feedback, which accumulatively enhance cognitive control effects as outcomes of TCC practice.

Firstly, deep breathing, one of the key components of TCC, is increasingly used for its relaxation effect as a complementary and alternative medicine for maintaining general health, as well as treating myriad diseases. Several studies investigating TCC have confirmed that TCC practice could increase vagal activity and the balance between sympathetic and parasympathetic activity via increasing heart rate variability (HRV) (Lu and Kuo, 2003, 2014). We have also previously observed improved vagal modulation during deep breathing by comparing the HRV of TCC practitioners and controls (Wei et al., 2016). Although the underlying neural mechanism is unclear as to how deep breathing establishes the temporary neural circuits relevant to the control system, we speculate that TCC practice is beneficial to modulating the cognitive control system by increasing functional connectivity between the frontoparietal network and the corresponding cortical and subcortical structures dominating deep breathing. Meditation studies have reported that some brain regions which have stable anatomical and functional connectivity with the frontoparietal network, such as anterior cingulate cortex and insula, are responsible for the activity of the autonomic nervous system (Tang et al., 2009). Hence, it is likely that the functional connectivity between the frontoparietal network and these brain regions contributes to the execution of indirect feedback toward the control system. Secondly, attention control is also considered one of the key characteristics of TCC for the requirement of harmony between movement and mental activity. There is growing evidence that mindful meditation could induce functional changes of dorsal lateral prefrontal cortex and anterior cingulate cortex supporting attention control processes (Tang et al., 2015). Prefrontal cortex is the critical flexible hub existing in the frontoparietal network (Cole et al., 2013), while anterior cingulate cortex has relatively close connections in anatomical and functional connectivity with frontoparietal network. This partly determines the role of this component of TCC in the feedback channel. Thirdly, aerobic exercise, the last but not least component of TCC, has been extensively demonstrated to change GM volume (Colcombe et al., 2006), cortical thickness (Wei et al., 2013), and regional brain function (Colcombe et al., 2004) at the cortical level in prefrontal and parietal cortex, which is consistently implicated in cognitive control processing. In view of a long-term strategy, TCC practice might facilitate such established, long-term, body feedback by these specific brain regions to execute control-related feedback. Taken together, multiple feedback

channels benefited by extensive TCC practice might be one of the key correlates underlying enhance cognitive control capacity to further perfect the “immune system” of mental health.

Limitations

The results and interpretations of this study must be considered with several limitations. One limitation is that the cross-sectional examination between mind-body practice and brain networks could not completely exclude the effect of some confounding factors such as predisposition, preexisting characteristics of brain structure and function, and intelligence, although we controlled for age, gender, and education level when examining the group differences in fALFF and the correlation between behavior and the altered brain networks. Longitudinal studies will help delineate the beneficial effect of mind-body practice on resting brain state and will be needed in future research. Second, the findings of this study should be interpreted with caution given the relatively small sample size. In particular, the data of half the sample's behavioral performance had not been collected, which possibly covers up more significant correlations in other brain networks. Third, although several clinical studies using gRAICAR showed good reliability in subject grouping, it still needs examination in normal populations with little variability compared to patients diagnosed for neurological diseases. Thus, an important area of future study could utilize this brain parcellation on the ICN to carry on the investigation in a normal population. Hopefully, it is promising to apply this to a longitudinal prevention study on mind-body practice.

CONCLUSION

In sum, the present investigation firstly jointly employ MRI and behavioral methodologies to examine the link between mind-body practice and large-scale brain networks. A clear association between the cognitive control and frontoparietal networks is demonstrated. It is inferred that frontoparietal networks is likely to be the neural correlates underlying the effect of TCC practice on cognitive control. The results also extend previous research that has been based on a brain-specific location approach. The decreased fALFF in the left-lateralized frontoparietal network in the TCC practice group relative to the control group might reflect the improved cognitive control capacity associated with TCC practice. Other functional networks including the default network, the right-lateralized frontal-parietal network, and the dorsal prefrontal-angular gyri network are also an indication of functional optimization possibly induced by mind-body practice. Furthermore, the findings also carry significant public health implications. Although this study did not focus on patients with cognitive impairment, the optimized functional pattern in the frontoparietal network still help to unravel the partial neural correlates underlying the effect of mind-body practice on enhancing cognitive control capacity. This will hopefully encourage government to consider mind-body intervention in the treatment and prevention of illness and disease.

AUTHOR CONTRIBUTIONS

G-XW designed the work, drafted and finalized manuscript; Z-QG, ZY, and X-NZ analyzed data and revised manuscript.

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The Role of Medial Frontal Cortex in Action Anticipation in Professional Badminton Players

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Some studies show that the medial frontal cortex is associated with more skilled action anticipation, while similar findings are not observed in some other studies, possibly due to the stimuli employed and the participants used as the control group. In addition, no studies have investigated whether there is any functional connectivity between the medial frontal cortex and other brain regions in more skilled action anticipation. Therefore, the present study aimed to re-investigate how the medial frontal cortex is involved in more skilled action anticipation by circumventing the limitations of previous research and to investigate that the medial frontal cortex functionally connected with other brain regions involved in action processing in more skilled action anticipation. To this end, professional badminton players and novices were asked to anticipate the landing position of the shuttlecock while watching badminton match videos or to judge the gender of the players in the matches. The video clips ended right at the point that the shuttlecock and the racket came into contact to reduce the effect of information about the trajectory of the shuttlecock. Novices who lacked training and watching experience were recruited for the control group to reduce the effect of sport-related experience on the medial frontal cortex. Blood oxygenation level-dependent activation was assessed by means of functional magnetic resonance imaging. Compared to novices, badminton players exhibited stronger activation in the left medial frontal cortex during action anticipation and greater functional connectivity between left medial frontal cortex and some other brain regions (e.g., right posterior cingulate cortex). Therefore, the present study supports the position that the medial frontal cortex plays a role in more skilled action anticipation and that there is a specific brain network for more skilled action anticipation that involves right posterior cingulate cortex, right fusiform gyrus, right inferior parietal lobule, left insula and particularly, and left medial frontal cortex.

Keywords: action anticipation, fMRI, functional connectivity, professional badminton players, medial frontal cortex

INTRODUCTION

From an evolutionary perspective, the anticipation of an upcoming action based on environmental cues is of great importance for individuals' survival, as it may help individuals estimate other people's or animal's intentions in order to better prepare adaptive reactions to approach safety and avoid harm. Action anticipation is still important to human life, be that in daily engagement, such as driving a car or avoiding a moving object, in sports, or in combat competition. Therefore, how action anticipation influences individuals' behavior and neural activity has been an important topic of research in psychology and human neuroscience. Expert players in interceptive sports such as badminton reacting under time pressure provide a helpful model to explore the aforementioned issue. Using the temporal occlusion paradigm, in which action is cut off at various time intervals relative to a crucial event, such as right when the shuttlecock contacts the racquet in badminton, behavioral studies have repeatedly demonstrated that compared with non-players, professional players can better predict the outcomes of other players' sequential movements (e.g., Abernethy, 1990; Vickers, 1992; Ripoll et al., 1995; Williams and Davids, 1998; Williams and Elliott, 1999; Wolpert et al., 2003; Wilson and Knoblich, 2005; Schutz-Bosbach and Prinz, 2007; Aglioti et al., 2008; Bubic et al., 2010; Brown and Brune, 2012).

Using similar approaches, studies have also investigated whether more skilled action anticipation can be reflected in neural activity. However, the findings were mixed, particularly regarding whether the medial frontal cortex plays a role in more skilled action anticipation. Wright et al. (2010, 2011) found that compared to novices, badminton players exhibited enhanced activation in the medial frontal cortex when anticipating the direction of a badminton stroke. However, other studies did not replicate this effect. For example, Wright et al. (2013) did not find differential activation of the medial frontal cortex between high-skilled and low-skilled male soccer players when the players observed video clips of a person dribbling a ball and were required to predict the direction of the ball. Similarly, when participants were asked to predict a basketball shot's fate, differential activation of the medial frontal cortex was not observed between athletes and novices (Abreu et al., 2012; Wu et al., 2013).

Previous studies have suggested that the medial frontal cortex plays a role in anticipation in general, when the anticipated content is unrelated to sports actions (Ridderinkhof et al., 2004; Moriguchi et al., 2007; Carrington and Bailey, 2009; Waszak et al., 2012; Corradi-Dell'Acqua et al., 2015). Therefore, activation of the medial frontal cortex should be expected during action anticipation. The fact that the studies mentioned above did not observe an effect of sports action anticipation on the medial frontal cortex may be related to the stimuli employed and the participants included in the control group. With respect to the stimuli, in previous studies, the movie clips contained information not only about the actions of the players but also about the trajectory of the ball. For example, the movie clips in Abreu et al. (2012) ended just before the ball hit the basket, fell short of it or surpassed it. Aglioti et al. (2008) found that the

accuracy of action anticipation of elite players was higher than that of novices when the clip stopped right at the point the ball left the player's hand and initiated its own trajectory; however, there were no differences between these two groups when the clips ended after the ball started its own trajectory, at the point just before the ball hit the basket. These findings suggest that the novices relied on the trajectory of the ball to perform the task. Additionally, our previous studies (Jin et al., 2010, 2011) showed that in novices, accuracy was at chance level when the clips ended right at the point that the shuttlecock contacted the racket; whereas accuracy was comparably high in this group when the shuttlecock had finished 1/3 or 2/3 of its trajectory between its point of contact with the racket and touching the ground. These findings also indicate that in novices, the ability to anticipate actions was enhanced when the clips displayed the trajectory of the shuttlecock. Showing more trajectories may allow individuals to have more cues to anticipate the outcome of the shuttlecock. This may help to enhance the accuracy of the anticipation and reduce the task difficulty for both groups, resulting in reducing group differences in the ability of anticipation and as a result, weakening the effect of more skilled action anticipation on the medial frontal cortex. Therefore, using stimuli that include the trajectory of the ball might reduce group differences in the ability of anticipation and, as a result, weaken the effect of more skilled action anticipation on the medial frontal cortex.

With regards to the participants included as the control group, while they should clearly not be professional or elite players of the related sports, in some studies, these participants were not complete "novices." That is, the participants had some training or watching experience. For example, Wright et al. (2013) recruited low-skilled players as the control group, some of whom had previous experience that included playing for local sport clubs or school teams for more than 1 year. This experience may have enhanced the action anticipation abilities of the controls, which could reduce the differential activation of the medial frontal cortex between the players and the controls.

Therefore, the first aim of the present study was to re-investigate whether the medial frontal cortex is involved in more skilled action anticipation. To address this issue, badminton players and novice participants were presented video clips from international badminton matches and were asked to anticipate the location of the shuttlecock. To remove the effect of the trajectory of the shuttlecock, the clips ended right at the point that the shuttlecock and the racket came into contact. Participants without any training or watching experience were recruited for the control group. Based on previous studies (Wright et al., 2010, 2011), we predicted that during action anticipation, activation of the medial frontal cortex would be stronger in the badminton players compared to the novices.

In addition, cognitive task performance likely depends on connections between several brain regions. Accordingly, if the medial frontal cortex plays a role in more skilled action anticipation, this brain region may be connected more strongly with other brain regions involved in action processing, especially for more skilled action anticipation. Therefore, the second aim of the present study was to investigate how the medial frontal cortex was functionally connected with other brain regions needed for

successful action anticipation. Based on evidence that experts in interceptive sports (e.g., badminton and basketball) are similar to experts in action video games, who exhibit a high level of multiple cognitive functions, including attention, executive control, and hand-eye coordination (Kida et al., 2005; Russo et al., 2006; Nakamoto and Mori, 2008a,b; Voss et al., 2010; Vestberg et al., 2012; Bejjanki et al., 2014), we predicted that expert players with more skilled action anticipation might exhibit greater functional connectivity between the medial frontal cortex and posterior regions, as observed in action video games experts during video game play (Gong et al., 2015, 2016).

MATERIALS AND METHODS

Participants

Sixteen badminton players (14–37 years, $M \pm SD = 22.54 \pm 5.15$ years, 11 males) were recruited for the study. Players were members of professional teams or professional university teams and had completed at least 3 years of badminton training (mean = 8.81 years, range from 3 to 16 years). The control group consisted of 18 healthy novices (17–37 years, $M \pm SD = 21.09 \pm 4.27$ years, 8 males) who had no professional or amateur training in badminton or other racquet sports or no watching experience of badminton or other racquet sports. Players and novices did not differ in age ($p > 0.05$) or level of education. All participants were right-handed as determined by the Edinburgh Handedness Inventory (Oldfield, 1971). Participants had normal or corrected-to-normal vision and no participants had a history of neurological illness. All participants gave written informed consent prior to the study. The study was approved by School of Psychology, South China Normal University.

Stimuli

The stimuli consisted of 40 color video clips (wmv format, 25 frames per second, including 16 practice clips) of single matches in world tournaments, as described in detail in our previous study (Jin et al., 2011, 2010). The clips depicted a player struck the shuttlecock away and an opponent receiving the shuttlecock and ended when the opponent's racket contacted the shuttlecock. The clips did not provide any visual information about the shuttlecock's final location or the trajectory of the shuttlecock. All of the clips lasted for 480 ms. The resolution of all the clips was 768×576 pixel. The resolution of the monitor was 1024×768 pixel with the refresh rate of 60 Hz. Examples of the stimuli were shown in **Figure 1**.

Procedure

fMRI scanning consisted of four runs. Each run included two blocks, which varied across tasks (anticipation and gender). The sequence of the blocks was counterbalanced across the participants. Each block consisted of 20 video clips. Therefore, there were 160 trials in total (20 trials per condition \times 2 conditions \times 4 runs). A 20 s fixation period took place between blocks. Each block started with a 2-s instruction period. The video clips were presented for 480 ms, and the mean interval

between two video clips was 3020 ms, with a range from 2520 to 3520 ms. During the presentation of the video clips, the participants were asked to pay attention to the clips. During the anticipation block, the participants were also asked to predict whether the shuttlecock would land in the forecourt or the backcourt of the players (but not the opponents') while the clips were presented or during the following random interval. During the gender block, the participants were told to guess the gender only of the competitors. Both of the tasks emphasized speed and accuracy. Responses were given by pressing the corresponding button on a two-button pad. Response assignments were counterbalanced across participants. Stimulus presentation and the recording of behavioral responses were accomplished with E-prime software.

Behavioral Data Recording and Analysis

For both the anticipation and the gender tasks, response accuracy and time were recorded for each video clip. Response accuracy was analyzed by one-sample test with a test value of 0.5 separately for task (anticipation versus gender) and group (player versus novice). Response accuracy and time were analysed by repeated measures analysis of variance (ANOVA) with task (anticipation versus gender) as a within-subject factor and group (player versus novice) as a between-subjects factor using SPSS 17.0 software (SPSS Inc., Chicago, IL, USA). All data are expressed as the $M \pm SD$ or SE .

fMRI Data Acquisition and Analysis

The subjects were scanned by a Siemens 3T Trio scanner with a standard single-channel head coil. The stimuli were presented through an LCD projector onto a rear projection screen, which was located behind the participant's head inside the magnet bore. The stimuli were presented centrally at a viewing distance of approximately 60 cm. We acquired the functional ($T2^*$ -weighted) images using blood oxygenation level-dependent (BOLD) contrast (TR = 2000 ms, TE = 30 ms, field of view = 200×200 mm, flip angle = 90° , matrix = 64×64 , 32 slices/volume, in-plane resolution = $3.125 \text{ mm} \times 3.125 \text{ mm}$, no gap). For each run, the first two volumes were discarded to ensure steady-state tissue magnetization. For each participant, a $T1$ -weighted anatomical MRI was also acquired (TE = 1.64 ms; field of view = 256×256 mm, flip angle = 7.0° , matrix = 256×256 , 176 slices, voxel size = $1 \text{ mm} \times 1 \text{ mm} \times 1 \text{ mm}$, no gap).

Preprocessing and whole-brain analyses were performed using SPM8 (Wellcome Trust Center for Neuroimaging, UCL, London, UK) implemented in Matlab 7.9. Functional connectivity analyses were conducted with the functional connectivity toolbox version 15 (CONN¹; Whitfield-Gabrieli and Nieto-Castanon, 2012). For the pre-processing, the volumes were realigned to the first volume to minimize the effects of head movements. Then, the functional and anatomical data were co-registered. During normalization, we resampled the images at a voxel size of $3 \text{ mm} \times 3 \text{ mm} \times 3 \text{ mm}$ and smoothed them with an $8 \text{ mm} \times 8 \text{ mm} \times 8 \text{ mm}^3$ FWHM Gaussian kernel.

¹www.nitrc.org/projects/conn

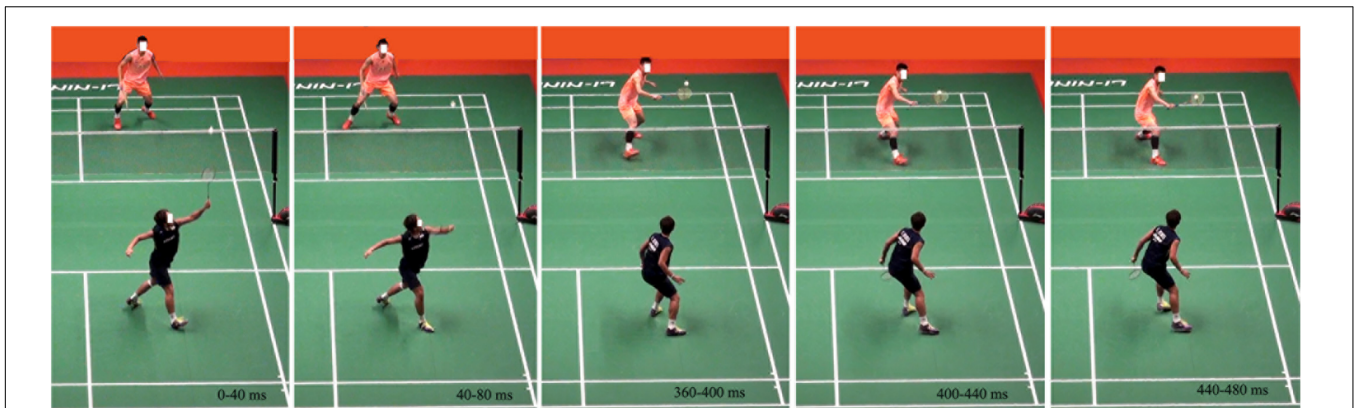


FIGURE 1 | Snapshots of a clip (480 ms in length) illustrating stimuli used in the experiment. The clips ended at the point when the opponent’s racket touches the shuttlecock. Numbers on each frame indicate the time range of each frame (40 ms per frame). Neither the numbers nor the white square occluding the face appeared in actual experimentation.

For the whole-brain analyses, statistical analyses based on a general linear model (GLM) were performed first at the participant level and then at the group level. At the participant level, a fixed-effects GLM was specified for each participant. Task-related changes in BOLD signal at the onset of each video clip were modeled as a delta function convolved with a haemodynamic response function (HRF). Task-related contrast was performed for each participant: anticipation > gender. The resulting contrast was then entered into separate second-level analyses, where group (player versus novice) served as a between-subjects variable in independent sample *t*-tests. The threshold in SPM was initially set to $p < 0.001$, uncorrected. Regions with $k > 10$ voxels that survived small volume correction at $p < 0.05$, FWE corrected, are reported.

In the functional connectivity analysis, a seed-based correlational analysis was used to identify the intrinsic functional connectivity of the seed, the left medial frontal cortex, across the whole brain. The left medial frontal cortex was defined based on the study by Wright et al. (2010) to avoid independent error (Vul et al., 2009). Regional time series for the seed region were extracted from bandpass-filtered images with a temporal filter (0.008–0.09 Hz). An anatomical-component-based noise correction method (aCompCor) was used to reduce noise (Behzadi et al., 2007). Global signal regression was not included to avoid potential false anti-correlations (Murphy et al., 2009). At the participant level, a GLM was used to assess significant BOLD signal correlation between the medial frontal cortex and each voxel with respect to anticipation/gender. The toolbox converted the resulting correlation coefficients to *Z*-values using Fisher transformation for subsequent *t*-tests. Differences were also examined at the group level. For the statistical parametric maps that were produced by voxel-wise analysis, clusters were considered statistically significant if they survived multiple comparisons correction. We used the approach implemented in AlphaSim² based on a 3D extension of the randomization procedure described by Forman et al. (1995). The voxel-level

threshold was initially set to $p < 0.001$ (uncorrected). The correction criterion was based on an estimate of the maps’ spatial smoothness and on an iterative procedure (Monte Carlo stimulation) for estimating cluster-level false-positive rates. After 1000 iterations, AlphaSim determined that an image-wide threshold of $p < 0.05$ required a cluster with 22 contiguous voxels for significance.

RESULTS

Behavioral Data

The response accuracy results showed that for players, the mean accuracy for the anticipation and gender tasks was above the 0.5 chance level [$t_{(15)} = 6.33, p < 0.001$; $t_{(15)} = 41.60, p < 0.001$]. For novices, the mean accuracy for the gender task was significantly higher than 0.5 [$t_{(17)} = 17.55, p < 0.001$], but this was not the case for the anticipation task ($ps. > 0.05$). Additionally, the ANOVA showed main effects of task [$F_{(1,32)} = 116.13, p < 0.001, \eta_p^2 = 0.78$] and group [$F_{(1,32)} = 38.70, p < 0.001, \eta_p^2 = 0.55$]. Accuracy was higher for the gender compared to the anticipation task and for players compared to novices. More importantly, the interaction between these two factors was also significant [$F_{(1,32)} = 10.73, p = 0.004, \eta_p^2 = 0.23$]. Further analysis showed that accuracy was higher for players compared to novices in both the anticipation [$F_{(1,32)} = 26.77, p < 0.001, \eta_p^2 = 0.46$] and the gender task [$F_{(1,32)} = 16.18, p < 0.001, \eta_p^2 = 0.34$], although to a different extent. For descriptive data, please refer to **Table 1**.

TABLE 1 | Mean accuracy (%) and its standard errors (SE) for each experimental condition.

	Predicted task		Gender task	
	<i>M</i>	<i>SE</i>	<i>M</i>	<i>SE</i>
Experimental group	74.06	3.81	95.56	1.75
Control group	47.00	3.59	85.89	1.65

²<http://afni.nimh.nih.gov/afni/doc/manual/AlphaSim>

Response times were shorter for the gender compared to the anticipation task [$F_{(1,32)} = 110.20, p < 0.001, \eta_p^2 = 0.78$]. In the present study, limited cues were provided for the anticipation task, particularly when the trajectory of the shuttlecock was not presented. However, this seems to be not the case for the gender task. Participants could be able to identify the gender relying on some other cues (e.g., hair and clothing). Therefore, it may be easier for participants to perform the gender as compared to the anticipation task, resulting in observing shorter reaction time. In addition, while the interaction between task and group was significant [$F_{(1,32)} = 11.78, p = 0.002, \eta_p^2 = 0.27$], we did not find a group effect in the anticipation ($ps. > 0.05$) and gender tasks ($ps. > 0.05$). For descriptive data, please refer to **Table 2**.

fMRI Data

Whole Brain Analysis

The results of the whole-brain analysis are shown in **Table 3**. The players compared to novices showed stronger activation in the

left middle frontal gyrus and left medial frontal gyrus (**Figure 2**) and in the right inferior frontal gyrus and right inferior occipital gyrus. In contrast, the novices showed no higher activation in any brain regions compared to players.

Functional Connectivity Analysis

The brain regions that were connected with the left medial frontal cortex in relation to more skilled action anticipation are shown in **Table 4**. The left medial frontal seed region showed greater functional connectivity with the left insula and with the right posterior cingulate cortex, fusiform gyrus and inferior parietal lobule in players compared to novices (**Figure 3**). However, the novices did not show stronger connections between the left medial frontal cortex and other brain regions compared to players.

DISCUSSION

The present study aimed to re-investigate whether more skilled action anticipation is associated with activation of the medial frontal cortex by circumventing the limitations of previous research. It also aimed to investigate how the medial frontal cortex had functional connectivity with other brain regions involved in action processing in more skilled action anticipation.

Consistent with previous studies, the behavioral results showed that response accuracy was higher for badminton players

TABLE 2 | Mean response times (ms) and its standard errors (SE) for each experimental condition.

	Predicted task		Gender task	
	M	SE	M	SE
Experimental group	1135.97	67.01	699.74	32.55
Control group	970.16	63.18	748.91	30.69

TABLE 3 | Results of the whole brain analysis

Region	BA	cluster	Peak t	MNI coordinates		
				X	Y	Z
Predicted task: experimental group > control group						
Left fusiform gyrus	37	22	4.24	-42	-39	-12
			4.21	-48	-45	-9
Left middle frontal gyrus	6	35	4.22	-42	12	51
			3.56	-36	3	60
Left superior frontal gyrus	8/32	70	4.21	-18	36	48
			4.1	-24	30	57
			3.7	-15	27	39
Right medial frontal gyrus	8	42	4.16	6	36	54
			3.82	-3	33	60
			3.61	18	33	51
Right medial frontal gyrus	10/32	33	4.15	18	48	0
			3.8	6	54	12
Left inferior parietal lobule		17	3.71	-48	-39	27
Gender task: experimental group > control group						
Left medial frontal gyrus	10	89	4.25	-12	36	-9
Predicted task - Gender task: experimental group > control group						
Left middle frontal gyrus	9	20	4.25	-39	18	30
Right inferior occipital gyrus	18/19	18	4.03	36	-78	-9
			3.78	39	-81	0
Right inferior frontal gyrus	47	14	3.94	39	27	-15
Left medial frontal gyrus	8/32	22	3.68	6	36	30
			3.63	-6	42	27

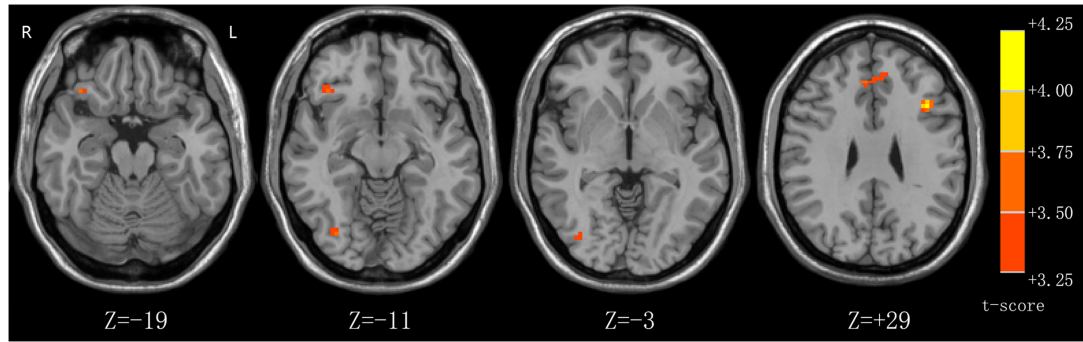


FIGURE 2 | Enhanced activations in the left medial frontal cortex to players (anticipation – gender) as compared to novices (anticipation – gender). Statistical parametric maps are overlaid on a T1 scan.

TABLE 4 | Results of functional connectivity analysis.

Seed	FC Region	BA	K	MNI coordinates			Beta	t
				X	Y	Z		
Experimental group > Control group: predicted task – gender task								
Left medial frontal gyrus(BA9)	Right fusiform gyrus		108	28	-66	-10	0.24	4.66
	Left insula		67	-26	24	8	0.22	4.65
	Right inferior parietal lobule	40	63	48	-54	56	0.19	5.08
	Right posterior cingulate	29	60	18	-42	24	0.2	5.27

FC region refer to which have functional connection with seed; K is the size of cluster; beta is the strength of connection; $p < 0.001$ (uncorrected), cluster size > 40.

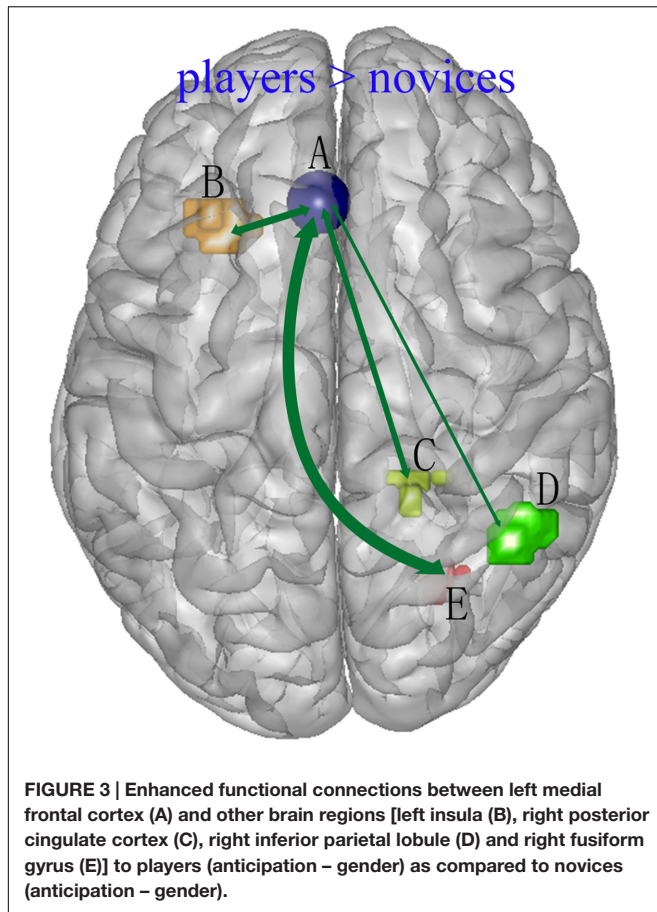
compared to novices; the novices performed the task at chance level, while this was not the case for the players (Singer et al., 1996; Abernethy et al., 2001; Rowe and McKenna, 2001; Williams et al., 2002, 2011; Cañal-Bruland et al., 2011; Jin et al., 2011). These findings demonstrate the validity of participant selection, which may be critical for successfully observing differential activation of the medial frontal cortex between the two groups.

The imaging data results showed that activation of the left medial frontal cortex was stronger in badminton players compared to novices during action anticipation, indicating that the medial frontal cortex plays a role in more skilled action anticipation. Previous studies have shown that the medial frontal cortex is involved in the anticipation of mental states (e.g., beliefs and desires; Keysers and Gazzola, 2007; Moriguchi et al., 2007; Carrington and Bailey, 2009; Corradi-Dell’Acqua et al., 2015). In a recent study by Corradi-Dell’Acqua et al. (2015), the medial orbitofrontal cortex was associated with predicting rule-based behavior. Taking previous studies and the present study together, the findings support that the left medial frontal cortex is associated with anticipation, including action anticipation.

Inconsistent with the findings of the present study, several previous studies did not find an effect of more skilled action anticipation (Abreu et al., 2012; Wright et al., 2013; Wu et al., 2013). As mentioned in the introduction, whether or not a significant effect is identified may be related to whether the participants were presented with the trajectory of the moving object and whether the participants in the control group

had any experience with the related sport. In badminton, when the trajectory of the shuttlecock was presented; the nature of the anticipation became more concrete, which may be easier for participants to anticipate the location of the shuttlecock and reduce the task difficulty as a result. The simplified task may reduce the involvement of medial frontal areas for both players and novices, resulting in failing to observe the effects of skilled action anticipation. In the present study, we used clips that displayed only body kinetic information of the opponent and not information about the trajectory of the shuttlecock. We also recruited novices without any training or even watching experience. Under these conditions, we found enhanced activation of the medial frontal cortex during action anticipation in players. Therefore, information about the trajectory of the moving object and the experience of novices may be important factors that influence the effect of action anticipation.

Another important finding of the present study was that the left medial frontal cortex exhibited greater positive functional connectivity with the posterior cingulate cortex, fusiform gyrus and inferior parietal lobule in players compared to novices. These brain regions are all associated with action processing in some respect, which further suggests the involvement of the left medial frontal cortex in more skilled action anticipation. For example, the bilateral (Battelli et al., 2003) or left (Buxbaum et al., 2005; Kalénine, 2010) inferior parietal lobule was found to play a role in the recognition of



actions (Battelli et al., 2003; Buxbaum et al., 2005; Kalénine, 2010). In addition, while the right or bilateral fusiform gyrus is often thought to be involved in face processing (Kanwisher et al., 1997; Duchaine and Yovel, 2015), Peelen and Downing (2005) observed that this right fusiform also plays a role in visual processing of the human body. Consistently, previous studies have shown that players were better than novices with respect to action recognition (Starkes, 1987; Abernethy et al., 1994). The posterior cingulate gyrus is thought to play an important cognitive role, and one influential hypothesis posits that the posterior cingulate cortex plays a central role in supporting internally directed cognition (Leech and Sharp, 2014). Small et al. (2003) found that right posterior cingulate cortex, together with the left medial frontal cortex, mediates the anticipatory allocation of spatial attention. Players showed a different attention pattern than non-players at key locations in a sports situation (Williams et al., 1994; Eccles et al., 2006) and paid more attention to an opponent's body kinetic information (Williams et al., 1994; Eccles et al., 2006). Cauda et al.'s (2011) study demonstrated an important role of the bilateral insula in sensorimotor integration. In addition, Gong et al. (2015) found that action video game experts exhibited enhanced functional connectivity and gray matter volume in bilateral insular sub-regions. Similarly, badminton playing may improve players' action

processing abilities, including action recognition and visual identification of key information, as well as action anticipation; these abilities may reflect enhanced functional integration of brain regions involved in action comprehension and action anticipation.

We would like to mention several limitations of our study and suggest outlines for future research. First, the present study investigated the effect of action anticipation only by asking badminton players to predict the path of the shuttlecock; players have more skilled experience in such predictions. However, whether similar effects would be found if the players were asked to predict more general daily action sequences (e.g., inserting a spoon with food into the mouth, Reid and Striano, 2008) remains unclear. Additionally, the present study found a role of the left medial frontal cortex only in professional badminton players; it remains unclear whether this brain region would be activated in normal adults asked to predict the conclusion of common action sequences. In future studies, we may investigate these issues in greater detail. Future studies may also investigate the role of gender and experience in gender identification and action anticipation.

CONCLUSION

In the present study, we observed stronger activation of the left medial frontal cortex in badminton players compared to novices during action anticipation. The left medial frontal cortex also showed stronger functional connectivity with the right cingulate and posterior cingulate cortex, right fusiform gyrus, right inferior parietal lobule and left insula in players compared to novices during action anticipation. Taken together, the findings indicate that the left medial frontal cortex is associated with more skilled action anticipation ability and that there is a specific brain network for action anticipation.

AUTHOR CONTRIBUTIONS

HX was involved in data analysis and manuscript drafting and revises. PW and ZY were involved in data collecting and analysis. XD and GX were involved in imaging data collecting. LM was involved in study design. HL was involved in manuscript drafting and revises. HR was involved in imaging data collecting and manuscript revises. HJ was involved in study design, execution, data analysis, and manuscript revises. We have read and approved the manuscript and agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

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The ERP Effects of Combined Cognitive Training on Intention-Based and Stimulus-Based Actions in Older Chinese Adults

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Age-related decreases in action are caused by neuromuscular weakness and cognitive decline. Although physical interventions have been reported to have beneficial effects on cognitive function in older adults, whether cognitive training improves action-related function remains unclear. In this study, we investigated the effects of combined cognitive training on intention-based and stimulus-based actions in older adults using event-related potentials (ERPs). A total of 26 healthy older adults (16 in the training group and 10 in the control group) participated in the study. The training group received 16 sessions of cognitive training, including eight sessions of executive function training and eight sessions of memory strategy training. Before and after training, both groups of participants underwent cognitive assessments and ERP recordings during both the acquisition and test phases with a motor cognitive paradigm. During the acquisition phase, subjects were asked to press one of two keys, either using a self-selected (intention-based) method or based on the preceding stimulus (stimulus-based). During the test phase, subjects were asked to respond to the pre-cues with either congruent or incongruent tasks. Using ERP indices—including readiness potential, P3 and contingent negative variation to identify motor preparation, stimulus processing and interference effect, respectively—we revealed the effects of training on both intention-based and stimulus-based actions. The correlations were also computed between the improved cognitive performance and the ERP amplitudes. It was shown that the improved executive function might extend substantial benefits to both actions, whereas associative memory may be specifically related to the bidirectional action-effect association of intention-based action, although the training effect of memory was absent during the insufficient training hours. In sum, the present study provided empirical evidence demonstrating that action could benefit from cognitive training. Clinical Trial Registration: www.chictr.org.cn, identifier: ChiCTR-OON-16007793.

Keywords: combined cognitive training, intention-based action, stimulus-based action, executive function, associative memory, ERPs, motor cognition, older adults

INTRODUCTION

Age-related decreases in action are caused by neuromuscular weakness and cognitive decline (Sterr and Dean, 2008; Seidler et al., 2010 for a review). Although the function of action and cognition declines with age, it has been well documented that brain plasticity can remain in older adults (Kelly et al., 2014 for a review; Lampit et al., 2015), which provided encouraging evidence for

effective interventions aiming to improve action and cognitive functions. The relationship between action and cognition has been continuously examined—for example, physical intervention has shown beneficial effects on cognitive function for older adults (Hillman et al., 2002; Colcombe and Kramer, 2003 for a review). However, the question of whether cognitive training improves action-related function remains unclear. In this study, we aimed to explore the electrophysiological effect of combined cognitive training on motor cognition in older Chinese adults.

It has been shown that the age-related decline of executive function is associated with motor deficits among older adults. On one hand, executive function has been reported to be responsible for age-related motor slowing in both the stimulus processing and motor planning stages (Sterr and Dean, 2008; Berchicci et al., 2012; Woods et al., 2015). On the other hand, exercise has been shown to benefit executive function. A meta-analytic study showed that the benefits of physical training in older adults are greatest for executive function (Colcombe and Kramer, 2003 for a review), suggesting that executive function may underpin motor-related cognitive processing in a fundamental and wide-ranging manner.

Recently, with the development and further exploration of ideomotor theory (Greenwald, 1970), it has been recognized that memory also plays an important role in motor cognition. For example, when people intended to achieve a desired goal or action effect, it was necessary to retrieve the related action through associative memory (Elsner et al., 2002). A stream of studies has revealed the related cognitive processing by comparing intention-based action (i.e., ideomotor action) with stimulus-based action (i.e., sensorimotor action) (Elsner and Hommel, 2001; Haggard et al., 2002; Waszak et al., 2005; Keller et al., 2006; Herwig et al., 2007; Herwig, 2015 for a review), which yields a reliable paradigm for the experimental investigation of intention-based action. Although intention-based action is a more complex and evolved action that plays an important role in everyday life (Herwig, 2015 for a review), stimulus-based action, which follows the simple stimulus-response rule, involves tasks that have been commonly used in past studies—such as the reaction task.

In previous behavioral studies comparing intention-based and stimulus-based action modes (Elsner and Hommel, 2001; Haggard et al., 2002; Herwig et al., 2007), participants were asked to perform tasks using an acquisition-test paradigm, which is similar to the study-test paradigm that is typically used in memory studies. In Herwig et al. (2007), during the acquisition phase, the participants in the intention-based action group were asked to make a self-selected keypress (the action) that was always followed by a certain tone with either a high or low pitch (the effect of the action), thus creating an *action-effect binding*. For the stimulus-based action group, participants were asked to make certain responses by pressing a key based on a specific rule. During the subsequent test phase, the participants were asked to make speeded keypresses in response to tones with high or low pitches based on either the congruent or incongruent rule that was learned during the acquisition phase. The results showed an interference effect of reaction time only in the intention-based action group—that is, faster performance occurred in congruent tasks than in incongruent tasks. This result was interpreted

as showing that the *action-effect binding* that was acquired in the acquisition phase was bidirectional and then activated in reverse (i.e., *effect-action retrieval*) during the test phase, which postponed the reaction time when the incongruent rule was applied. However, for stimulus-based action, the interference effect of the reaction time was missing because it was based on simpler stimulus-response linkages that are unidirectional (Herwig et al., 2007). Notably, these processes of *action-effect binding* and *effect-action retrieval* resembled the processes of associative memory coding and retrieval, respectively. The involvement of associative memory in intention-based action was further improved by neuroimaging studies that revealed activations in both the supplementary motor area (SMA) and the medial temporal memory system, including hippocampus and parahippocampal gyrus, during either action image or execution (Elsner et al., 2002; Melcher et al., 2008, 2013; Pfister et al., 2014).

The different processing modes between the two types of actions have also been revealed by event-related potentials (ERPs) (Waszak et al., 2005; Keller et al., 2006). The results showed that during the acquisition phase, the readiness potential (RP) (e.g., Shibasaki and Hallett, 2006) component, which is a slow, negative going potential prior to an action execution that reflects the general preparation of voluntary movement, was more negative under intention-based conditions, but the P3 component—a positive potential observed approximately 300 ms after the stimulus presentation that reflects the formation of a link between stimulus evaluation and response selection (e.g., Petruo et al., 2016)—was more positive under stimulus-based conditions. However, the ERP mode was not included during the test phase. The interference effect of the reaction time between congruent and incongruent tasks that was discussed above was observed during the test phase, revealing the *effect-action retrieval* process involved in intention-based action. Therefore, the ERP mode must be investigated during the test phase in this study with the expectation of the interference effect of ERPs. According to previous studies, the contingent negative variation (CNV) component, which is thought to reflect action preparation that is identical to the RP component (Leuthold et al., 2004), tends to increase with the amount of advanced information provided by the precue, either because this information enables greater sufficient preparation for the following action (Leuthold et al., 2004) or because the subjects can access additional resources to complete the task (Falkenstein et al., 2003). In this study, we used a precuing task during the test phase to investigate the interference effect of CNV amplitude. We assumed that for intention-based actions, the CNV amplitude increases during congruent tasks because the congruent response rule facilitated action preparation more than the CNV amplitude during incongruent tasks because the incongruent response rule interfered with action preparation. This difference in CNV amplitude between congruent and incongruent tasks might be observed with intention-based action but not with stimulus-based action.

Based on these points, we can see that executive function and associative memory are involved in the cognitive processing of action. A growing number of studies have examined the training benefits of executive function (Karbach and Verhaeghen, 2014

for a review; Kelly et al., 2014 for a review) and associative memory (Derwinger et al., 2003; Bottiroli and Cavallini, 2009; Gross et al., 2012 for a review). It has also been reported that cognitive training, combined with multiple components, produced a broader effect on multiple cognitive domains than single cognitive training (Cheng et al., 2012; Walton et al., 2014). Therefore, we aimed to use combined cognitive training—including both executive function and associative memory—in this study of older Chinese adults to improve their action functions, which were measured by the related cognitive processes under specific action modes. The action modes of intention-based and stimulus-based actions were measured electrophysiologically separately with the pre- and post-tests of neuropsychological tests, respectively. We assumed that combined cognitive training would benefit both actions in terms of cognitive processing, which is reflected by the training effects observed in the amplitudes of the related ERP indices. Enlarged amplitudes after training are expected in the RP and P3 components under intention-based and stimulus-based action modes, respectively, suggesting beneficial gain in either the action-preparation or stimulus-processing components. For the CNV component, the interference effect—the amplitude difference between congruent and incongruent tasks—is also expected in intention-based actions after training, suggesting an improvement in action-effect association. Additionally, the training effect correlation between the ERP amplitudes and cognitive performance will be analyzed for further investigation. We predicted that after training, the improved executive function may benefit both intention-based and stimulus-based actions because of the fundamental and extensive role of executive function that may occur during action preparation. However, improved associative memory was predicted to benefit intention-based action only based on its specific role in action-effect association. In summary, we hypothesized that (1) combined cognitive training would benefit both intention-based and stimulus-based actions in their ERP modes and (2) the increased performance in executive function may be responsible for the training benefits of both actions, whereas the increased performance in associative memory may benefit only intention-based action.

MATERIALS AND METHODS

Participants

The participants were a subset of the subjects enrolled in a larger study that was conducted by the Institute of Psychology, Chinese Academy of Sciences called Cognitive Training for Healthy Older Adults: Combined Training versus Memory Training¹. In the larger study, healthy older adults were enrolled from the local community through advertisements. Forty subjects were randomly divided into memory training group and combined cognitive training group, with 20 subjects included in each training group. Eighteen subjects were recruited later from the local community for the control group. All subjects were

(1) right-handed, (2) scored ≥ 24 on the Mini Mental State Examination (MMSE, Folstein et al., 1975), (3) scored ≤ 16 on the Center for Epidemiologic Studies Depression Scale (CES-D, Roberts and Vernon, 1983), (4) had normal or corrected-to-normal vision and hearing, (5) had no history of severe psychiatric or neurological disease, and (6) did not use drugs that might have adversely affected cognition (i.e., benzodiazepines or antipsychotics). In this study, 34 subjects in the combined training group and control group volunteered to participate in the ERP study. Of those who completed the baseline assessments, six subjects dropped out of the study, and two subjects were rejected due to an ERP data collapse caused by electrode failure. In the present ERP study, there were 16 final subjects ($N = 16$) in the combined training group and ten final subjects ($N = 10$) in the control group.

All the subjects participated in both the acquisition and test phases. During the test phases, the subjects in each age group were divided into two subgroups that performed congruent or incongruent tasks. The order of the intention-/stimulus-based acquisition and the congruent/incongruent tasks was counterbalanced for each group. For the training group, eight subjects performed congruent tasks and eight subjects performed incongruent tasks. For the control group, six subjects performed congruent tasks and four subjects performed incongruent tasks.

As shown in **Table 1**, the subjects in both groups did not differ significantly ($p > 0.05$) in terms of age, gender, education, global cognition (MMSE) or depression (CES-D). All the subjects were given written informed consent and were financially reimbursed for their participation. The study was approved by the Ethics Committee of the institute of Psychology, Chinese Academy of Sciences.

Procedures and Tasks

Both groups of subjects received cognitive assessments and ERP recordings of two action modes before and after training. The cognitive assessments and ERP recordings were conducted in two separate sessions that lasted approximately 100 and 150 min, respectively. The pre-training assessments and recordings were conducted within 2 weeks before training, and the post-training assessments and recordings were conducted within 1 week after training. The combined cognitive training group received 16 training sessions over the course of approximately 6 weeks, including eight initial sessions of executive function training and eight sessions of memory strategy training thereafter, in which it was supposed that enhanced executive

TABLE 1 | Demographic and cognitive characteristics.

	Training ($n = 16$) M (SD)	Control ($n = 10$) M (SD)	<i>P</i> -value
Age (years)	69.6 (4.6)	69.0 (3.4)	0.74
Education (years)	12.4 (3.5)	13.1 (3.0)	0.62
Female/Male	9/7	6/4	0.85
MMSE	27.7 (1.5)	28.3 (1.5)	0.32
CES-D	4.6 (4.3)	3.7 (4.5)	0.60

¹<http://www.chictr.org.cn/showproj.aspx?proj=13140>

function may facilitate the effective utilization of memory strategy. Each session lasted approximately 60 min (50 min of training with a break of 10 min). The subjects in the training group were asked to come to the institute three times per week and received 16 h of training in total.

Combined Cognitive Training

The combined cognitive training and the outcome measures used in this study were part of another interventional study (Li et al., 2016) with fewer subjects who participated in the ERP study of the two action modes. The more detailed content of combined cognitive training and outcome measures were presented in supplements to this article because the main focus of this study was the ERP modes of the training effect on actions.

Executive function training

Updating training. The updating training was adapted from the keep-track task (Yntema and Mueser, 1962) that included word-updating and picture-updating tasks from identical training sessions. The subjects were instructed to continuously update the items (i.e., either words or pictures) of targeted categories (e.g., animals, clothes, vegetables, and fruits) and verbally report the last item of each category at the end of the trial. Task difficulty was self-adaptive based on each subject, and the difficulty was manipulated by varying the number of categories presented (2 or 3) and the number of items in each category (2, 3, or 4).

Switching training. The switching training was adapted from the task-switching paradigm (Kray and Lindenberger, 2000), including only mixed-task blocks (two-task and three-task switching). The subjects were instructed to input responses by pressing one of two keys based on the cues presented at the bottom of the screen indicating the switched response rule. Task difficulty was also self-adaptive to each subject by varying the number of subtasks (two and three) and the number of items in each subtask (8, 10, and 12).

Memory strategy training

Method of loci. The method of loci (Bower, 1970) was used to recall a list of names of common objects (such as fruits and animals) with a mental image. The subjects were required to imagine a familiar route with several landmarks and the words associated with these landmarks using a mental map. The subjects were then asked to retrieve the words by revisiting the landmarks in their brain. The size of the wordlist was initially set at 8 and gradually increased to 10, 12, 14, and 16 with the training.

Face-name mnemonic. The face-name mnemonic (Yesavage et al., 1983) was taught to subjects to build an association between prominent facial features and name. In the recall phase, the subjects were required to retrieve the related mental image of the learned association and then recall the name. The number of practiced face-name mnemonics began from 1 face and increased to 7 faces progressively.

Outcome measures

Both the trained and untrained tasks were measured in the pre- and post- assessments. The detailed description of outcome measures was included in the Supplements Materials.

The trained executive function tasks included word- and picture-updating tasks and the switching task. The untrained executive function included the Trail Making Test (B-A) (TMT (B-A)), the difference value between the reaction time of TMT-B and TMT-A (Reitan, 1955), the Stroop Test (Stroop, 1935) and the Backward Digit Span Task (from the Wechsler Adult Intelligence Scale-Revised in China; Gong, 1992).

Trained memory tasks included the wordlist task and face-name task. The untrained memory task included the Associative Learning Test (ALT, from the Clinical Memory Scale, Xu and Wu, 1986) and the Logical Memory Test (LMT, from the Wechsler Memory Scale-Revised in China; Gong, 1989).

ERP Recordings

Apparatus and Stimuli

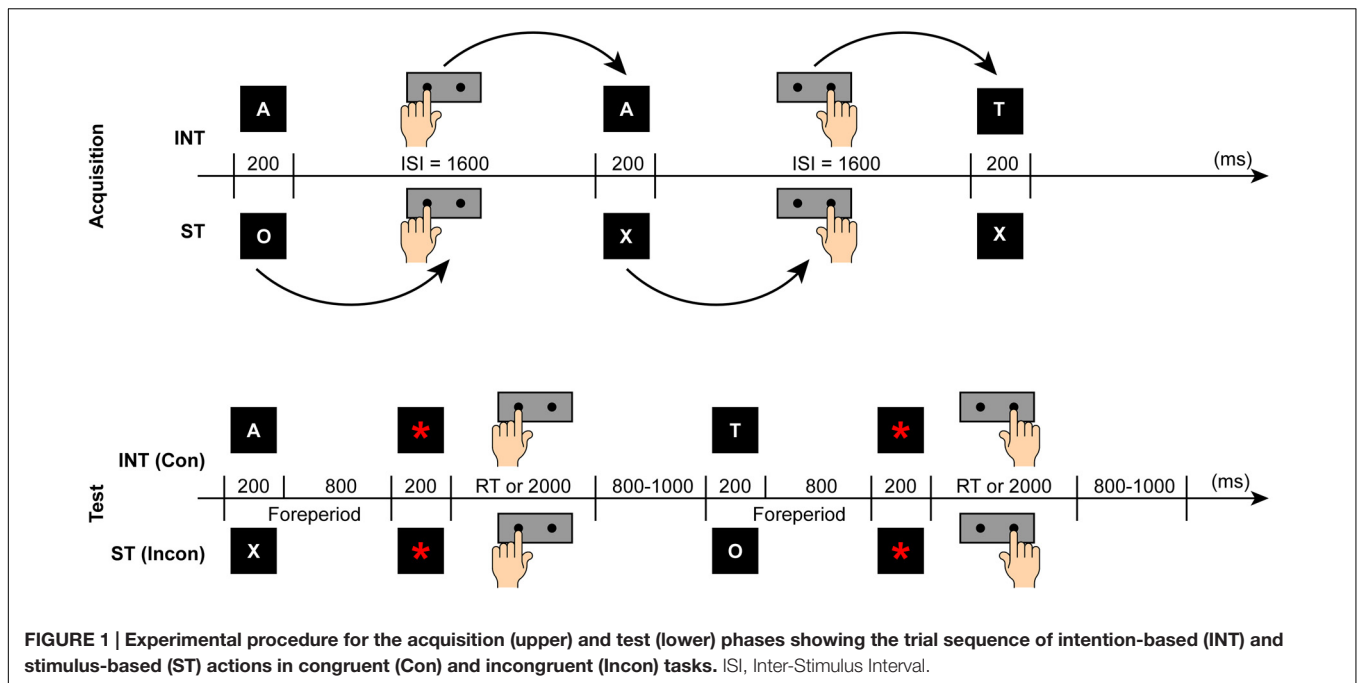
The subjects sat in front of a computer screen on a table that was 75 cm away from the subjects' eyes. A keyboard with two response keys separated by a horizontal distance of 45 mm was placed on the table in front of the screen. A white cross ("+") depicted on a black background at a visual angle of $0.8^\circ \times 0.8^\circ$ served as the eye fixation site. Possible visual stimuli included the capitalized letters A, T, O, and X presented in white (height: 1.7°) against a black background at the center of the computer screen. During the test sessions, a centrally presented red asterisk ("*") with a visual angle of $0.8^\circ \times 0.8^\circ$ was used as the imperative signal for a key press response. An auditory pacing signal consisting of sine tones (600 Hz; 100 ms in duration) was presented at the beginning of each block (see below) at a comfortable volume through mini-sound box sets placed on either side of the computer screen. E-prime (Version 2.0) software for Windows XP was used to present the visual stimuli and auditory signals to the subjects and to collect the behavioral data.

Tasks

Each subject completed four experimental sessions, including the acquisition and test phases, for both the intention-based and stimulus-based actions. As shown in **Figure 1**, all the subjects performed intention-based and stimulus-based acquisition during the acquisition phase. In other words, they pressed the left or right key either in a self-selected way or based on a specific rule. Following the acquisition phase, the subjects entered a test phase in which they were required to respond to the stimulus presented on the screen. During the test phase, half of the subjects performed a congruent task, and the remaining half performed an incongruent task. The order of the intention- and stimulus-based acquisition for the congruent and incongruent tasks was counterbalanced across each group.

Acquisition phase

A temporal bisection task adapted from Keller et al. (2006) was used during the acquisition phase. First, the subjects were presented with instructions and performed practice trials of the timed bisection *per se*, which was a pure timing task. This task required them to randomly press one of the two response



keys to bisect a 1,600 ms time duration (i.e., the inter-stimulus interval, ISI) during which a fixation cross (“+”) was presented at the center of the screen, which was initiated by a centrally placed number eight (“8”) that appeared on the screen for 200 ms. Each practice block contained 20 trials, after which the subjects were apprised of their time performance to inform them of how frequently their responses fell within 350 ms of the exact bisection time point. The practice session ended when the subjects indicated that they were ready for the formal experiment and when their response accuracy rates were above the 80% criterion.

Electroencephalogram (EEG) electrodes were then fixed to the subject’s scalp. The subjects were then instructed concerning the performance of each type of action via both the visually presented instructions on the computer screen and a verbal explanation by an instructor. After a brief practice period, the subjects proceeded with the formal experiment. EEG recordings were collected during both intention-based and stimulus-based action sessions. As shown in **Figure 1**, in the intention-based acquisition session, the subjects were required to press the left or right key to bisect the 1,600 ms ISI. Each key press produced an “action effect” by determining the subsequent letter (i.e., A, T, O, or X) that appeared for 200 ms at the end of each ISI (e.g., the selection of the left key determined the appearance of A, and the selection of the right key determined the appearance of T). The subjects were instructed to attempt to implement a random sequence of letters, similar to tossing a coin, rather than to produce a sequence in a certain order. During the stimulus-based acquisition session, the subjects were required to press the left or right key at the bisection point of the ISI based on the rules associated with the appearance of a specific letter (A, T, O, or X; e.g., the appearance of O signaled a left key press, and the appearance of X signaled a right key press); these letters were

presented in a randomized and counterbalanced sequence. To eliminate carry-over effects, different letters were presented for the same subject across the two actions.

Each block contained 40 trials, and the subjects were informed that they could take a break between blocks. For the first 10 trials of each block, an auditory pacing signal indicating the true ISI midpoint to assist the subjects with their timing performance. The subjects were required to synchronize their key presses with pacing signals and were told to try to maintain this rhythm when the auditory pacing signal ceased for the remaining 30 responses. As with the practice trials, in the experimental trials, the responses that were considered correct were within 350 ms (± 350 ms) of the true bisection point. To ensure that a sufficient number of trials fit the ERP average under each condition, the blocks were repeated until 200 trials with correct responses (excluding auditory signaled trials) were performed.

Test phase

Upon completion of the acquisition phase, the participants proceeded to the test phase. Two sessions of the movement precueing task that were congruent or incongruent with the intention-based and stimulus-based acquisitions were administered. After the fixation cross (“+”) was presented for 200 ms, a letter serving as the precue (A, T, O, or X) appeared at the center of the screen for the next 200 ms to indicate whether the subsequent key pressed should be the left or right key. Additionally, the subjects were instructed not to respond directly to the precueing letter presented on the screen to ensure sufficient time for motor preparation before the motor response (Falkenstein et al., 2003; Leuthold et al., 2004). Although they already knew which key to press upon the appearance of the letter, they were instructed to withhold their responses until a red asterisk (“*”) was presented (see **Figure 1**). This cue served

as an imperative response signal and appeared at the center of the screen during the response period. Any responses occurring beyond this period were classified as incorrect. The response period ended with either a key press or ended after 2000 ms and was followed by a black screen for a random duration of between 800 and 1000 ms. Each test session consisted of 200 trials. The experiment ended after the two test sessions were completed.

During the test phase, the subjects were subdivided into two groups: one group performing congruent tasks and the other performing incongruent tasks. In the congruent task, the subjects were required to press a key based on the rules that they had learned during the acquisition phase. For instance, the subjects who had acquired left key press→A/right key press→T associations were then required to respond to A with a left key press and to T with a right key press. In the incongruent task, the subjects were required to perform the opposite key presses. For instance, the subjects who had acquired O→left key press/X→right key press associations were then required to respond to O with a right key press and to X with a left key press. The congruent and incongruent tasks described above are shown in **Figure 1**.

Analysis

Analysis of the Cognitive Assessment Score

The composite scores were calculated separately for the executive function and memory measures to index the overall performance of these two cognitive functions. The raw scores for the five executive function tasks (word and picture updating tasks, TMT (B-A), Stroop Test and backward digit span) and four memory tasks (wordlist task, face-name task, ALT and LMT) were standardized into z scores and then summed separately for the two cognitive functions and converted into a composite t score ($M = 50$, $SD = 10$).

Before the performance of one-way analysis of variance (ANOVA), the training and control groups' baseline performances on the cognitive assessments were compared. Significant differences ($p < 0.05$) were found for the ALT, face-name task, memory composite t -score and backward digit span. Therefore, to control the baseline performance between the two groups, a covariance analysis (ANCOVA) was conducted on these four tasks. The posttest-minus-pretest change scores were compared between the two groups with the pretest scores on the above four tasks included as covariates. The ANCOVA was performed for each outcome measure and composite t -score. The switching task scores of the pretest from eight subjects in the control group were lost due to technical issues. Therefore, the switching task was not included in the analysis of this study.

ERP Recording and Data Analysis

The EEG data were recorded continuously from 64 cap-mounted Ag/AgCl electrodes (Neuroscan Inc.) arranged according to the International 10–20 System. The left mastoid served as an online reference, and the average of both mastoids was calculated offline to enable algebraic re-referencing. The

EEG data were amplified with a band-pass filter of 0.05–100 Hz and were digitized at 500 Hz. The vertical and horizontal electrooculograms (VEOG and HEOG, respectively) were recorded from two pairs of electrodes, one pair placed approximately 1 cm above and below the left eye and the other pair placed approximately 1 cm lateral to the outer canthi of both eyes. Inter-electrode impedances were maintained below 5 k Ω .

The EEG data were processed offline, and the ocular artifacts were removed using a regression procedure (Semlitsch et al., 1986). The data were low-pass filtered using a cutoff frequency of 30 Hz. Epochs of RP were time-locked to the onset of a motor response and were segmented from a –900 ms pre-response to a 400 ms post-response, with –900 ms to –800 ms serving as the baseline period. Epochs of 700 ms (including the pre-stimulus baseline time of 100 ms) were extracted for P3, and 2,200 ms segments (including the pre-stimulus baseline time of 200 ms) were extracted for CNV; both were time-locked to the onset of the stimulus. Epochs exceeding $\pm 100 \mu\text{V}$ were considered artifacts and excluded from further analysis.

For the acquisition phase, RP was measured as the mean amplitude at electrodes F3, Fz, F4, 384 C3, Cz, C4, P3, Pz, and P4 within a window of –400 to 0 ms because these signals reflect general motor preparation prior to action (Shibasaki and Hallett, 2006). The mean amplitude of P3 was measured within 400–500 ms over the centro-parietal sites (C3, Cz, C4, CP3, CPz, CP4, P3, Pz, and P4). Finally, for the test phase, the mean CNV amplitude was measured within 500–1,000 ms over the centro-parietal sites. The data were analyzed via a repeated-measures ANOVA considering time (pretest and posttest), action (intention-based and stimulus-based), anterior–posterior scalp location (anterior, medial, and posterior) and scalp laterality (left, middle, and right) as within-subject factors and group (training and control) as the between-subject factor. An analysis of the congruent and incongruent task subgroups was conducted exclusively for the CNV within the two groups while considering the task (congruent and incongruent) as a between-subject factor. Trials that were classified as incorrect were excluded from the ERP analysis. The Greenhouse-Geisser correction was used to adjust for sphericity violations. A *post hoc* analysis for significant main effects was performed using the Bonferroni method when necessary. Significant interactions were analyzed using simple effects models.

Correlation analyses between cognitive assessments and ERP mean amplitudes were also conducted when an ERP training effect was found. The change scores of the posttrain-minus-pretrain were calculated for cognitive assessments, except for the Stroop task and TMT (B-A). In these two tasks, the change scores of the pretrain-minus-posttrain were calculated because the lower scores indicated better performance. In calculating the ERP amplitude changes, the posttrain-minus-pretrain was used for positive ERP components, and the pretrain-minus-posttrain was used for negative ERP components, so that larger values indexed more enlarged amplitudes after training. Spearman's rho was computed as two-tailed.

RESULTS

Training Effects on Cognitive Assessments

Table 2 shows the baseline performance, the means of change scores, significances and effect sizes (partial eta squared). The ANCOVA results showed significant training effects ($p < 0.05$) on the measure of the executive function composite score, word-updating and picture-updating tasks.

Training Effects on ERPs of Action Modes

The values of ERP indices under each condition was showed in Table 3.

Readiness Potential (−400–0 ms)

Figure 2 presents the RP measurements of the pretest and posttest for both groups. There was a significant main effect of action [$F_{(1,24)} = 40.325, p < 0.001, \eta_p^2 = 0.627$], indicating that the RP amplitude for intention-based action was significantly more negative than that of the stimulus-based action. The interactions between time, action and group were also significant [$F_{(1,24)} = 4.67, p = 0.041, \eta_p^2 = 0.163$]. A simple effects analysis revealed that after training, the RP amplitude of stimulus-based action was significantly more negative ($p = 0.005$) than that of the baseline measure in the training group, but this measure was not significant in the control group ($p = 0.387$).

P3 (400–500 ms)

The P3 measurements of the pretest and posttest are shown in Figure 3 for both the training and control groups. There was a significant main effect of action [$F_{(1,24)} = 11.487, p = 0.002, \eta_p^2 = 0.324$], indicating that the P3 amplitude

for stimulus-based action was significantly more positive than that for the intention-based action. No interaction was found to be significant between time, action and group.

Contingent Negative Variation (CNV) (500–1,000 ms)

Figure 4 shows the CNV measurements of the pretest and posttest for both groups, in which a significant interaction between time and action was found [$F_{(1,22)} = 14.156, p = 0.001, \eta_p^2 = 0.392$]. A simple effects analysis revealed that after training, the CNV amplitude of intention-based action was significantly more negative ($p = 0.017$) than that of the baseline measure for both groups. However, such an effect was not found for the stimulus-based action ($p = 0.757$). This training effect was further analyzed within each group. The significant interaction between time and action was found in only the training group with a larger effect size [$F_{(1,14)} = 17.155, p = 0.001, \eta_p^2 = 0.551$] and was not found in the control group.

According to our assumption, the interference effect of CNV, which was reflected by the amplitude difference between congruent and incongruent tasks, was expected for intention-based action rather than stimulus-based action after training. Therefore, the pattern of CNV amplitudes between the two tasks and action modes was investigated within each group during each measure time. For the pretest, no significance was found for the main effect or the interaction of the action and task for either group. For the posttest, there was a significant action main effect [$F_{(1,14)} = 25.98, p < 0.001, \eta_p^2 = 0.65$] in the training group, indicating that the CNV amplitude of intention-based action was more negative than that of stimulus-based action. A significant interaction was also found between action and task [$F_{(1,14)} = 5.905, p = 0.029, \eta_p^2 = 0.297$]. A simple effects analysis showed that, in contrast with our

TABLE 2 | Training effects on cognitive assessments of two groups.

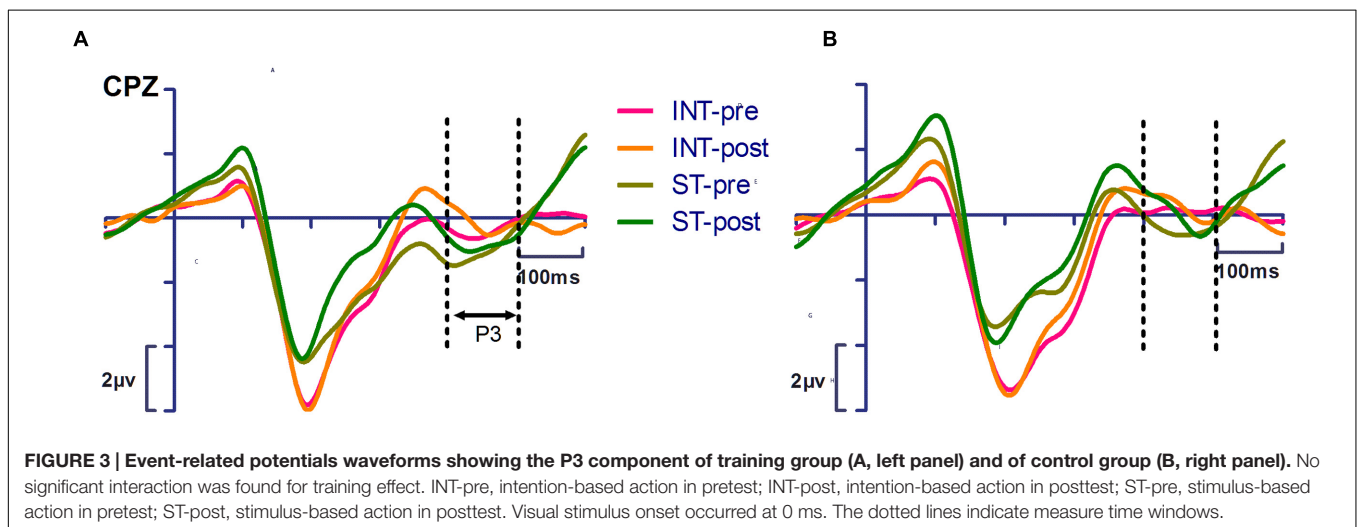
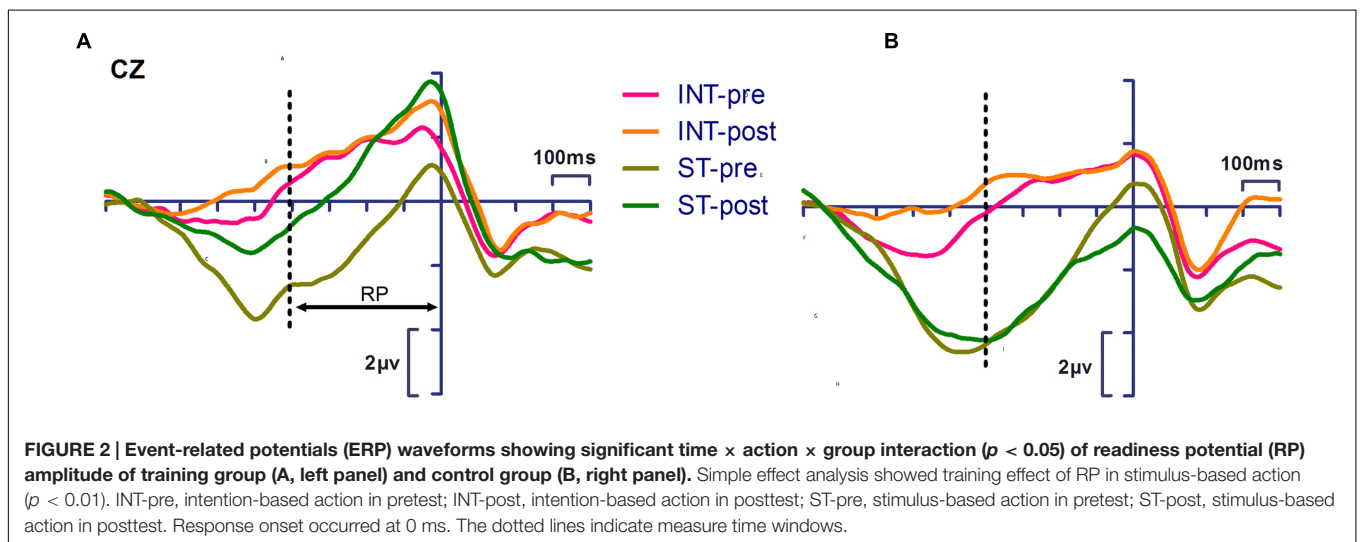
	Training (n = 16)		Control (n = 10)		P ^b	Partial eta squared
	Baseline mean (SD)	Change mean (95% CI)	Baseline mean (SD)	Change mean (95% CI)		
Executive function						
EF (composite t score)	48.6 (27.5)	10.37 (−4.60–25.35)	52.2 (23.0)	−16.20 (−35.14–2.74)	0.032	0.18
Stroop test ^a	11.6 (13.7)	−1.19 (−7.40–5.02)	16.6 (8.5)	−3.92 (−11.78–3.94)	0.579	0.01
TMT (B-A) ^a	32.0 (18.0)	−8.63 (−15.99–−1.26)	26.9 (13.1)	0.09 (−9.22–9.42)	0.143	0.09
Digital span backward	4.3 (1.3)	0.38 (−0.19–0.95)	5.6 (1.4)	0.50 (−0.25–1.24)	0.807	0.00
Word updating	3.5 (2.3)	3.75 (2.71–4.79)	5.5 (2.7)	1.00 (−0.31–2.31)	0.002	0.90
Picture updating	6.0 (2.5)	2.94 (2.21–3.66)	6.0 (1.9)	1.60 (0.68–2.52)	0.027	0.19
Memory						
Memory (composite t score)	41.1 (30.5)	4.48 (−6.84–15.80)	64.3 (15.9)	−7.16 (−21.80–7.47)	0.224	0.06
ALT	10.2 (2.3)	2.36 (0.65–4.06)	12.9 (2.5)	1.93 (−0.30–4.16)	0.770	0.00
Face-name	2.4 (1.9)	−0.11 (−0.84–0.62)	4.1 (2.0)	−0.18 (−1.12–0.76)	0.904	0.00
LMT	7.4 (2.6)	1.69 (0.89–2.49)	8.5 (2.1)	0.70 (−0.32–1.72)	0.128	0.09
Wordlist	8.9 (2.4)	1.50 (−0.48–3.48)	9.1 (1.7)	−0.20 (−2.70–2.30)	0.281	0.05

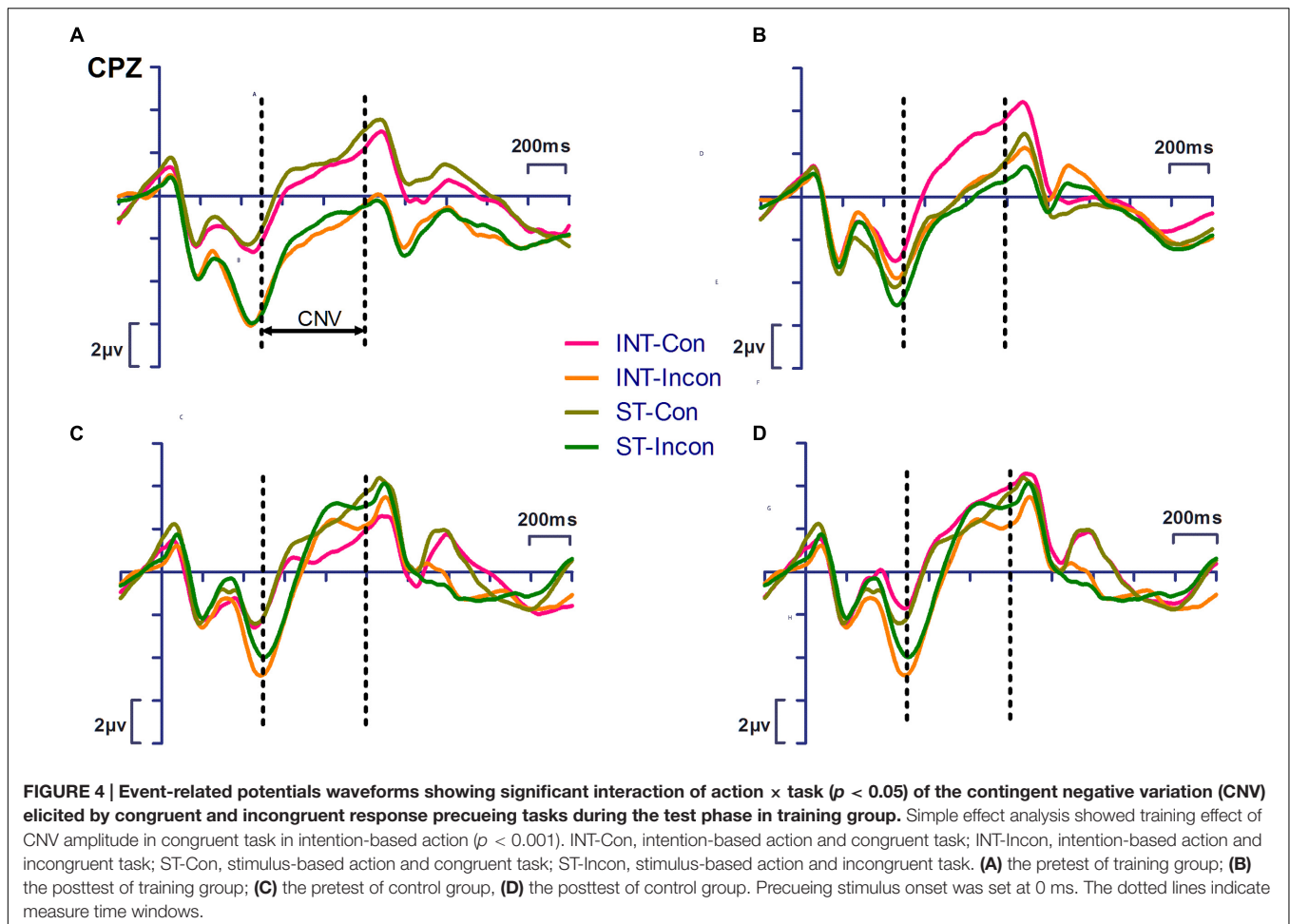
SD, standard deviation; CI, confidence interval; ALT, Associative Learning Test; LMT, Logical Memory Test; EF, executive function; TMT, Trail Making Test. ^aLower scores indicate better performance. ^bAnalyses of covariance of posttraining-minus-baseline-change mean scores, controlled for baseline performance of backward digit span composite score of memory, ALT and face-name.

TABLE 3 | The means and 95% confidence interval (μV) of RP, P3, and CNV amplitudes in all conditions.

	Training group ($n = 16$)		Control group ($n = 10$)	
	Pretest mean (95%CI)	Posttest mean (95%CI)	Pretest mean (95%CI)	Posttest mean (95%CI)
RP				
INT	-0.57 (-1.42-0.27)	-0.44 (-1.52-0.64)	0.52 (0.72-1.76)	0.33 (-1.68-2.34)
ST	2.04 (0.96-3.11)	0.42 (-0.97-1.81)	2.65 (1.40-3.90)	3.33 (1.68-4.99)
P3				
INT	0.40 (0.09-0.71)	0.36 (-0.11-0.83)	-0.08 (-0.40-0.24)	-0.13 (-0.50-0.25)
ST	0.93 (0.54-1.32)	0.80 (0.24-1.36)	0.53 (-0.09-1.14)	0.20 (-0.61-1.02)
CNV				
INT-Con	-0.44 (-3.08-2.19)	-0.82 (-3.17-1.53)	-0.60 (-2.06-0.86)	-1.46 (-3.14-0.21)
INT-Incon	2.55 (-0.49-5.58)	0.42 (-1.39-2.22)	1.44 (-0.17-3.06)	-0.08 (-1.63-1.47)
ST-Con	-1.03 (-3.98-1.91)	0.53 (-1.92-2.99)	-0.92 (-2.35-0.51)	-0.74 (-1.91-0.43)
ST-Incon	1.75 (-0.86-3.86)	0.90 (-0.88-2.67)	0.08 (-1.07-1.24)	-0.66 (-1.96-0.64)

CI, Confidence Interval; RP, readiness potential; CNV, contingent negative variation; INT, intention-based condition; ST, stimulus-based condition; -Con, congruent task; -Incon, incongruent task.





assumption, the CNV amplitude of intention-based action was significantly more negative than that of the stimulus-based action ($p < 0.001$) in the congruent task but not in the incongruent task ($p = 0.08$). No significance was found for the main effect or the interaction of action and task in the control group.

Correlation Results

As revealed by the ANOVA analysis, two training effects reflected by the increased amplitudes of RP in the stimulus-based action and CNV of the intention-based action were found in the combined cognitive training group. Marginal significance was found for the correlation between the change scores of the picture-updating task and the changed RP amplitude of stimulus-based action ($r_s = 0.482$, $p = 0.059$), showing that improved performance of executive function after training was positively related with the increased RP amplitude in stimulus-based action. Significant correlations were also found between the change scores of the composite executive function ($r_s = 0.738$, $p = 0.037$), the Stroop task ($r_s = 0.719$, $p = 0.045$) and the TMT (B-A) ($r_s = 0.857$, $p = 0.007$) and the changed CNV amplitude of intention-based action in the congruent task, showing that improved performances of executive function after training

were positively related with the increased CNV amplitude in congruent task in intention-based action during test phase. The change scores of the Stroop task ($r_s = -0.778$, $p = 0.023$) were significantly correlated with the changed CNV amplitude of intention-based action in the incongruent tasks, showing that improved performances of executive function after training were negatively related with the decrease CNV amplitude in incongruent task in intention-based action.

DISCUSSION

The present study investigated the training effects of combined cognitive training on intention-based and stimulus-based actions using ERP indices, including RP, P3, and CNV components. The training effects showed significant improvements for executive function performance but not for memory performance, consistent with Li et al.'s (2016) study. In comparison with the baseline performance, enlarged amplitudes of RP components in stimulus-based action and in the CNV component in intention-based action were revealed after training. Correlation analysis indicated that the ERP training effects observed in this study were related to improved executive function performance.

Based on our assumptions, the combined cognitive function training used in this study benefits both executive function and memory. However, the results showed that only executive function but not memory benefited from the training. The possible reasons for this finding have been discussed in Li et al.'s (2016) article, addressing insufficient training hours for both cognitive functions. In Li et al.'s (2016) study, a comparable memory training group receiving 16 sessions of memory training in total demonstrated a significant training effect of memory, whereas the combined cognitive training group demonstrated a significant training effect for the executive function and marginal significance for memory. In this study, the same combined cognitive training group and control group were involved, with fewer participants completing the ERP tasks before and after training, revealing a significant training effect on executive function but not on memory.

The ERP results showed a significant main effect of action for both RP and P3 amplitudes during the acquisition phase. This result replicated the findings of previous studies with regard to the modal differences between the two types of actions and has been discussed in depth (Waszak et al., 2005; Keller et al., 2006). In other words, a higher RP amplitude was observed for intention-based action than for stimulus-based action, reflecting that the process of motor preparation was more prominent for the intention-based condition. A larger P3 amplitude was observed for stimulus-based actions than for intention-based actions, indicating that the process of stimulus evaluation and response selection was more prominent for the stimulus-based condition. This mode difference between two actions was consistently presented in this study for both groups before and after training, suggesting that the two action modes were acquired under experimental manipulation.

The training effects revealed by ERPs were investigated separately during the acquisition and test phases. Consistent with our assumption, during the acquisition phase, it was revealed that the RP amplitude of the stimulus-based action was enhanced after training in comparison to that of the baseline. Thus, the stimulus-based action was improved after cognitive training, which may be related to enhanced executive function. In previous studies, the relationship between executive function and motor preparation has been reported in older adults and the related brain activations in prefrontal cortex has been discussed (Sterr and Dean, 2008; Berchicci et al., 2012). Moreover, tasks indexing stimulus-based action such as reaction time task were mostly used in previous laboratory studies. Therefore, our result was consistent with previous studies. The training effect was not observed for the RP amplitude of intention-based action and the P3 amplitude of both actions. These results demonstrate that executive function training benefited motor preparation rather than stimulus processing. A previous ERP study (Berchicci et al., 2012) has revealed that the age-related decline in action was more pronounced for motor preparation than for stimulus processing. The over-recruitment of the prefrontal cortex (PFC) was observed during the motor preparation for both simple and complex tasks in older adults compared to young adults. Therefore, it might be that the executive function training in this study enhanced the activities of the PFC in older adults,

which improved the motor preparation of the stimulus-based action. The relationship between executive function and motor preparation has been demonstrated in previous studies (Sterr and Dean, 2008; Berchicci et al., 2012; Woods et al., 2015). However, it is notable that the motor preparation of intention-based action in this study did not benefit from the cognitive training. One possible explanation might be that the intention-based action was more complex and that older adults favor simpler stimulus-based action to show the age-related decline. The other explanation, according to our assumption, was that associative memory was responsible for the action-effect binding of intention-based action during the acquisition phase. The absence of the memory training effect in this study may account for the lack of improvement of intention-based actions.

During the test phase, based on the results and grand average of the CNV component, the training effect was mainly demonstrated as an enlarged amplitude of intention-based action in congruent tasks, which correlated with the changed scores of executive function after training and suggested that improved executive function was responsible for improved intention-based action after the combined cognitive training. It has been demonstrated in previous studies that the executive function was responsible for motor preparation (Sterr and Dean, 2008; Berchicci et al., 2012) and gained the greatest cognitive benefits from physical training in older adults (Colcombe and Kramer, 2003 for a review). Therefore, executive function may play a fundamental and extensive role in motor preparation. In the present study, we observed that improved executive functions after cognitive training were responsible for improvements in both intention-based action, which was indexed by an enlarged CNV amplitude, and stimulus-based action, which was indexed by an enlarged RP amplitude. Thus, consistent with our assumption, the improved executive functions benefits both intention-based and stimulus-based actions and provides evidence for the feasibility that action might gain benefits from cognitive training.

Nevertheless, the training effect of CNV amplitude was inconsistent with our assumption. The interference between congruent and incongruent tasks reflected by the CNV amplitude was not observed as we assumed—in other words, for intention-based action, the CNV amplitude in congruent tasks enlarges with the facilitation effect whereas the CNV amplitude in incongruent tasks decreases with the interference effect. In another study focused on the age effects on two actions (Niu et al., submitted) in which a young age group was involved, the interference effect of CNV amplitude replicating the RT interference between congruent and incongruent tasks was found only in intention-based action for young adults but not for older adults. Therefore, the absence of interference effects of CNV indicated the deficit of intention-based action in older adults, which was probably related to memory deficit in older adults.

According to the dual-process theory of memory, memory judgment is controlled either by the retrieval of specific details (i.e., recollection) or by the general strength (i.e., familiarity) of information (Rhodes et al., 2008). As suggested by the recall-to-reject theory (Rotello and Heit, 2000; Gallo et al., 2004), for the ability to reject the rearranged pairs of old items from the

learned intact pairs, recollection was essentially required. For older adults, the recollection deficit is more sensitive to aging, and familiarity may become more favorable and reliable for associative memory retrieval (Naveh-Benjamin, 2000; Rhodes et al., 2008). As a result, older adults tend to endorse the rearranged pairs as intact pairs with high levels of false alarm observed, particularly when the items were repeatedly learned (Rhodes et al., 2008). In the present study, the effect-action association in the incongruent task involved a type of rearranged pair, in which the effects and action were learned repeatedly during the acquisition phase. Therefore, recollection should be necessary for the older participants to correctly recognize the rearranged association of effect and action that causes the interference effects. However, the recollection deficit of older adults may endorse the incongruent effect-action association, and the interference effect thus does not appear. In this study, although the improved executive function after training enlarged the CNV amplitude of the congruent task, unfortunately, memory performance did not benefit from the training, which might be responsible for the absence of the interference effect, which is reflected by the CNV amplitude of the incongruent task. Given more enduring memory training, a memory benefit may be gained—as shown in Li et al.'s (2016) study—and may hence improve recollection in older adults, which provides evidence for potential intervention targeting the improvement of intention-based action by means of cognitive training. In other words, although the executive function could benefit, the essential improvement of intention-based action may require the sufficient and enduring training of associative memory.

Previous studies have suggested that extensive executive function training could benefit the performance of episodic memory despite the small transfer effect (Buschkuhl et al., 2008; Dahlin et al., 2008). The brain activities involved in associative memory includes both the PFC and the medial temporal lobe (Naveh-Benjamin, 2000; see Yonelinas, 2002 for a review; see Blumenfeld and Ranganath, 2007 for a review). It was also demonstrated that memory strategy utilization required the involvement of executive function (Nyberg et al., 2003; Jones et al., 2006). The interaction between executive function and memory may indicate that although associative memory was responsible for the bidirectional association of action-effect in a specific manner, executive function may also play an important role in intention-based action. As shown in the results of this study, improved executive functions were related to either enlarged CNV amplitudes in congruent tasks or decreased CNV amplitudes in incongruent tasks, which appeared to demonstrate that the interference effect of CNV amplitude was caused by improved executive function. However, the inhibitive function reflected by the Stroop task was designed to help the participants focus on the presently required task by excluding the interference of irrelevance, which may possibly reduce the interference effect between congruent and incongruent tasks. Therefore, we believe that the interference effect reflecting the bidirectional action-effect association is caused by other cognitive functions such as associative memory, based on previous research and our assumptions. Unfortunately, the training effect of memory were not covered in this study, which may account for the

absent interference effect of CNV in intention-based action after training. Nevertheless, the improved executive function after training may mainly benefit intention-based action by means of the enlarged CNV amplitude in the congruent task.

Some limitations should be noted in this study. First, the training hours used for both cognitive functions were relatively short, which may lead to the absence of the memory training effect and hence hinder the demonstration of the correlation between associative memory and bidirectional action-effect association in intention-based action. Further studies targeting intention-based action may establish sufficient training time for memory practice to further investigate the correlation and brain mechanism of associative memory in intention-based action. Second, the sample size was small and the individual differences were relatively large because fewer participants completed both the pretest and posttest of ERP tasks and there was a high dropout rate in the control group, which led to a limitation in the statistical power of the test hypothesis. As the results, on one hand, some potential positive effects may not be significantly observed in this study. In Li et al.'s (2016) study, marginal significance was found for the training effect on composite memory scores after the combined cognitive training, whereas no significance was observed for such an effect in this study, which had fewer participants. On the other hand, the limit statistical power could not support the present discussion firmly. As a pilot study, what was more important, it provided a new perspective to investigate the relationship between age-related cognitive decline and action, and discussed the potential mechanism of associative memory underlying intention-based action. Further studies using large sample sizes are also required to reveal the relationship between associative memory and intention-based action.

In sum, the present study provided empirical evidence demonstrating that action could benefit from cognitive training. The potential relationships of executive function and associative memory were discussed in the context of specific action modes. As we assumed, combined cognitive training benefitted both intention-based and stimulus-based actions. The improved executive function may produce extensive benefits to both actions, whereas associative memory may be specifically beneficial to the bidirectional action-effect association of intention-based action. A comparison of the cognitive processes between intention-based and stimulus-based action was introduced in this study to investigate the age-related decline and the brain plasticity of motor cognition in older adults, which offers a new pathway to investigate age-related motor cognition (in addition to the present research concerning movement disorders such as motor slowness, gait balance disorders and motor discoordination).

AUTHOR CONTRIBUTIONS

Y-NN and JL conceptualized the design. Y-NN took part in the data collection and wrote the draft. X-YZ took part in the data collection and contributed to discussion. JL critically reviewed and edited the manuscript. J-NF took part in the data analysis.

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SUPPLEMENTARY MATERIAL

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The Benefits of Working Memory Capacity on Attentional Control under Pressure

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The present study aimed to examine the effects of working memory capacity (WMC) and state anxiety (SA) on attentional control. WMC was manipulated by (a) dividing participants into low- and high-WMC groups (Experiment 1), and (b) using working memory training to improve WMC (Experiment 2). SA was manipulated by creating low- and high-SA conditions. Attentional control was evaluated by using antisaccade task. Results demonstrated that (a) higher WMC indicated better attentional control (Experiments 1 and 2); (b) the effects of SA on attentional control were inconsistent because SA impaired attentional control in Experiment 1, but favored attentional control in Experiment 2; and (c) the interaction of SA and WMC was not significant (Experiments 1 and 2). This study directly manipulated WMC by working memory training, which provided more reliable evidence for controlled attention view of WMC and new supportive evidence for working memory training (i.e., far transfer effect on attentional control). And the refinement of the relationship between anxiety and attentional control proposed by Attentional Control Theory was also discussed.

Keywords: attentional control, working memory capacity, state anxiety, working memory training, antisaccade

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INTRODUCTION

Attentional control is one of the key components of human perception, which requires an individual to focus on the task-relevant information and resist the interference of task-irrelevant information (i.e., distractor) (Knudsen, 2007). It is important for people who would like to maintain concentrated for certain task, especially under some stressful situations that may induce anxiety, such as examination, surgery, aviation, and competitive sports.

The controlled attention view of working memory capacity (WMC; Kane et al., 2001; Engle, 2002) suggests that WMC is not about individual differences in how many items can be stored per se but about differences in the ability to control attention to suppress interference, avoid distraction and maintain information in an active, quickly retrievable state. More important, WMC and attentional control share a similar neural system (i.e., prefrontal cortex) (Kane and Engle, 2002; van Veen and Carter, 2006). The controlled attention view of WMC proposed that high-WMC individuals are generally better able to maintain top-down attentional control and remain focused, whereas low-WMC individuals are likely to experience failures in goal maintenance due to their inability to inhibit distraction or interference (Kane et al., 2001; Engle, 2002; Barrett et al., 2004; Unsworth et al., 2004). There

Abbreviations: OSPAN, operation-word span task; SA, state anxiety; TA, trait anxiety; WM training, working memory training; WMC, working memory capacity.

are numerous studies using various paradigms support this prediction (e.g., Kane et al., 2001; Unsworth et al., 2004; Colflesh and Conway, 2007; Fukuda and Vogel, 2011; Furley and Memmert, 2012; Hiebel and Zimmer, 2015).

Another separate line concerning attentional control is about anxiety and attentional control. Attentional Control Theory (Eysenck et al., 2007) proposed that anxiety creates an imbalance between two attentional systems: goal-directed (top-down) system (responsible for the maintenance of task goals) and stimulus-driven (bottom-up) system (sensitive and responsive to salient stimuli). Successful attentional control requires the processing of goal-directed system, whereas anxiety will decrease the influence of goal-directed system and increase the influence of the stimulus-driven system. In other words, anxiety will impair attentional control and lead to distraction. This prediction was supported by many researchers (e.g., Derakshan et al., 2009; Wilson et al., 2009a; Edwards E.J. et al., 2015; for reviews see Derakshan and Eysenck, 2009; Eysenck and Derakshan, 2011), especially sport psychologists (e.g., Wilson et al., 2009b; Wood and Wilson, 2010; Nieuwenhuys and Oudejans, 2010; Causer et al., 2011; Navarro et al., 2013).

One important issue in Attentional Control Theory is the type of anxiety. Anxiety can be differentiated into trait anxiety (TA) and state anxiety (SA) (Spielberger et al., 1983). TA is a personality dimension characterized by a stable and chronic propensity to experience moderate to high levels of anxiety in general, whereas SA is a more acute and transient emotional experience of anxiety triggered by situational stress or pressure. Previous studies about Attentional Control Theory provided consistent evidences for the prediction that TA will impair attentional control (e.g., Ansari et al., 2008; Derakshan et al., 2009; Moser et al., 2012; Edwards E.J. et al., 2015). However, there were inconsistent evidences for the effect of SA on attentional control, which challenged Attentional Control Theory (e.g., SA showed negative effect: Wood and Wilson, 2010; Navarro et al., 2013; Allsop and Gray, 2014; SA showed null effect: Moser et al., 2012; Edwards E.J. et al., 2015; Hoskin et al., 2015; SA showed positive effect: Booth and Sharma, 2009). It is worthy to further explore the effect of SA on attentional control. We think that the inconsistent effects of SA could attribute to the different SA levels in different studies, because the SA conditions are different in different studies so that the induced SA levels are different. Relationship between SA levels and task performance is supposed as an inverted-U curve, and a moderate SA will favor task performance (Yerkes and Dodson, 1908; Spielberger et al., 1983; Jones, 1995). Although task performance is not equal with attentional control, we think that the effect of SA on task performance could prompt the effect of SA on attentional control. We infer that there could be two different effects of SA on attentional control (i.e., both negative and positive effects, which conflict with each other). Considering that the effect of SA is unclear in contrast to TA as discussed above, the present study would focus on SA.

Given the views of Attentional Control Theory (i.e., anxiety impairs attentional control) and controlled attention view of WMC (i.e., high-WMC favors attentional control), it is very

possible that WMC could modulate the effect of anxiety on attentional control, that is, high-WMC individuals with high-TA will be better at attentional control compared with low-WMC individuals with high-TA; or high-WMC individuals will be better at attentional control under pressure (i.e., under SA condition) compared with low-WMC individuals. To date, several studies have explored the effects of WMC and anxiety (including TA and SA) on attentional control (Booth and Sharma, 2009; Johnson and Gronlund, 2009; Edwards M.S. et al., 2015; Wood et al., 2015; Wright et al., 2014), but the results of these studies are inconsistent. For example, two studies (Johnson and Gronlund, 2009; Wright et al., 2014) found significant interaction of WMC and anxiety (i.e., the deficit of attentional control under anxiety was less obvious for high-WMC individuals compared with low-WMC individuals, and it should be mentioned that the “anxiety” in these two studies was TA), but Wood et al. (2015) did not find this interaction (the “anxiety” in this study was SA). Furthermore, Booth and Sharma (2009) even found a significant interaction with opposite pattern (i.e., the increase, not deficit, of attentional control under anxiety was more obvious for high-WMC individuals compared with low-WMC individuals, and the “anxiety” here was SA).

There might be two reasons for obtaining these different results. First, the manipulations of anxiety are different. Studies employed TA showed consistent results, whereas studies employed SA showed inconsistent results. This pattern is similar with studies on Attentional Control Theory mentioned above. So we did a similar inference that different SA conditions induce different SA levels so that the effects of SA were inconsistent. For example, Edwards M.S. et al. (2015) might have induced relatively low SA level, because they did not manipulate SA conditions directly (they only measured the SA levels using questionnaires after experiment); Booth and Sharma (2009) might have induced relatively moderate SA level, because they used single-source SA condition: noise punishment (that is why the SA effect in this study was positive, because moderate SA might favor performance); Wood et al. (2015) might have induced relatively high SA levels, because they used multi-sources SA condition: gun shooting threat and peer comparison. So we argue that multi-sources SA condition could be a better way to induce SA, and we would also employ this SA condition in the present study.

Second, some attentional control measurements in these studies (e.g., Stroop task, Booth and Sharma, 2009; highly demanding dual-task, Johnson and Gronlund, 2009; attentional shifting task, Edwards M.S. et al., 2015) confounded attentional control (e.g., the eye movement data) and task performance (e.g., accuracy or reaction time), which might make the indicators less sensitive. Attentional Control Theory claims that anxiety impairs attentional control but did not affect task performance directly (Eysenck et al., 2007), because one can invest more efforts to maintain good performance when attentional control is impaired. So a better attentional control measurement is eye movement (Wright et al., 2014; Wood et al., 2015), because the fixation often cued the focus of attention. For example, antisaccade task is often used in studies on Attentional Control

Theory (e.g., Derakshan et al., 2009; Wright et al., 2014), and we would also employ this task in the present study.

Another problem with studies concerning the interaction of anxiety and WMC on attentional control is that no study manipulates WMC directly. A regular approach to explore the effect of WMC is to divide participants into low-WMC group and high-WMC group based on performance of WMC tasks (operation-word span task, i.e., OSPAN is one of the most commonly used measurements). One could argue that the causal link between WMC and attentional control is not solid due to the lack of experimental manipulation. Here we could manipulate WMC directly by using working memory training (WM training). The plasticity of WMC was widely explored in the past decade, numerous studies indicated that WM training could improve WMC, which is regarded as “near transfer” (e.g., Klingberg et al., 2002; Dahlin et al., 2008; Chein and Morrison, 2010; Jaeggi et al., 2011; for review see Shipstead et al., 2012). And the benefits of training could transfer to other aspects such as fluid intelligence (see Au et al., 2015 for meta analyses) or attentional control (e.g., Klingberg et al., 2005; Brehmer et al., 2011; Borella et al., 2014; see Karbach and Verhaeghen, 2014, for meta analyses on older adults), which is regarded as “far transfer.” It should be mentioned that the effectiveness of far transfer is still unclear (some researchers did not support far transfer effect such as Shipstead et al., 2012; Melby-Lervåg and Hulme, 2013; Melby-Lervåg et al., 2016, and the previous studies about far transfer effect on attentional control also produce inconsistent evidences), but the effectiveness of near transfer is widely supported. So we could use WM training to manipulate WMC directly to further confirm the causal link between WMC and attentional control and, at the same time, examine the far transfer (i.e., attentional control) of WM training.

In the present study, we conducted two experiments to examine the effect of WMC and SA on attentional control. Considering the existing problems of studies on this topic mentioned above, multi-sources SA conditions were used to induce relatively high-SA level (Experiments 1 and 2), and antisaccade task was used to evaluate attentional control for a more direct attentional control measurement and separating attentional control from task performance (Experiments 1 and 2). OSPAN was used to evaluate WMC, we divided participants into low- and high-WMC groups based on original OSPAN scores (Experiment 1), and we also manipulated WMC directly using WM training (adaptive n-back training) (Experiment 2). Results of Experiment 1 showed that SA impairs attentional control and high-WMC individuals were better at attentional control, but the interaction of SA and WMC was not significant. Results of Experiment 2 showed that individuals with WM training (i.e., WMC had improved) were better at attentional control compared with individuals without WM training (i.e., WMC had not improved).

EXPERIMENT 1

Previous studies showed inconsistent results of the effects of WMC and anxiety on attentional control as mentioned in

Introduction. Here we conducted Experiment 1 to examine this effect again and attempted to solve potential problems in previous studies as mentioned above. Antisaccade task was used to evaluate attentional control. We divided participants into low- and high-WMC groups based on OSPAN scores and manipulated low- and high-SA using multi-sources SA condition. One reason for choosing SA rather than TA as an anxiety independent valuable is that SA could be manipulated in experiment by setting stressful situation. Nevertheless, we also measured TA as a covariant variable, because previous studies provided consistent evidences for TA impairs attentional control as mentioned in Introduction (e.g., Ansari et al., 2008; Derakshan et al., 2009; Moser et al., 2012; Edwards E.J. et al., 2015). The present study, however, focused on SA (that is, we did not concern about the effect of TA, but there still might be individual differences on TA), so we considered to regard TA as covariant variable to balance the contribution of TA on attentional control. There were three hypotheses in Experiment 1:

H1-1: SA impairs attentional control, that is, the first correct antisaccade latency (latency) would be longer and the percentage of incorrect saccades (error rate) would be higher under high-SA condition compared with low-SA condition. This hypothesis aimed to explain previous inconsistent results of SA on attentional control.

H1-2: high-WMC individuals have better attentional control, that is, the latency would be shorter and the error rate would be lower for high-WMC group compared with low-WMC group. This hypothesis is a replication of previous studies.

H1-3: WMC modulates the effect of SA on attentional control, that is, the increase of latency and error rate under high-SA would be less obvious for high-WMC group compared with low-WMC group. This hypothesis aimed to explain previous inconsistent results of interaction between SA and WMC.

Method

Participants

Sixty four healthy young adults were recruited by flyer. All participants provided informed consent in advance and received ¥50 payment for their participation. This experiment was approved by Beijing Sport University Institutional Review Board (BSUIRB) (Approval Number: 2015037). However, one participant claimed that SA condition was not effective for him/her (he/she felt that he/she was more anxious under low-SA condition compared with high-SA condition. More important, the data of his/her manipulation check also indicated that he/she was strangely far more anxious under low-SA condition compared with high-SA condition) so this data were excluded. Furthermore, seven participants who performed over 40% invalid trials were also excluded (according to Derakshan et al., 2009, for the detail of exclusion criteria, see “measurement of attentional control” below). After the data collection, 56 participants were included (13 males, 43 females; mean age 21.339 ± 2.414 years. In terms of the education level, there were 40 undergraduate students and 16 graduate students in these 56 participants).

Measurement of WMC

Operation-word span task (La Pointe and Engle, 1990) was used to evaluate WMC, which has been widely approved (e.g., Engle et al., 1999; Kane et al., 2001; Unsworth et al., 2004; Wright et al., 2014; Wood et al., 2015). In OSPAN, an operation-word string [e.g., $(3 \times 3) - 5 = 4?$ Train] will be displayed on the screen and participant is required to read the operation aloud, verify aloud whether the operation is correct (“right” vs. “false”; 85% accuracy criterion on the operations is required for all participants), and then finally read the word aloud. Once the participant has read the word aloud, the experimenter presses a key to move onto the next operation-word string. Pausing was not permitted during this process until three question marks (???) cued the participant to recall the words from that set in the correct order (write the words on an answer sheet). The operation-word strings can vary from two to six items in length and each length has three sets (the different set sizes appear in an unpredictable order). Thus, the OSPAN score is the sum of the recalled words for all sets recalled completely in correct order (if participant has recalled words completely but in wrong order, half of the words in this set will be included in the score), and all possible scores that ranged from 0 to 60. Higher OSPAN scores imply higher WMC. This OSPAN task program was developed using E-prime 2.0.

Measurement of Attentional Control

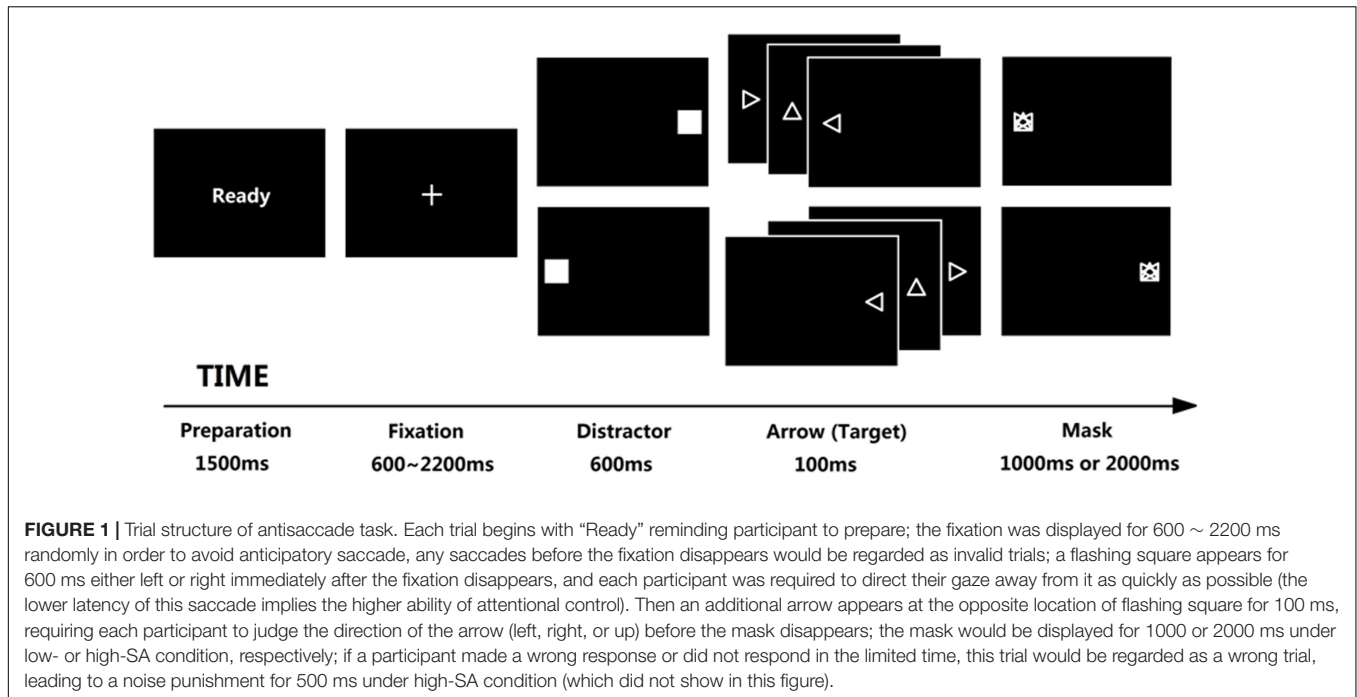
Tobii T120 was used as the eye-tracking device with 120 Hz sampling rate. The stimuli was displayed on the Tobii Eye Tracker (subtending $32.47^\circ \times 25.79^\circ$, resolution is 1024×768 pixels, and refresh rate is 60 Hz). The distance between participant’s eyes and the center of screen was 60 cm. Antisaccade task (Hallett, 1978) was used to evaluate attentional control (Kane et al., 2001; Unsworth et al., 2004; Hutton and Ettinger, 2006; Derakshan et al., 2009; Wright et al., 2014), which was developed using E-prime 2.0. In this task, attentional control was required to suppress a reflexive saccade toward a distractor, and generate a volitional saccade to its mirror position (see **Figure 1** for trial structure). Each trial begins with “Ready” in the center of screen for 1500 ms and participant is required to fixate a cross ($1.2 \text{ cm} \times 1.2 \text{ cm}$, subtending $1.15^\circ \times 1.15^\circ$, displaying for random 600 ~ 2200 ms) until it disappears. A flashing square (i.e., distractor, $5.5 \text{ cm} \times 5.5 \text{ cm}$, subtending $5.25^\circ \times 5.25^\circ$, displaying for 600 ms) then appears either left or right of the center at 13.37° with equal possibility. The participant is then required to direct their gaze AWAY from the flashing square in the opposite location as quickly as possible. Immediately after the presentation of square, a triangle arrow ($1 \text{ cm} \times 1 \text{ cm}$, subtending $0.96^\circ \times 0.96^\circ$, displaying for 100 ms) appears at 13.37° from the center in the opposite direction of the square and followed with a mask ($1 \text{ cm} \times 1 \text{ cm}$, subtending $0.96^\circ \times 0.96^\circ$, displaying for 1000 or 2000 ms in high- and low-SA condition, respectively). The participant needs to identify the arrow’s direction (up, left or right) within limited time by pressing the relevant keys on the keyboard. In fact, we don’t concern the arrow-judgment because it is an explicit task requirement represented for task performance, not the indicator of attentional control (we concern attentional control rather

than task performance). In contrast, the eye movement data in antisaccade task are rather implicit indicators, which are suitable as indicators of attentional control (as mentioned in Introduction, the advantage to use antisaccade task is that we could separate attentional control (AC) from task performance). At last, we only analyzed the eye movement data and ignored the task performance data. Nevertheless, the requirement to identify the arrow direction ensured participants were more engaged in the task.

The two main dependent variables (eye movement data) were the latency of first correct saccade (latency, being the elapsed time between the onset of the distractor and a saccade in the correct direction before the onset of arrow, which reflects the effort of suppressing the attraction of distractor and implies the deficit of attentional control if the participant needed more time to complete a correct saccade) and the percentage of incorrect saccades (error rate, being the percentage of the trials that saccade in the distractor’s direction, which reflects the trends of being attracted by distractor and also implies the deficit of attentional control if participant performed more incorrect saccades). According to Derakshan et al. (2009), a first correct saccade was defined as a first eye movement with a velocity $> 30^\circ/\text{s}$ and amplitude $> 3^\circ$ toward the mirror position of distractor that was made after the onset of the distractor and before the onset of arrow. Similarly, an incorrect saccade was the first saccade toward the position of the distractor after it onset. Trials that latency shorter than 83 ms (i.e., anticipatory) or longer than 600 ms (i.e., saccade failed) were excluded. Also, trials would be regarded as invalid trial when the eye tracker failed to sample that trial. There were 36 trials in 1 block. Participants’ data would be excluded if they performed more than 15 trials (over 40%) should be excluded (all these exclusion criteria above were according to Derakshan et al., 2009, and 7 data were excluded in Experiment 1).

The SA Condition

Multi-sources SA condition (i.e., limited wrong response, limited reaction time, noise punishment, and electric shock threat) were used to induce SA. There were two SA conditions: low- and high-SA. In the low-SA condition, each participant was required to keep the number of wrong judgments of the arrow direction (error times) within three (i.e., maximum error times were three), and the reaction time to judge the arrow direction was limited within 2 s, or this trial would be regarded as a wrong judgment (i.e., maximum RT was 2 s). Whereas in the high-SA condition, the maximum error time was only one and the maximum RT was 1 s. Once a participant made a wrong judgment, he/she would be punished by white noise (lasting 500 ms, which was generated by Cool Edit Pro V2.1, and presented via EDIFIER headphones. The actual intensity of the noise is 95.730 ± 2.545 dB measured by using BENETECH GM1356 decibel device for 10 times. The participant was told the intensity is 110 dB, and would listen to the noise before high-SA condition task in order to ensure the effectiveness of inducing high-SA). Moreover, the experimenter would stand aside and hold an electric stimulator (TAIMENG BL-420S Biological Experimental System with 24V DC power). Participant was told that he/she could be shocked by the electric



stimulator at any time (may not immediately) if a wrong judgment was made (but it actually was just a kind of electric threat, because participant would never be shocked during the experiment). Besides, both physiological and psychological measurements were used as SA manipulation check: participant’s heart rate and skin conductance were recorded during the experiment using biofeedback device (NEXUS-10 MARK II) as physiological measurement, and participant was also required to fill the mental readiness form-3 (MRF-3, Krane, 1994) as psychological measurement. MRF-3 is an 11-point Likert scale with three items to evaluate cognitive anxiety, somatic anxiety and state confidence, which is applicable to measure the SA (Krane, 1994; Wilson et al., 2009b; Wood and Wilson, 2010).

Procedure

Participants completed the Trait-Anxiety Inventory (T-AI, part of the State-Trait Anxiety Inventory, Spielberger et al., 1983) after informed consent. Then, they completed the OSPAN, including practice (2 sets with 5 operation-word strings) and formal task (15 sets with 60 operation-word strings). After that, participants could have a break and the experimenter would help them equipping the biofeedback (heart rate and skin conductance would be recorded from now on till the end of experiment). Participants subsequently conducted a practice block of antisaccade task (12 trials), and then completed a practice block again after calibration of the eye-tracker. In the formal antisaccade task, participants needed to conduct a low-SA block and a high-SA block, respectively. Each block had 36 trials, and participants were required to complete the MRF-3 after each block (the sequence of low- and high-SA block was determined by random lottery). At last, participants were interviewed briefly.

Results

We did the manipulation check of SA condition first. The evaluations of SA level (heart rate, skin conductance, and MRF-3 scores) were analyzed. The heart rate, $F(1,55) = 38.320$, $p < 0.001$, $\eta_p^2 = 0.411$, skin conductance, $F(1,55) = 22.639$, $p < 0.001$, $\eta_p^2 = 0.292$, and MRF-3 scores, $F(1,55) = 92.905$, $p < 0.001$, $\eta_p^2 = 0.628$, were all significantly increased in the high-SA condition, implying that SA manipulation was successful.

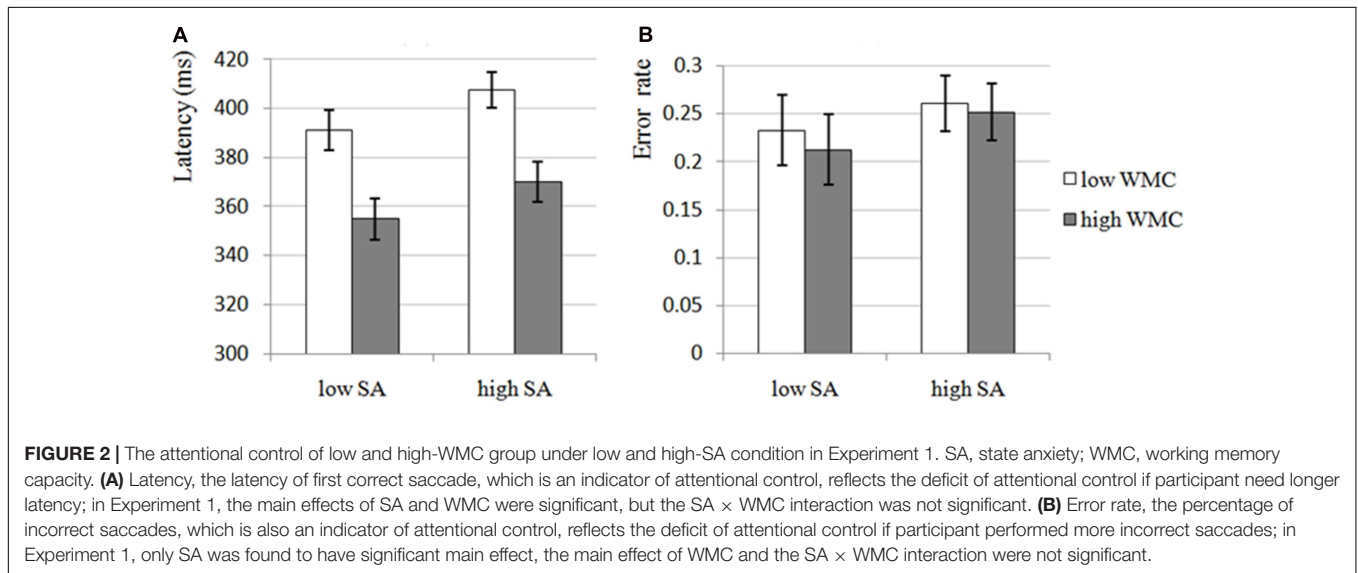
Then, 56 participants were sorted based on the OSPAN scores, and half of them were selected from the top half of the distribution as high-WMC group ($n = 28$), and another half as low-WMC group ($n = 28$), see **Table 1** for the descriptive data of OSPAN scores. We originally would like to conduct a 2 (SA Condition as within-participants factor: low vs. high) \times 2 (WMC Group as between-participants factor: low vs. high) ANCOVA for the two indicators (i.e., latency and error rate, respectively) of attentional control, and the covariant variable was TA measured by T-AI. But the preliminary analysis revealed that TA was not applicable as a covariant variable¹, so typical 2 \times 2 ANOVA was then conducted for latency and error rate respectively, regardless of TA (it should be noted there was no significant difference on T-AI scores between low- and high-WMC group, $t(54) = -0.439$, $p = 0.662$, demonstrating that that TA is not the main contributor of the effects on latency or error rate). See **Table 1** for the descriptive data of latency and error rate.

¹An applicable covariant variable should meet two prerequisites at the same time: (a) correlate with dependent variable and (b) independent with independent variable. Here the preliminary analysis of ANCOVA revealed that TA was not applicable as a covariant variable, because the main effect of TA was not significant, that is, the covariant variable was unrelated with dependent variables, for latency: $F(1,52) = 0.771$, $p = 0.384$, for error rate: $F(1,52) = 1.703$, $p = 0.198$, which did not meet the first prerequisite, so we exclude TA and conducted typical ANOVA.

TABLE 1 | Working memory capacity and attentional control in Experiment 1 (means, with standard deviations in parentheses).

Indicators	Low WMC		High WMC	
	Low-SA	High-SA	Low-SA	High-SA
WMC				
OSPANs	11.679 (3.418)		22.875 (5.319)	
Attentional control				
Latency	391.094 (43.065)	407.443 (44.490)	354.965 (38.808)	370.109 (43.580)
Error rate	0.233 (0.194)	0.261 (0.194)	0.213 (0.155)	0.252 (0.160)

WMC, working memory capacity; SA, state anxiety; WM training, the working memory training group; Control, the control group; OSPANs, operation-word span task scores; Latency, the latency of first correct saccade; Error rate, the percentage of incorrect saccades.



The results of 2×2 ANOVA for latency and error rate showed that the main effects of SA Condition were significant for both latency, $F(1,54) = 12.988$, $p = 0.001$, $\eta_p^2 = 0.194$, and error rate, $F(1,54) = 6.199$, $p = 0.016$, $\eta_p^2 = 0.103$, that is, there were significant increases in high-SA condition compared with low-SA condition for both latency (see **Figure 2A**) and error rate (see **Figure 2B**), which was consistent with H1-1, demonstrating that high-SA impairs attentional control. Furthermore, the main effects of WMC Group were significant for latency, $F(1,54) = 12.246$, $p = 0.001$, $\eta_p^2 = 0.185$, but not for error rate, $F(1,54) = 0.103$, $p = 0.749$, $\eta_p^2 = 0.002$, that is, there was a significant decrease in high-WMC group compared with low-WMC group for latency (see **Figure 2A**), but not for error rate (see **Figure 2B**), which was still consistent with H1-2, demonstrating that high-WMC individuals have better attentional control (the non-significant result for error rate would not affect this inference too much, and it would be discussed in the Section “General Discussion”). Unfortunately, none of the interactions were significant (all $ps > 0.663$), which was inconsistent with H1-3, demonstrating that the effects of SA and WMC on attentional control seems to be independent with each other.

Discussion

In Experiment 1, we manipulated low- and high-SA and divided participants into low- and high-WMC group to examine the effects of SA and WMC on attentional control. Results completely supported Attentional Control Theory (i.e., H1-1): attentional control was impaired under high-SA (see also Wilson et al., 2009a; Wood and Wilson, 2010; Navarro et al., 2013; Allsop and Gray, 2014). Given that we used multi-sources SA condition to induce relatively high-SA and got this result, it implies that the inconsistent results of SA on attentional control might be explained by the different SA levels induced by different SA conditions in different studies. Comparing with TA, the effect of SA is more complex due to the inference that the effect depends on the SA level: the positive effects of SA (e.g., improvement of motivation or arousal) might counteract the negative effects of SA (e.g., impairment of attentional control) if the SA level is relatively low or moderate.

Furthermore, the results of Experiment 1 supported controlled attention view of WMC (i.e., H1-2): high-WMC individuals have better attentional control (see also Conway et al., 2001; Kane et al., 2001; Unsworth et al., 2004; Colflesh and Conway, 2007; Fukuda and Vogel, 2009, 2011; Unsworth and Spillers, 2010; Unsworth and Robison, 2016). High-WMC individuals may get

more attentional resources to cope with the distractor (distractor cannot be avoided in many cases) during the time when they were doing the main task. It should be mentioned, however, that this kind of benefit was not observed for error rate (in fact, we would see a similar null-effect again in Experiment 2 and this will be discussed in the Section “General Discussion”).

Unfortunately, the results of Experiment 1 did not support the prediction of the possible interaction (i.e., H1-3): the SA \times WMC interaction did not affect attentional control (see also Wright et al., 2014; Wood et al., 2015). Wright et al. (2014) suspected that the mathematical operations in the OSPAN task may be anxiety provoking, leading to an underestimation of WMC. This implies that OSPAN may lead to negative feeling (e.g., low self efficacy) due to higher difficulty than other WMC measurements, which may affect task performance. But OSPAN is one of the most effective measurements of WMC (Engle et al., 1999). So we speculate that the effect of SA and WMC on attentional control might be independent with each other, that is, SA and WMC might affect different aspects of attentional control (this will be discussed in Section “General Discussion”).

EXPERIMENT 2

In Experiment 1, we divided participants into low- and high-WMC group based on OSPAN scores, just like most of previous studies (e.g., Conway et al., 2001; Kane et al., 2001; Unsworth et al., 2004; Colflesh and Conway, 2007; Fukuda and Vogel, 2011; Furley and Memmert, 2012; Hiebel and Zimmer, 2015; Wood et al., 2015). However, one shortcoming of these studies (including Experiment 1) was that researchers did not manipulate WMC directly (they divided participants into low- and high-WMC groups based on original WMC rather than directly manipulate WMC), which relied heavily on samples, and it was not enough to infer the relationship between WMC and attentional control. Experiment 2 was conducted to explore the effect of SA and WMC on attentional control again, and we manipulated both SA (the same with Experiment 1) and WMC (using WM training).

It is reasonable to use WM training to manipulate WMC, because the near transfer effect (i.e., WMC would be improved after WM training) has been widely supported in previous studies (see Melby-Lervåg and Hulme, 2013; Karbach and Verhaeghen, 2014; Au et al., 2015; Melby-Lervåg et al., 2016 for meta analysis). Besides, however, the far transfer effect (i.e., whether the benefits of WM training would transfer to attentional control) is still unclear. Shipstead et al. (2012) suggested that studies in this area have far relied heavily on the Stroop task to evaluate attentional control, future studies should employ a variety of tasks that converge on the attention construct such as antisaccade task. So Experiment 2 could also examine the far transfer effect of WM training using antisaccade task (which is a novel task in this field) to evaluate attentional control. There were three hypotheses in Experiment 2 (it should be mentioned that we did not find the SA \times WMC interaction in Experiment 1 and we speculate that the effects

of SA and WMC are independent with each other, so we did not propose any prediction about interaction effects in Experiment 2):

H2-1: WMC will be improved after WM training (i.e., near transfer), that is, the OSPAN scores of WM training group would be higher compared with control group after training. This hypothesis is a replication of previous studies.

H2-2: individuals with WM training have better attentional control after training (i.e., far transfer), that is, the latency and error rate of WM training group would be lower compared with control group after training. This hypothesis would provide a more reliable evidence for the causal relationship of WMC and attentional control.

H2-3: SA impairs attentional control, that is, the latency and error rate would be higher under high-SA condition compared with low-SA condition. This hypothesis would be helpful for explain previous inconsistent results of effects of SA on attentional control (taken together with H1-1).

Method

Participants

Thirty two participants of Experiment 1 were selected as the participants of Experiment 2. The screening process was (a) 7 of 56 participants in Experiment 1 were excluded first (they performed more than 10 invalid trials in one antisaccade block) to ensure more acceptable data in Experiment 2; (b) the experimenter invited participants to attend Experiment 2 one by one from the rest of 49 participants based on the order of OSPAN scores in Experiment 1 (from lowest to highest) to ensure more obvious training effect in Experiment 2; (c) at last, we had sent 36 invitations and 32 participants accepted. And the training sessions were started 1 week after Experiment 1 (without training, the WMC should be stable in this kind of short period). Participants provided informed consent in advance and received ¥200 payment for their participation. This experiment was approved by Beijing Sport University Institutional Review Board (BSUIRB) (Approval Number: 2015037). These 32 participants' data in Experiment 1 were regarded as the pre-training data in Experiment 2, and they were randomly (toss) matched into WM training group ($n = 16$) and control group ($n = 16$) based on OSPAN scores, there was no significant difference on pre-training OSPAN scores between WM training and control group, $t(30) = -0.156$, $p = 0.877$. At last, no data were excluded according to the same criteria in Experiment 1, so all 32 participants were included (9 male, 23 female; mean age 21.000 ± 1.481 years).

Training Task

Adaptive spatial n-back training (Jaeggi et al., 2011, 2014) was utilized for the WM training group, and adaptive spatial 1-back training was utilized for the control group. In a spatial n-back task, participants were presented with a sequence of stimuli (i.e., a blue square) appearing at random spatial locations on the screen, one at a time at a rate of 3 s (stimulus length is 500 ms; judgment interval is 2500 ms). Participants were required to press a key whenever the currently presented stimulus was at the same

TABLE 2 | The difficulty levels for WM training group and control group.

Level	WM training group					Control group				
	Task	spatial	Block	Time	Error(s)	Task	spatial	Block(s)	Time	Error(s)
1	1-back	7	1	2500	2	1-back	25	1	1000	2
2	1-back	9	1	2500	2	1-back	25	2	1000	4
3	2-back	9	1	2500	2	1-back	25	2	700	4
4	2-back	11	1	2500	2	1-back	25	3	700	6
5	3-back	11	1	2500	2	1-back	25	3	600	6
6	3-back	13	1	2500	2	1-back	25	4	600	8
7	4-back	13	1	2500	2	1-back	25	4	550	8
8	4-back	15	1	2500	2	1-back	25	5	550	10
9	5-back	15	1	2500	2	1-back	25	5	500	10
10	5-back	17	1	2500	2	1-back	25	5	500	5
11	6-back	17	1	2500	1	1-back	25	5	400	5
12	6-back	19	1	2500	1	1-back	25	5	400	1
13	7-back	19	1	2500	1	1-back	25	5	300	5
14	7-back	21	1	2500	1	1-back	25	5	300	1
15	8-back	21	1	2500	1	1-back	25	5	250	5
16	8-back	23	1	2500	1	1-back	25	5	250	1
17	9-back	23	1	2500	1	1-back	25	5	230	5
18	9-back	25	1	2500	1	1-back	25	5	230	1
19	10-back	25	1	2500	1	1-back	25	5	220	5
20	10-back	27	1	2500	1	1-back	25	5	220	1

Task, the task participants should complete in this level; Spatial, the number of possible locations of the stimuli; Block(s), the number of blocks participants should complete in this level; Time, maximum response time of each trial in this level (ms); Errors, maximum error times allowed to pass this level.

TABLE 3 | Working memory capacity and attentional control in Experiment 2 (means, with standard deviations in parentheses).

Indicators	Pre-training				Post-training			
	WM training		Control		WM training		Control	
	Low-SA	High-SA	Low-SA	High-SA	Low-SA	High-SA	Low-SA	High-SA
WMC								
OSPANs	14.656 (5.036)		14.938 (5.147)		24.719 (11.312)		19.969 (9.177)	
Attentional control								
Latency	359.179 (33.018)	379.813 (45.241)	369.916 (52.576)	387.399 (51.719)	333.286 (32.058)	338.738 (30.633)	382.246 (58.466)	370.708 (49.867)
Error rate	0.168 (0.101)	0.200 (0.095)	0.191 (0.138)	0.271 (0.114)	0.174 (0.115)	0.200 (0.164)	0.171 (0.131)	0.205 (0.124)

WMC, working memory capacity; SA, state anxiety; WM training, the working memory training group; Control, the control group; OSPANs, operation-word span task scores; Latency, the latency of first correct saccade; Error rate, the percentage of incorrect saccades.

location as the one n item(s) back in the series (targets), and press another key if that was not the case (non-targets). There were 7 targets and 14 non-targets of trials per block (which included $21 + n$ trials). Whereas in a spatial 1-back task, the most important difference with the n -back task above is that participants were required to press a key whenever the currently presented stimulus was at the same location as the previous one in the series, and press another key if that was not the case.

In each training session, participants in both groups were required to complete 15 blocks, which lasted 20~30 min. For WM training group, the difficulty level was adjusted according to the participants' performance after each block (see **Table 2**,

participants started from level 1. If the error times were less than the requirement, then participants would pass this level and the present level would increase one; if the error times were more than the requirement, then participants would stay in the present level and repeat this level; if participants failed to pass one certain level for three times, than the present level would decrease one). For the control group, the number of n was always 1 (which would not affect WMC in general), and the difficulty level was also adjusted according to participants' performance. The difficulty level was manipulated by changing the interval of judgment, number of blocks in one level, and the error times allowed in one level (see **Table 2** for details),

that is, we designed the tasks for control group like a fast-response game (i.e., the response interval would be shorter and shorter along with the increasing of difficulty level), besides, the blocks that participants should complete would also increase along with the increasing of difficulty level, and the error times allowed in one level would also be changed based on difficulty level. This kind of design could ensure that the treatment of WM training and control group was almost the same (including the improvement of achieved difficulty level based on participants' performance which is known as "adaptive task"), but the control group was always conducting 1-back task. The last achieved difficulty level would be recorded after participants had finished one training session (i.e., 15 blocks), and then participants would start the next training session from this level.

There were 15 training sessions for both groups, and participants would complete one session per day. After each training session, participants were required to answer 3 manipulation check questions which were (a) how concentrated do you think you were in this training session (i.e., perceived attention level); (b) how difficult do you think the task was in this training session (i.e., perceived difficulty level); and (c) how attractive do you think the task was in this training session (i.e., perceived attraction level). All these questions were 7-points Likert evaluation. Participants could get "points" after they achieved a higher level, and they could get extra monetary reward based on how many points they have at the end of all training sessions.

Measurement of WMC, Measurement of Attentional Control, and the SA Condition

All were the same with Experiment 1.

Procedure

Participants were invited to take part in Experiment 2 and matched into WM training group and control group based on pre-training OSPAN scores (i.e., OSPAN scores measured in Experiment 1). Both groups started training at the same time, and participants were required to go to the lab every day to complete one training session within a certain period of time. If someone could not go to the lab due to any problem, the experimenter would send participants an Email with the training program attached. Participants would then complete the training session whenever convenient, and send the training data back to experimenter (This occurred 1 time for 8 participants, 2 times for 1 participant, and 3 times for 3 participants in the total 15 times of training). If someone even did not have time to complete one training session 1 day, there would be a break and participants needed to be trained one more day to achieve the required number of training sessions (This occurred 1 time for 1 participant, and 2 times for 2 participants in the total 15 times of training). After 15 training sessions, participants conducted OSPAN and antisaccade task like Experiment 1 (also under same SA conditions as in Experiment 1). At last, participants were interviewed briefly.

Results

Profile of WM Training

The manipulation check of WM training was conducted first. There were no significant differences on perceived attention level, perceived difficulty level, or perceived attraction level between WM training group and control group (all $ps > 0.373$), indicating that these extra variables unlikely contributed to the differences between these two groups. More improvement, we could see from **Figure 3A** that the performance of the trained task (i.e., n-back task) of WM training group has increased overtime, and in order to demonstrate that this kind of improvement was significant, we compared the mean level ($M = 6.719$, $SD = 1.798$) achieved in the first two training sessions and the mean level ($M = 11.688$, $SD = 3.219$) achieved in the last two training sessions (according to Jaeggi et al., 2011): there was a significant improvement on the achieved difficulty level (i.e., the performance of trained task) for the WM training group, $t(15) = -8.242$, $p < 0.001$. In contrast, for the control group, the training task was always 1-back, which would not affect WMC in general as mentioned above (i.e., although the apparent performance of control group seemed increasing from **Figure 3A**, the actual "performance" of control group on the n-back task was always 1-back with the increasing of difficulty level). In summary, the profile of training indicated that participants in WM training group showed a significant improvement on a trained WMC task (i.e., n-back task) after training, but the participants in control group did not, that is, the WMC of WM training group should be higher than control group after training.

Near Transfer of WM Training

Near transfer refers to the effect of WM training transferring to other untrained WMC task. In the present study, the training task was n-back task (see the "Profile of WM training" above) and the near transfer task was OSPAN (see **Table 3** for the descriptive data). We used the change of OSPAN scores (i.e., post-training OSPAN scores minus pre-training OSPAN scores, the average OSPAN change was 10.063 ($SD = 8.181$) for WM training group, and 5.031 ($SD = 7.288$) for control group) as dependent valuable and conducted a one-way ANOVA (Training Group as between-participants factor: WM training vs. control). Results indicated that there was a trend-level difference between WM training group and control group for the improvement of OSPAN scores after training, $F(1,30) = 3.374$, $p = 0.076$, $\eta_p^2 = 0.101$, demonstrating that participants in WM training group showed better performance on untrained WMC task (i.e., OSPAN) after WM training compared with control group (i.e., a near transfer effect, see **Figure 3B**). Combined this result with the "Profile of the WM training" above, we inferred that WMC was improved for the WM training group after WM training, which supported H2-1. Now we have successfully manipulated the WMC, and we could examine the effect of SA and WMC on attentional control again.

Re-examination for the Effects of SA and WMC on Attentional Control

The manipulation check of SA was conducted first: the heart rate, skin conductance and MRF-3 scores were all significantly

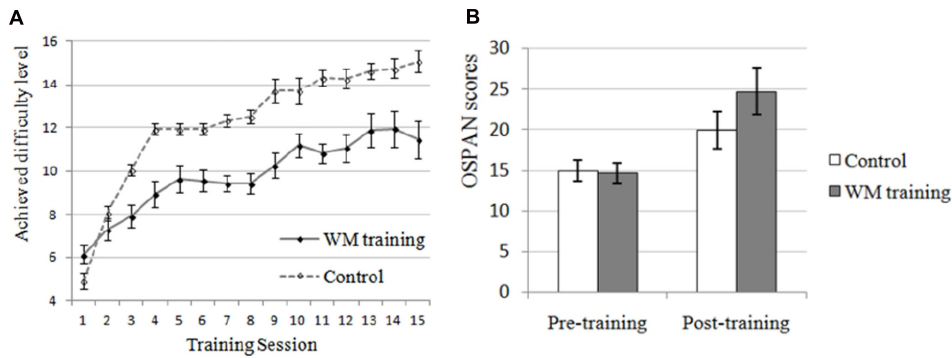


FIGURE 3 | Effects of working memory (WM) training on trained and untrained task in Experiment 2. **(A)** WM training group performed better and better on the training task (n-back) along with the increase of training sessions, demonstrating that the WMC of participants in WM training group was improved (they completed n-back task, and the number of n was increased along with the enhancement of achieved difficulty level). Although the apparent performance of control group seemed increasing, the actual “performance” of control group on the n-back task was always 1-back with the increasing of difficulty level. In contrast, the performance of WM training group for the n-back task was increasing from 1-back to 1 + n-back with the increasing of difficulty level. It is unnecessary and improper to compare the apparent performance of n-back task and 1-back task. **(B)** OSPAN, operation-word span task, which is an untrained WMC task as an evaluation of near transfer effect. The post-training OSPAN score for the WM training group was higher than the control group (trend-level), demonstrating higher WMC for the WM training group after training. It should be mentioned that the post-training OSPAN score was also improved after training for the control group, which might due to the familiarity effects, placebo effects of training, or easier training tasks for the control group that might improve self-efficacy.

increased in the high-SA condition (all $ps < 0.002$ for both pre- and post-training), which implied that SA manipulation was successful for both pre- and post-training. We used the change of latency [i.e., post-training latency minus pre-training latency]. The average change of latency under low-SA condition was -25.894 ($SD = 20.702$) for WM training group, and 12.330 ($SD = 47.953$) for control group, whereas under high-SA condition was -41.075 ($SD = 36.636$) for WM training group, and -16.691 ($SD = 33.313$) for control group] and the change of error rate [i.e., post-training error rate minus pre-training error rate]. The average change of error rate under low-SA condition was 0.006 ($SD = 0.087$) for WM training group, and -0.020 ($SD = 0.125$) for control group, whereas under high-SA condition was 0.001 ($SD = 0.132$) for WM training group, and -0.067 ($SD = 0.079$) for control group] as dependent variables. We originally would like to conduct 2 (Training Group as between-participants factor: WM training vs. control) \times 2 (SA Condition as within-participants factor: low vs. high) ANCOVA for the change of latency and error rate, respectively, and the covariant variable was TA measured by T-AI. But the preliminary analysis showed that TA was not a applicable covariant variable², so typical 2 \times 2 ANOVA was then conducted for the change of latency and error rate respectively, regardless of TA (see Table 3 for the descriptive data). It should be noted that there was no significant difference on T-AI between WM training and control group, $t(30) = -1.404$, $p = 0.171$, demonstrating that that TA is not the main contributor of the effects for attentional control.

²The reason to include TA as covariant variable here is that there might be differences on TA between WM training group and control group, which might affect attentional control but we don't concern it. Here the preliminary analysis of ANCOVA revealed that TA was not applicable as a covariant variable, because the main effect of TA was not significant, that is, the covariant variable was unrelated with dependent variables, for the change of latency: $F(1,28) = 0.946$, $p = 0.339$, for the change of error rate: $F(1,28) = 0.011$, $p = 0.918$, which did not meet the first prerequisite of ANCOVA, so we exclude TA and conducted typical ANOVA.

The results of 2 \times 2 ANOVA showed that: for the change of latency, (a) the main effect of Training Group was significant, $F(1,30) = 7.012$, $p = 0.013$, $\eta_p^2 = 0.189$, that is, the latency of WM training group decreased more than control group after training (see Figure 4A), demonstrating that WM training group have better attentional control than control group after training, which supported H2-2. (b) The main effect of SA condition was significant, $F(1,30) = 22.082$, $p < 0.001$, $\eta_p^2 = 0.424$, that is, the latency under high-SA condition decreased more than under low-SA condition after training (see Figure 4A), demonstrating that SA favors attentional control after training, which was contrary to the H2-3, and also inconsistent with the results of Experiment 1 (i.e., H1-1). (c) The Training Group \times SA Condition interaction was not significant, $F(1,29) = 2.165$, $p = 0.152$, $\eta_p^2 = 0.067$, which was consistent with the results of Experiment 1. Furthermore, for the change of error rate (see Figure 4B), none of these effects were significant (all $ps > 0.162$).

Far Transfer of WM Training

Far transfer refers to the effect of WM training transferring to other untrained related tasks but not WMC tasks (e.g., attentional control tasks, fluid intelligence tasks, etc). In the present study, the training task was n-back task (see the “Profile of WM training” above), the near transfer task was OSPAN (see the “Near transfer of WM training” above), and the far transfer task was antisaccade task. As described above, the performance of WM training group on the trained task (i.e., n-back task) and untrained task (i.e., OSPAN) were improved after training. More important, WM training group have better attentional control than control group after training, which supported H2-2 and also implied that the benefit of WM training has transferred to the untrained, non-WMC task (i.e., far transfer effect).

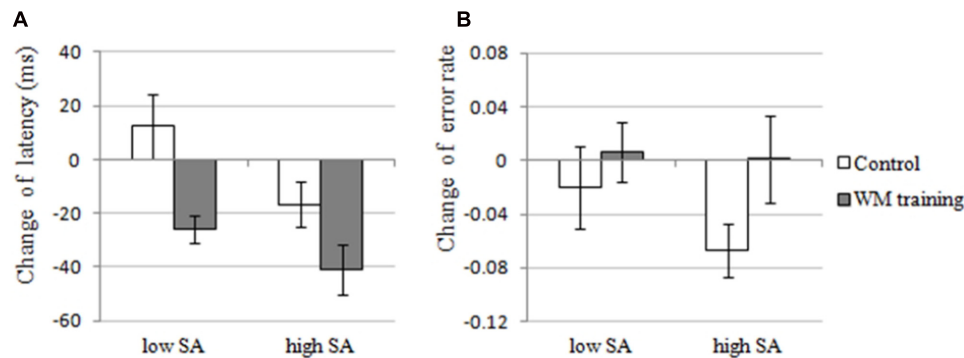


FIGURE 4 | The changes of attentional control of the WM training group and the control group under low and high SA in Experiment 2. **(A)** Latency, the latency of first correct saccade, which is an indicator of attentional control, reflects the deficit of attentional control if participant need longer latency, and the change of latency was calculated by post-training latency minus pre-training latency. In Experiment 2, the main effects of Training Group and SA Condition were significant, but the interaction effect was not significant for the change of latency. **(B)** Error rate, the percentage of incorrect saccades, which is also an indicator of attentional control, reflects the deficit of attentional control if a participant performed more incorrect saccades, and the change of error rate was calculated by post-training error rate minus pre-training error rate. In Experiment 2, none of the effects were significant for the change of error rate.

Discussion

In Experiment 2, we sought to examine the effect of SA and WMC on attentional control again by manipulating both SA (the same manipulation with Experiment 1) and WMC (using adaptive n-back training). Results revealed that the performance of n-back task (the trained WMC task) had been enhanced after training (see **Figure 3A**, see also Dahlin et al., 2008; Schmiedek et al., 2010; Jaeggi et al., 2011), and the performance of OSPAN (an untrained WMC task) had also been enhanced after training, demonstrating a near transfer effect of WM training (H1-1 was supported, see **Figure 3B**, see also many other WM training studies mentioned in Section “Introduction”). So we claim the manipulation of WMC was successful. More important, the latency of WM training group (the individuals who have improved WMC) in antisaccade task was decreased after training (supported H2-2, see **Figure 4A**), demonstrating that improved WMC closely related with better attentional control, which is consistent with the results of Experiment 1 (i.e., high-WMC individuals have better attentional control), and this might be a more direct evidence than previous studies that did not manipulate WMC directly. Strangely, the results also showed that SA favors attentional control (did not support H2-3, see also Booth and Sharma, 2009), which seems inconsistent with the results of Experiment 1 (we will try to explain it fully in Section “General Discussion”). And the SA \times Training Group interaction (i.e., compared to the SA \times WMC interaction in Experiment 1) was not significant, which is consistent with the speculation in Experiment 1: the effects of SA and WMC on attentional control are independent with each other (this will be discussed in Section “General Discussion,” too).

Besides, results of Experiment 2 also demonstrating a far transfer effect of WM training, and it might be a considerable evidence for the debate of far transfer effect mentioned in Section “Introduction.” Because (a) we used OSPAN, which is one of the most representative WMC tasks according to Engle

et al. (1999), as a measurement of WMC. OSPAN is the most commonly used task to evaluate WMC in studies focused on controlled attention view of WMC, but we have not seen it was used in studies of WM training, so OSPAN here is a reliable measurement in general and also a novel measurement for WM training study. (b) Studies concerning WM training and attentional control relied heavily on the Stroop task to evaluate attentional control (Shipstead et al., 2012), Stroop task confounds attentional control and task performance, which is not a good measurement of attentional control, so the antisaccade task might be a better measurement of attentional control. Future studies should also employ different measurements of attentional control to provide different perspective of far transfer on attentional control.

However, the results of Experiment 2 must be concluded carefully given that the near transfer effect was trend-level ($p = 0.076$), which might be attributed to the unexpected enhancement of OSPAN score for control group (see **Figure 3B**). We suppose that was the reason of (a) familiarity effects, (b) placebo effects of training, and (c) the training task of control group: it might be easier than the training task of the WM training group (see **Figure 3A**, the apparent performance of control group increased faster than WM training group), leading to higher self efficacy, which might affect the performance of OSPAN. Besides, we did not found the correlation between amount of improvement on the trained task, and the amount of improvement on the near-transfer OSPAN task. That might be due to (a) the number of participant is too small (considering that WM training is very tough, we preferred to have a smaller sample size to ensure the effectiveness of training). For the WM training group, there is 16 participants, which might be insufficient for a correlation test, and (b) the individual difference of training sensitivity. Overall, all participants in WM training group performed better on OSPAN after training compared with before training, the training effect might be better for some participants who performed worse on

training task (even they still performed bad after all training session).

GENERAL DISCUSSION

The main purposes of present study were to examine the effects of SA and WMC on attentional control using two experiments. We manipulated SA and divided participants into low- and high-WMC group in Experiment 1, and we manipulate both SA and WMC in Experiment 2. Results shows consistent positive effects of WMC on attentional control (i.e., higher WMC means better attentional control in Experiments 1 and 2), inconsistent effects of SA on attentional control (i.e., SA impaired attentional control in Experiment 1 and favored attentional control in Experiment 2), and a consistent null-effect of SA \times WMC on attentional control (i.e., the possible interaction effect was not found). Here we attempted to explain these results in detail.

First, the consistent positive effect of WMC on attentional control replicated most of the previous studies about the controlled attention view of WMC. The unique contribution of present study is that we manipulated WMC directly rather than just divided participants into low- and high-WMC group, which provided more direct evidence to the controlled attention view of WMC. The exact mechanism of WMC favors attentional control is still unclear, attentional control and WMC might be the different representation of same psychological variable (or attentional control is part of the function of WMC), because WMC and attentional control have similar neural basis (i.e., prefrontal cortex) (Kane and Engle, 2002; van Veen and Carter, 2006), and ERP study showed that after WM training, the amplitudes of P300 and N160 increased significantly whereas that of P200 decreased (Zhao et al., 2013). The increase of P300 and N160 respectively implied stronger ability of updating (Gevins and Smith, 2000) and stronger concentration of task-relevant information (Mcevoy et al., 2001), and the decrease of P200 implied stronger inhibition of task-irrelevant information (Mcevoy et al., 2001). These effects above are highly correlated with attentional control, but this study is about WM training. Future research could attempt to divide attentional control and WMC in a physiological way. Besides, we manipulated WMC by WM training, which also provide new evidence of far transfer effect on attentional control, but the effectiveness of far transfer effect still needs more evidences from different attentional control indicators due to that there are also many negative evidences (e.g., Shipstead et al., 2012; Melby-Lervåg and Hulme, 2013; Melby-Lervåg et al., 2016).

Second, the inconsistent effects of SA on attentional control reflected similar inconsistent results as previous studies (see Introduction). We claim that different SA levels would lead to different effects of SA on attentional control: relatively high SA would impair attentional control, whereas relatively low or moderate SA would have little effect (or even benefit) on attentional control, because SA also has some positive effects such as enhancing arousal level and

motivation, which might counteract the negative effects of SA on attentional control (sometimes the positive effects of SA might stronger than negative effects). This inference could explain the inconsistent results of Experiments 1 and 2: relatively high SA impaired attentional control in Experiment 1, whereas relatively low or moderate SA favored attentional control in Experiment 2. Evidences that support this inference are (a) the brief interview after post-training test revealed that all participants felt more relaxed in post-training test than in pre-training test, because they were more familiar and confident in completing the antisaccade in post-training test; (b) More important, we compared the pre- and post-training SA manipulation data, the heart rate and MRF-3 score in post-training test were significantly lower than in pre-training test (all $ps < 0.003$). So, the inconsistent results of SA on attentional control could be explained by our novel inference, which is also the contribution of present study. Future studies should pay more attention to induce relatively high SA when exploring similar topics. A standard multi-sources SA condition should be proposed so that we could induce relatively high SA and easily compare results of different studies.

Third, we claim that the consistent null-effect of SA \times WMC on attentional control implied that the effects of SA and WMC might be independent with each other, that is, SA and WMC might affect different aspects of attentional control. We think that SA will affect stimulus-driven system, whereas WMC will affect goal-directed system. The evidence for SA affects stimulus-driven system is that Pacheco-Unguetti et al. (2010) had explored the effects of TA and SA on attentional network (orienting, alerting, and executive control), they found that TA impaired executive control (which is more like goal-directed system), whereas SA was associated with an over-functioning of the alerting and orienting (which are more like stimulus-driven system). As for the evidence for WMC affects goal-directed system is that high-WMC individuals perform better top-down attentional control such as they were better at resisting distractors (Kane et al., 2001; Unsworth et al., 2004; Unsworth and Spillers, 2010), they could amplify task-relevant information or inhibit task-irrelevant information according to task requirement (Colflesh and Conway, 2007), and they searched object (top-down) by keeping the features in their minds (Bleckley et al., 2014). Besides, the neural basis of WMC is prefrontal cortex, which is also the basis of top-down attentional control (Kane and Engle, 2002; van Veen and Carter, 2006). Future research could consider exploring this speculation about the relationship between SA and WMC.

It should be highlighted that the relationship between anxiety and attentional control predicted by Attentional Control Theory needs to be refined, because anxiety could be divided into TA and SA and the effects of SA on attentional control are complex as mentioned above. According to Pacheco-Unguetti et al. (2010), TA impairs attentional control through impairing goal-directed system, and SA impairs attentional control by favoring stimulus-driven system. This explanation is also helpful for understanding the null-effect of the SA \times WMC interaction (e.g., the present study, see also

Edwards M.S. et al., 2015; Wood et al., 2015). SA and WMC affect different aspects of attentional control, so it is more difficult to observe this interaction compared with TA \times WMC interaction (TA and WMC affect the same aspects of attentional control, e.g., Johnson and Gronlund, 2009; Wright et al., 2014). Another refinement should be considered is the SA level, that is, relatively high SA level might be necessary for observing the negative effect of SA on attentional control (already discussed above).

One shortcoming of present study is that the effects on antisaccade error rate are mostly null-effect in present study (except for the SA on error rate in Experiment 1). High-WMC individuals did not show lower error rate in Experiments 1 and 2, and SA had little effect on error rate in Experiment 2. A similar pattern was reported by Derakshan et al. (2009): they argued that error rate is more suitable to become an evaluation of antisaccade task performance rather than attentional control. It seems that error rate is probably not a sensitive enough indicator of attentional control in antisaccade task. Future studies could consider regarding error rate as a task performance indicator rather than an attentional control indicator. Besides, given that the education levels in the present study were inconsistent (i.e., we included both undergraduate and graduate students, and people who have higher education level might imply higher ability, higher WMC or attentional control), future studies could pay more attention to the education levels of participants in order to provide more reliable evidence and extend the result to people with different education levels.

Taken in sum, the present study implies a complex relationship between SA and attentional control, emphasizes the important promotion of WMC on attentional control, and denies the possible interaction of SA and WMC. In detail: (a) we found that the effect of SA on attentional control will depend on the SA level, that is, relatively high SA level might be necessary for observing the negative effect of SA on attentional control; (b) we manipulated WMC directly by WM training, and provided more reliable evidence for the importance of high-WMC on better attentional control and a new supportive evidence on far transfer effect of WM training; (c) we did not found the interaction of SA and WMC, and we speculated that the effects of SA and WMC on attentional control might be

independent with each other, that is, SA and WMC might affect different aspects of attentional control (e.g., SA will affect stimulus-driven system, whereas WMC will affect goal-directed system).

ETHICS STATEMENT

The present study was approved by Beijing Sport University Institutional Review Board (BSUIRB) (Approval Number: 2015037). Before experiment, all participants provided informed consent to confirm that they were clear about the details and possible uncomfortable feelings in the experiment. Only when participants agreed can we start experiment. Participants could drop out at anytime during the experiment, and we also have debriefing period after experiment. Vulnerable populations were not involved in the present study.

AUTHOR CONTRIBUTIONS

XL and LZ contributed to the conception and design of the work. XL contributed to the acquisition of data. XL and LZ contributed to the analysis and interpretation of data for the work. XL, LZ, and JW wrote and revised the manuscript. XL, LZ, and JW approve the final version of the manuscript. XL, LZ, and JW agrees to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

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The Effect of Diaphragmatic Breathing on Attention, Negative Affect and Stress in Healthy Adults

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A growing number of empirical studies have revealed that diaphragmatic breathing may trigger body relaxation responses and benefit both physical and mental health. However, the specific benefits of diaphragmatic breathing on mental health remain largely unknown. The present study aimed to investigate the effect of diaphragmatic breathing on cognition, affect, and cortisol responses to stress. Forty participants were randomly assigned to either a breathing intervention group (BIG) or a control group (CG). The BIG received intensive training for 20 sessions, implemented over 8 weeks, employing a real-time feedback device, and an average respiratory rate of 4 breaths/min, while the CG did not receive this treatment. All participants completed pre- and post-tests of sustained attention and affect. Additionally, pre-test and post-test salivary cortisol concentrations were determined in both groups. The findings suggested that the BIG showed a significant decrease in negative affect after intervention, compared to baseline. In the diaphragmatic breathing condition, there was a significant interaction effect of group by time on sustained attention, whereby the BIG showed significantly increased sustained attention after training, compared to baseline. There was a significant interaction effect of group and time in the diaphragmatic breathing condition on cortisol levels, whereby the BIG had a significantly lower cortisol level after training, while the CG showed no significant change in cortisol levels. In conclusion, diaphragmatic breathing could improve sustained attention, affect, and cortisol levels. This study provided evidence demonstrating the effect of diaphragmatic breathing, a mind–body practice, on mental function, from a health psychology approach, which has important implications for health promotion in healthy individuals.

Keywords: breathing technique, mental health, real-time feedback, relaxation, sustained attention

INTRODUCTION

Breathing practice, also known as “diaphragmatic breathing” or “deep breathing,” is defined as an efficient integrative body–mind training for dealing with stress and psychosomatic conditions. Diaphragmatic breathing involves contraction of the diaphragm, expansion of the belly, and deepening of inhalation and exhalation, which consequently decreases the respiration frequency

and maximizes the amount of blood gases. Benefits of diaphragmatic breathing have been investigated in association with meditation and ancient eastern religions (such as Buddhism) and martial arts (Lehrer et al., 2010). It is considered to be a core component of yoga and Tai Chi Chuan (TCC) and contributes to emotional balance and social adaptation (Sargunraj et al., 1996; Beauchaine, 2001; Porges, 2001), as well as special rhythmic movements and positions.

Psychological studies have revealed breathing practice to be an effective non-pharmacological intervention for emotion enhancement (Stromberg et al., 2015), including a reduction in anxiety, depression, and stress (Brown and Gerbarg, 2005a,b; Anju et al., 2015). A 1-day breathing exercise was found to relieve the emotional exhaustion and depersonalization induced by job burnout (Salyers et al., 2011). A 30-session intervention with a daily duration of 5 min can significantly decrease the anxiety of pregnant women experiencing preterm labor (Chang et al., 2009). In addition, similar effects on anxiety was observed in a 3-days intervention study, where breathing practices were performed 3 times per day (Yu and Song, 2010). Further evidence from a randomized controlled trial (RCT) suggested that a 7-days intensive residential yoga program that included pranayama (breathing exercises) reduced anxiety and depression in patients with chronic low back pain (Tekur et al., 2012). Supportive evidence has also come from a line of RCTs of TCC and yoga (Benson, 1996; Telles et al., 2000; Oakley and Evans, 2014). Currently, breathing practice is widely applied in clinical treatments for mental conditions, such as post-traumatic stress disorder (PTSD) (Sahar et al., 2001; Descilo et al., 2010; Goldin and Gross, 2010), motion disorders (Russell et al., 2014), phobias (Friedman and Thayer, 1998), and other stress-related emotional disorders.

Earlier studies have observed an attention/vigilance impairment related to breathing dysfunction in dementia and sleep-disordered breathing in individuals across all ages (Chervin et al., 2006). More recent studies have suggested that there is a bidirectional association between breathing and attention. A growing number of clinical studies have demonstrated that breathing-including meditation may represent a new non-pharmacological approach for improving specific aspects of attention. Mindfulness, for instance, contributes to alerting and orienting, but conflicts with monitoring. In addition, an 8-weeks mindfulness-based stress reduction yielded a larger effect than a 1-month intensive mindfulness retreat, on the attention altering component (Jha et al., 2007). Focused attention meditation is a Buddhist practice, whereby selective attention and the sensation of respiration must be sustained (Gunaratana, 1993/2002; Gyatso and Jinpa, 1995). Three months of intensive focused attention meditation have been found to reduce variability in attentional processing of target tones and to enhance attentional task performance (Lutz et al., 2009). Some studies have investigated cognitive and emotional improvement simultaneously, and have indicated that a brief mental training could enhance sustained attention as well as reduce fatigue and anxiety (Zeidan et al., 2010). Some researchers believe that the relaxation generated by peaceful breathing helped to manage inattention symptoms among children with attention deficit-hyperactivity disorder

(ADHD) (Amon and Campbell, 2008). These results led to the development of a breath-controlled biofeedback game called ChillFish, which improved children's sustained attention and relaxation levels (Sonne and Jensen, 2016).

Studies orientated toward the physiological mechanism of breathing intervention effects have indicated a shared physiological basis underlying breathing, emotion, and cognition, involving the autonomic nervous system. Physiological evidence has indicated that even a single breathing practice significantly reduces blood pressure, increases heart rate variability (HRV) (Wang et al., 2010; Lehrer and Gevirtz, 2014; Wei et al., 2016) and oxygenation (Bernardi et al., 1998), enhances pulmonary function (Shaw et al., 2010), and improves cardiorespiratory fitness and respiratory muscle strength (Shaw et al., 2010). A daily 15-min breathing training for 2 weeks significantly promoted mean forced expiratory volume in 1 s and peak expiratory flow rate (Bernardi et al., 1998). Breathing with a certain frequency and amplitude was found to relieve clinical symptoms in patients of all ages with sleep-disordered breathing (Chervin et al., 2006). Evidence from yoga practice also confirms a reduction of sympathetic and an increase of parasympathetic nervous system activity (Vempati and Telles, 2002; Raghuraj and Telles, 2003). Cardiac vagal tone is assumed to form part of the shared physiological basis of breathing and emotion. It is influenced by breathing and is also integral to vagal nerve stimulation that is closely associated with the physiological basis of emotion, including emotional regulation, psychological adaptation (Sargunraj et al., 1996; Beauchaine, 2001), emotional reactivity and expression, empathic responses, and attachment (Porges, 2001). Moreover, dysfunction of the autonomic nervous system is observed in adults with anxiety (Kawachi et al., 1995; Thayer et al., 1996; Friedman and Thayer, 1998), depression (Carney et al., 1995; Lehofer et al., 1997), PTSD (Sahar et al., 2001), panic disorder (Friedman and Thayer, 1998), and other stress-related mental and physical disorders (Benson, 1996; Becker, 2000; Bazhenova et al., 2001; Jacobs, 2001).

The shared physiological basis of attention and breathing can be detected in part in the autonomic nervous system of patients with ADHD (Beauchaine, 2001), but more evidence is provided by electroencephalographic (EEG) studies and functional magnetic resonance imaging (fMRI) studies (Lutz et al., 2004). For instance, EEG studies have suggested that regular breathing practice during yoga and meditation can increase β -activity in the left frontal, midline, and occipital brain regions (Bhatia et al., 2003; Snayder et al., 2006), which has been associated with enhanced cognitive performance, such as during attention, memory, and executive functions (Freeman et al., 1999). In addition, fMRI studies have also detected a significant increase in activation in the bilateral inferior frontal and temporal regions under meditation, as compared to a relaxation condition. Such studies implicated the right inferior frontal cortex/right insula and right middle/superior temporal cortex as the regions involved in meditation (Hernández et al., 2015).

Cortisol, a steroid hormone of the glucocorticoid class, is released in response to stress. Cortisol release is associated with depression, anxiety, and other negative emotions. The underlying mechanism may be grounded in its sensitivity for the

activity of the hypothalamic–pituitary–adrenal (HPA) axis (Clow et al., 2010), which regulates metabolism, immunity, and some mental processing, including memories and emotional appraisal (Pariante and Lightman, 2008). Plasma cortisol levels reflect changes in the activation of the HPA axis with changes in CO₂ inhalation (Argyropoulos et al., 2002), while salivary cortisol levels have been associated with fast withdrawal of attention in response to angry faces (van Honk et al., 1998). However, the associations between breathing, emotion, attention, and cortisol have not been tested together.

Although breathing practice offers an integrated benefit for mental and physical health, the results of studies on this topic are inconsistent, because of methodological limitations in the experimental design, a lack of measurable breathing feedback, and limited sample sizes. Most cross-sectional and longitudinal studies have focused on how breathing treatment benefits individuals with particular conditions, such as women during pregnancy (Schmidt et al., 2000; Booth et al., 2014) and clerks experiencing job burnout (Salyers et al., 2011), rather than on its health promotion function in a healthy population. Most importantly, most studies have investigated physiological effects, emotional benefits, and cognitive benefits separately, which prevents an understanding of the possible mental and physiological mechanisms of breathing in terms of its potential benefit for both mental and physical health.

The present study was a pilot RCT with visible feedback breathing recordings used to monitor the breathing performance overall and to evaluate the outcomes of breathing practice. The aims of this study were to investigate the mental benefits and the hormone levels in healthy volunteers who completed an 8-weeks breathing training scheme. An emotional self-reporting scale and cognitive tests were used to measure mental benefits. Additionally, cortisol a major HPA-axis-related stress hormone in humans (Matousek et al., 2010), was also measured to examine whether the breathing practice could be a buffer for modulating stress levels in the working population. We hypothesized that an 8-weeks breathing training course would significantly improve cognitive performance, and reduce negative affect (NA) and physiological stress.

MATERIALS AND METHODS

Participants

Participants were recruited from a local IT company in Beijing, China. The Institute Review Board of Beijing Normal University approved this study. This study was performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments. The procedure of the study was fully explained to the participants, and informed written consent was obtained from each participant before the study.

All participants completed the following screening forms: (1) a health approval from a recent physical check-up at a medical center and (2) a demographic questionnaire that included basic demographic information and mind–body training experience. Participants who had a history of physical health problems, such as cardiovascular or cerebrovascular diseases, respiratory

diseases, autoimmune diseases, diabetes, neuropathy, and drug or alcohol abuse, were excluded from the study. In addition, participants who had yoga, TCC, or Qigong experience, as well as other mind–body training, were excluded.

Experimental Protocol

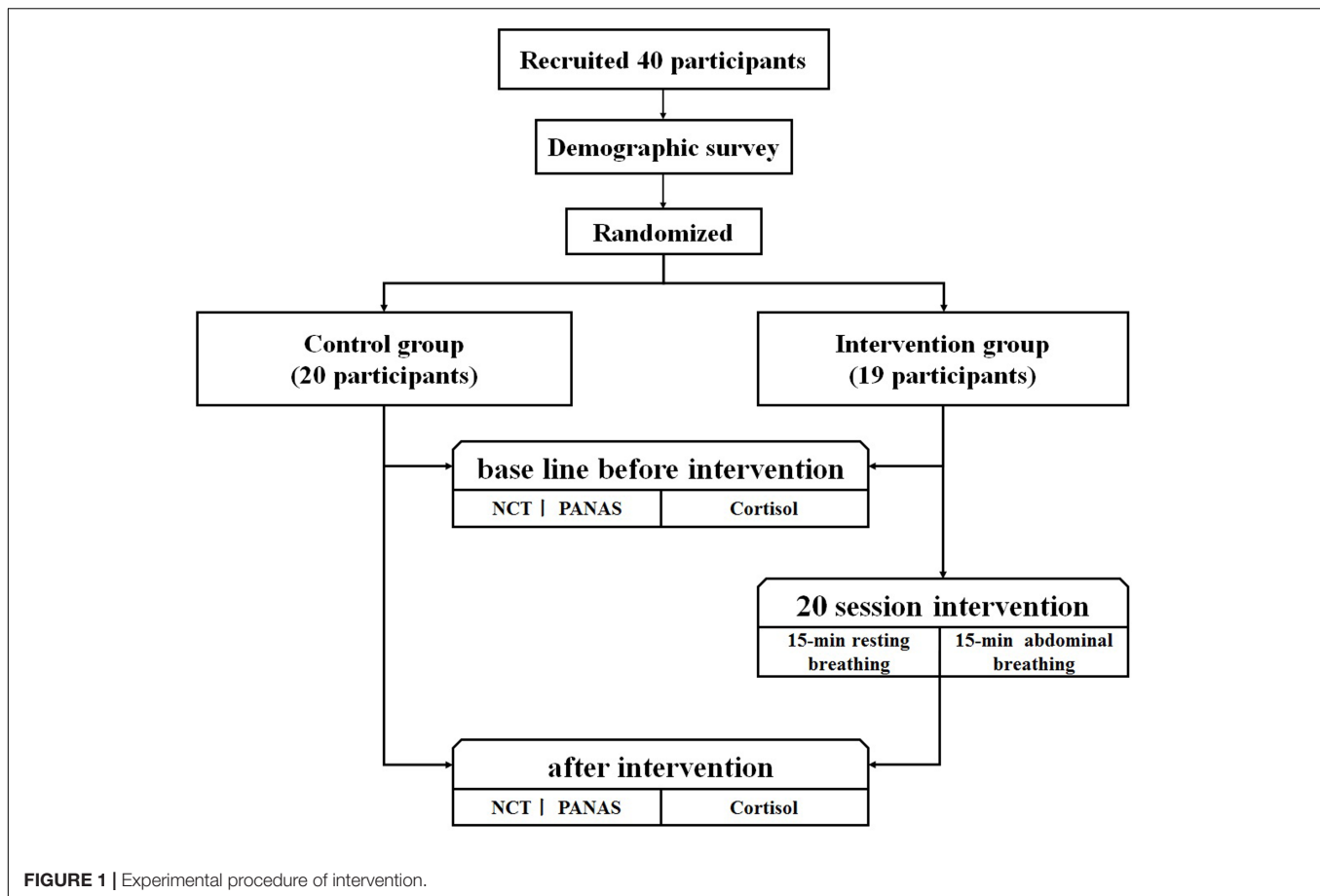
All interventions and tests were performed in a sunny, soundproof, open-air conference room at the IT company rather than in our laboratory. We reasoned that this would avoid any potential anxiety that could be brought on by rushing from the workplace to the laboratory. Participants sat comfortably in leather armchairs throughout the study.

A final total of 40 participants were included in this study. They were assigned to a breathing intervention group (BIG, 10 females and 10 males) or a control group (CG, 10 females and 10 males) by alternating the order of their registration. Gender balance was also taken into consideration during this sampling procedure.

The BIG learnt basic knowledge and essential skills about diaphragmatic breathing, and became familiar with experiencing breathing in as deeply as possible and then exhaling almost all the air from the lungs, slowly, in a self-controlled, slow rhythm, under the guide of a coach. All participants were instructed to focus on their breathing and the sensations produced in the body, while sitting comfortably in chairs with their eyes closed. Participants were considered as performing diaphragmatic breathing if their respiratory rate decreased while their respiratory amplitude increased in waveform.

After this learning phase, both groups completed the baseline tests. These included the Positive and Negative Affect Schedule (PANAS), the Number Cancellation Test (NCT), and a cortisol test. Thereafter, the BIG received 20-sessions of breath-controlling intervention. Each intervention involved a 15-min resting breathing session and a 15-min diaphragmatic breathing session consequently. The diaphragmatic breathing session began with general verbal guidance from the breathing coach, who spoke at a slow speed to help participants to become more easily involved. A final test, similar to the baseline test, was implemented at the end of the 20th intervention. In contrast, the CG received only an introduction of breathing and rest, a baseline test, and a final test, without any other intervention.

The experimental training procedure consisted of 20 sessions over a period of 8 weeks. Each session was conducted every other day on weekdays. Both groups were informed of the purpose and the procedures of this study after training. In the BIG, 20 participants were required to conduct resting breathing for 15 min and diaphragmatic breathing for 15 min in each session (Figure 1). During resting breathing, participants were instructed to breathe in a normal state. With closed eyes while sitting comfortably. During diaphragmatic breathing, they were instructed to inhale as deeply as they could while their abdomen expanded, and to exhale as slowly as they could while their abdomen contracted, in a self-paced rhythm, under the instruction of a breathing training coach and with feedback via a recording device. The two breathing conditions were recorded during the entire 30-min training. Therefore, each participant's breathing waves were visible and monitored by



the experimenter to ensure that participants were following the instructions completely. For the CG, data of resting breathing and diaphragmatic breathing from 20 participants were collected on the first day of training and on the last day of training. Before and after the entire training, the two groups completed the PANAS, NCT, and cortisol level test (Figure 1).

Breath Recording

Synchronous breathing signals were recorded using breathing monitors (Dongtuo Science and Technology Ltd., Beijing, China) that recorded participants' respiration with a high temporal resolution. All data were collected by inductive sensors (JD/PW-5; Boda Electron Co., Beijing, China), which were kept against the chest of each participant during the rest condition, or alternatively against the abdomen during diaphragmatic breathing. Data were transferred to the host computer (Lenovo, M4600 P3.0HT 25640VN) via Bluetooth and expressed on-screen as continuous visual waveforms. The height of the wave crest represented the amplitude of a single breath, while the wavelength indicated the duration. Each computer was connected to two inductive sensors; 10 recordings were made simultaneously.

All five research assistants were postgraduate students in psychology. They attended a meeting in which training was given and breath recording was practiced for approximately 1 week

prior to the formal experiment, in order to standardize the data collection procedure. Each of them monitored breathing processes for two participants concurrently and ensured that data input and output proceeded smoothly.

Throughout the breathing process, all participants kept their eyes closed and breathed through the nose. The assistants supervised their recordings and obtained the help of the coach if the recording vibrated or if the breathing frequency remained higher than 6 breaths/min by 2 min after the intervention began. The coach offered special guidance to help these participants attain diaphragmatic breathing.

Psychological Measurements

All participants were asked to complete behavioral measurements before training and after training. The behavioral measurements included the PANAS and the NCT. The PANAS (Watson et al., 1988) is a 5-point Likert scale that measures participants' feelings during the past week. It includes 20 self-reported items that are equally divided into a positive affect (PA) subscale and a NA subscale. Items are scored on a scale of 1–5, indicating very slightly or not at all, a little, moderately, quite a bit, and very much, respectively. The PANAS scale is highly internally consistent, largely uncorrelated, stable, and reliable across cultures, and can distinctively estimate PA and NA at the same time (Thompson, 2007).

The NCT includes one short practice sheet and four test sheets for evaluate attention sustainability, which is reflected by the scores generated according to the accuracy in the test. Each sheet contains 200 single digits with special symbols below or above them. The targets are “9” digits with two symbols below, above, or on either side. Participants were asked to cross out targets with a slash and ignore targets placed subsequent to a “5” as quickly as possible, within 1 min for each sheet. Participants had a 10- to 20-s interval break between two sheets. The final scores were yielded by the sum of the correct number of target digits for each sheet.

Neuroendocrine Test

Salivary cortisol was collected four times with the Salivette® Cortisol (Art. No. 51.1534) (Sarstedt AG and Co., Nümbrecht, Germany) at two time points: before and after diaphragmatic breathing at baseline, and before and after the diaphragmatic breathing at the final test. All salivary cortisol samples were processed according to the manufacturer’s instructions. Before collection, all participants were required to take a 5-min break. Each participant was asked to refrain from eating or drinking (except water) within 20 min before saliva collection. In both the BIG and CG, the saliva sample was collected between 11:00 and 12:00 to control for the variation in cortisol levels over the circadian rhythm.

Salivary cortisol was analyzed using a competitive enzyme immunoassay (ELISA, DiaMetra®, Perugia, Italy), which had a 3–10 g/mL limit of cortisol, for collections before lunch, according to specifications provided by the manufacturer DiaMetra®. An intra-assay coefficient variation (9.8%) and an inter-assay coefficient variation (15%) were calculated according to Jaedicke et al. (2012) (Validation and quality control of ELISAs for the use with human saliva samples) (Kirschbaum and Hellhammer, 1994; Ogbureke and Ogbureke, 2015).

RESULTS

Demographic Characteristics

Demographic characteristics of all the participants in each group are summarized in **Table 1**. The paired *t*-tests showed that there were no between-group differences in terms of age [*t*(38) = 1.31; *p* = 0.56, Cohen’s *d* = 0.420], years of education [*t*(38) = 1.47; *p* = 0.56, Cohen’s *d* = 0.465], or work experience [*t*(38) = 1.05; *p* = 0.22, Cohen’s *d* = 0.331]. These results indicate that the BIG and CG were well balanced in age, years of education, and work experience.

TABLE 1 | Demographic characteristics in the breathing intervention group (BIG) and control group (CG).

Characteristics	BIG (n = 19)	CG (n = 20)	<i>t</i>	<i>p</i>
Age (years)	30.16 ± 5.11	28.25 ± 3.91	1.31	0.56
Education (years)	16.21 ± 1.03	15.85 ± 0.37	1.47	0.10
Work experience (years)	7.24 ± 5.34	5.75 ± 3.31	1.05	0.22

Respiratory Rate

Descriptive statistics showed the average respiratory rate to be 4 times/min in the diaphragmatic breathing condition (*M* ± *SD* = 3.45 ± 1.86) and 17 times/min in the resting breathing condition (*M* ± *SD* = 17.51 ± 5.02) (**Table 2**).

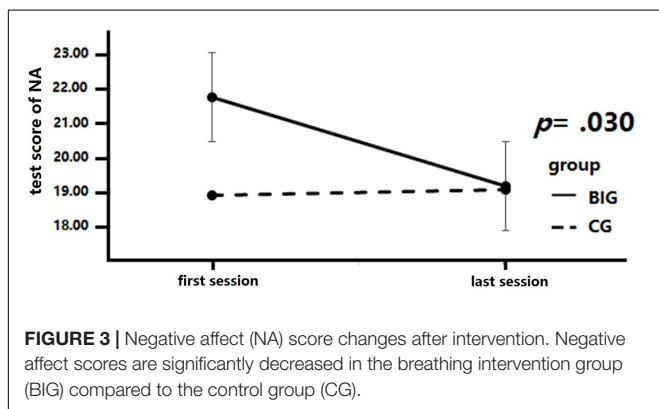
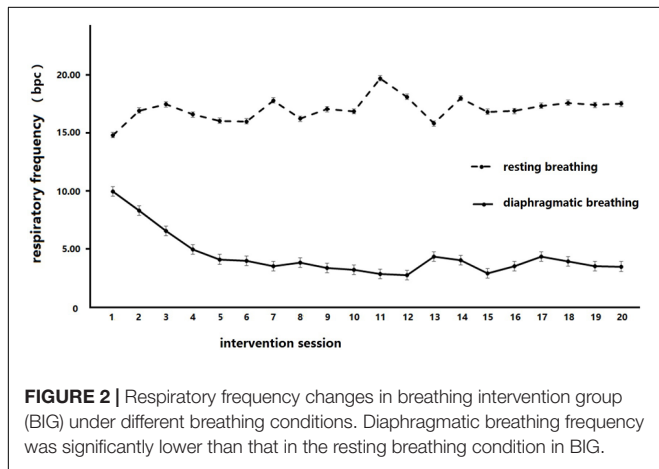
A 2 × 20 within-subject repeated measures analysis was conducted to analyze the change in respiratory rate between the resting and diaphragmatic breathing conditions across all time-points, after each of the 20 sessions, in the BIG. The within-group factors were breathing conditions (diaphragmatic vs. resting) and intervention times (20 assessments). This analysis revealed a significant effect of time, *F*(19,133) = 2.09, *p* = 0.008, η_p^2 = 0.23, and a significant effect of breathing condition, *F*(1,7) = 99.60, *p* < 0.000, η_p^2 = 0.93. There was also a significant interaction between time and condition, *F*(19,133) = 5.28, *p* < 0.000, η_p^2 = 0.43. The simple effect revealed that there were significant frequency drops (compared to time point 1) at time point 8, *MD* = 2.36, *p* = 0.027, time point 11, *MD* = 1.77, *p* = 0.018, time point 13, *MD* = 2.38, *p* = 0.019, time point 15, *MD* = 2.53, *p* = 0.006, time point 16, *MD* = 2.28, *p* = 0.014, time point 18, *MD* = 1.61, *p* = 0.033, and time point 20, *MD* = 2.23, *p* = 0.023. The simple effects on breathing shows that the respiratory frequencies in diaphragmatic breathing were significantly below that in resting breathing at every intervention time point, with the MDs ranging from 5.63 to 15.93, all *ps* < 0.002. These results indicated that the breathing intervention successfully reduced the

TABLE 2 | Respiratory rate in resting breathing and diaphragmatic breathing.

Session	Resting breathing (n = 19)		Diaphragmatic breathing (n = 19)	
	Mean	Standard deviation	Mean	Standard deviation
1	14.82	2.95	9.95	3.09
2	16.92	4.00	8.30	2.04
3	17.46	3.59	6.51	4.73
4	16.60	2.44	4.91	1.60
5	16.04	2.71	4.07	1.08
6	16.01	4.04	3.95	1.06
7	17.80	4.40	3.48	1.41
8	16.22	4.68	3.81	1.13
9	17.06	4.62	3.32	1.00
10	16.86	3.91	3.19	1.30
11	19.73	9.95	2.80	1.13
12	18.12	5.83	2.72	0.60
13	15.81	4.44	4.32	1.49
14	18.00	3.28	4.01	1.63
15	16.84	5.35	2.88	0.96
16	16.89	3.58	3.46	1.08
17	17.34	4.32	4.30	1.46
18	17.59	4.88	3.89	1.24
19	17.43	3.77	3.46	0.97
20	17.51	5.02	3.45	1.67

breathing frequency in the diaphragmatic breathing condition in the BIG.

A 2×2 mixed repeated measures analysis was conducted to analyze the change in the respiratory rate between the resting and diaphragmatic breathing conditions across time points and groups (see **Figure 2**). The within-group factor was the intervention session (baseline test and final line test), while the between factor was group (BIG vs. CG). The reduction of breathing frequency between diaphragmatic and resting conditions was employed as the measure. This analysis revealed a significant main effect of condition, $F(1,36) = 23.36$, $p = 0.000$, $\eta_p^2 = 0.39$, and an interaction between condition and groups, $F(1,36) = 7.66$, $p = 0.009$, $\eta_p^2 = 0.175$. A simple effect measurement was conducted and revealed that there was no significant between-group difference during resting breathing, $MD = 0.43$, $p = 0.861$, but diaphragmatic breathing frequency was significantly lower than that during resting breathing, $MD = 7.12$, $p = 0.000$. The respiration frequency in diaphragmatic breathing was significantly less than that in resting breathing in the BIG, $MD = 9.19$, $p = 0.000$, but no similar result was detected in the CG, $MD = 2.50$, $p = 0.153$. These results indicated that the diaphragmatic breathing intervention was effective in both the BIG and the CG, but a significant breathing frequency decrease was only observed in the BIG.



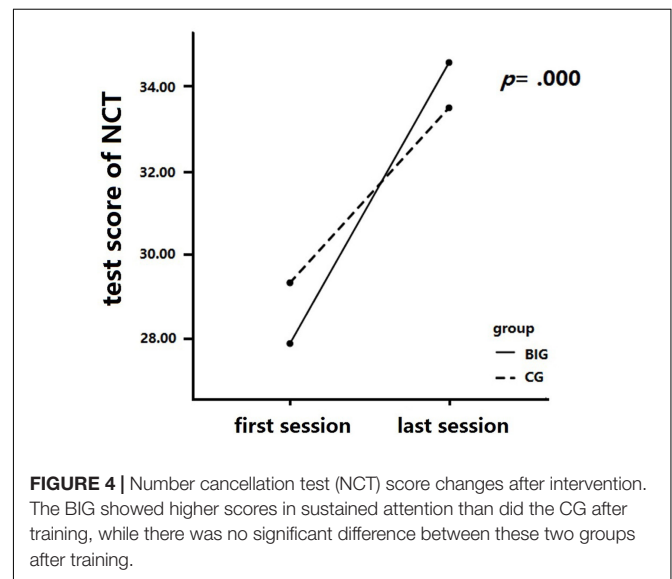
Positive and Negative Affect

A 2×2 mixed repeated measures analysis was conducted to analyze the change in NA across the intervention (see **Figure 3**). The between-group factor was group (BIG vs. CG), while the within-group factor was test time (baseline test vs. final test). Time and group revealed a marginally significant interaction, $F(1,37) = 3.43$, $p = 0.07$, $\eta_p^2 = 0.09$. A simple effect measurement was conducted and revealed that the BIG demonstrated a significant reduction in NA score after the intervention, $MD = 2.55$, $p = 0.02$, while no similar results were detected in the CG, $MD = -0.15$, $p = 0.88$. There was no significant main effect of time, $F(1,37) = 2.72$, $p = 0.11$, $\eta_p^2 = 0.07$, or of group, $F(1,37) = 0.9$, $p = 0.34$, $\eta_p^2 = 0.02$.

A 2×2 mixed repeated measures analysis was conducted to analyze the change in PA across the intervention. We measured the between-group differences (BIG vs. CG) in PA at the baseline test and final test and detected an insignificant interaction between group and test times, $F(1,37) = 0.17$, $p = 0.68$, $\eta_p^2 = 0.005$, a non-significant main effect of time, $F(1,37) = 0.96$, $p = 0.33$, $\eta_p^2 = 0.03$, and a non-significant main effect of group, $F(1,37) = 0.29$, $p = 0.60$, $\eta_p^2 = 0.008$.

Sustained Attention

A 2×2 mixed repeated measures analysis was conducted to analyze the change in the NCT score across the intervention (see **Figure 4**). We measured between-group differences (BIG vs. CG) in the NCT score change at the baseline and final tests. The NCT result revealed a significant interaction between time and group, $F(1,37) = 9.68$, $p = 0.004$, $\eta_p^2 = 0.21$. A simple effect measurement was conducted and revealed that the BIG showed a significant increase in the NCT score after the intervention, $MD = 6.728$, $p = 0.000$, and similar results were detected in the CG, $MD = 4.19$, $p = 0.000$. The main effect of time was significant, $F(1,37) = 191.48$, $p = 0.00$, $\eta_p^2 = 0.84$, so was the main effect of group, $F(1,37) = 0.01$, $p = 0.93$, $\eta_p^2 = 0.00$.

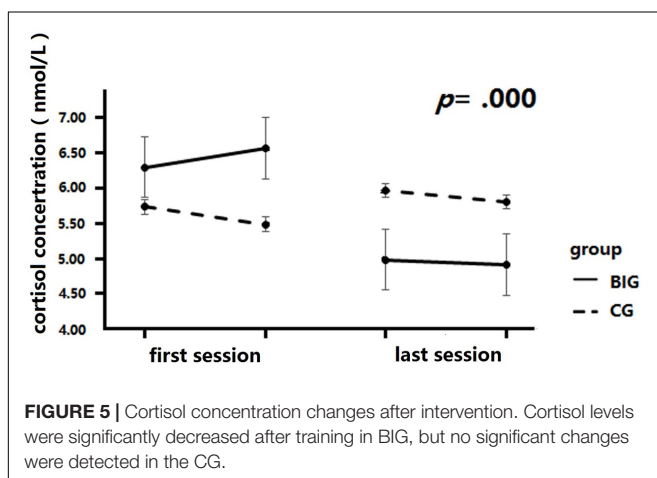


Salivary Cortisol

A 2×4 mixed repeated measures analysis was conducted to analyze the change in salivary cortisol concentration across the intervention (see **Figure 5**). The between-group factor was group (BIG vs. CG), while the within-group factor was test time (test 1, test 2, test 3, and test 4). The salivary cortisol samples were collected before and after diaphragmatic breathing for both baseline and final tests. The concentration result revealed a significant interaction of time and group, $F(3,111) = 9.06$, $p = 0.000$, $\eta_p^2 = 0.20$. A simple effect measurement revealed that the BIG showed a significant decrease in salivary cortisol concentration after the intervention, whereby the concentration was significantly lower in test 3 and test 4 as compared to test 1 and test 2, $MD_{1-3} = 1.32$, $p = 0.003$, $MD_{1-4} = 1.39$, $p = 0.002$, $MD_{2,3} = 1.59$, $p = 0.00$, $MD_{1-3} = 1.66$, $p = 0.00$. However, no similar result was found in the CG, $p > 0.05$. The main effect of time was significant, $F(1,37) = 4.17$, $p = 0.008$, $\eta_p^2 = 0.10$, but there was no significant main effect of group, $F(1,37) = 0.01$, $p = 0.92$, $\eta_p^2 = 0.00$.

DISCUSSION

Using a randomized controlled design, the present study examined whether 8 weeks of intensive diaphragmatic breathing training, a core component of mind-body practices, could influence cognition, emotion, and physiological responses. As expected, the lowered frequency of respiration after the intervention suggested that the diaphragmatic breathing intervention had been inculcated. The NA score decreased after the intervention, but the PA did not change. Sustained attention scores increased after the intervention. Moreover, a significant time effect of diaphragmatic breathing on cortisol levels was observed (before training vs. after training tests), although no group differentially influence cortisol. We interpret the findings as illustrating the potential benefits of diaphragmatic breathing practice for improving cognitive function and reducing negative affect and physiological responses to stress in healthy adults.



The Effectiveness of Breathing Practice

Numerous studies in health psychology and clinical treatment have demonstrated that diaphragmatic breathing is an effective relaxation technique in complementary and alternative medicine, with beneficial effects on physical and mental health (Cahalin et al., 2002; Tsang et al., 2015; Chen et al., 2016). However, in most studies of mind-body intervention, diaphragmatic breathing worked as a latent component or an essential preparation for the core intervention regime, such as meditation, TCC, or yoga (Telles et al., 2000; Oakley and Evans, 2014). In the present study, we monitored the breathing as an independent mind-body intervention form to discuss the health contribution to cognition, emotion, and stress response when respiration slowed. In order to achieve this aim, we adopted a unique breathing control method, which combined a monitoring device and coach supervision simultaneously.

In previous studies, practice duration and times were key characters for depicting an intervention protocol, but these seldom reported the final breathing rate and the manner in which respiration frequency decreased. In a study of slow-breathing training on chronic heart failure (Drozd et al., 2016) reported its protocol as a 10–12 weeks' (15 min per day) slow-breathing training with a breathing rate at 6 breaths/min, which meets the key requirements of diaphragmatic breathing. A clear respiration frequency provides a reliable and reproducible operation standard for estimating whether the intervention successfully decreased the respiration frequency. In the present study, the significant difference in respiration rate between the diaphragmatic breathing and resting breathing conditions confirmed that participants correctly followed the protocol for diaphragmatic breathing. The non-significant between-group difference in the respiration frequency in the resting breathing condition confirmed the distinction between the two breathing conditions. In addition, the continuous decrease in respiration rate observed in the BIG represented the process of learning and practice. All these results indicate that the change in respiratory rate could be attributed to the effectiveness of the diaphragmatic breathing practice over the 8 weeks. They also indirectly implied that the positive effects in this study were induced by intensive diaphragmatic breathing practice, rather than any confounding variables.

The present study employed a breathing monitor with results visualized on a screen and with a senior yoga coach providing professional guidance and instruction. Previous studies have demonstrated that most participants could follow self-paced breathing instructions via audio or video cues. Clinical studies usually adopt a breathing device with quantitative feedback parameters, such as respiratory rate or HRV (Sherlin et al., 2010). Respiratory sinus arrhythmia biofeedback has also been used to examine the effect of breathing practice on HRV and symptoms of PTSD, which revealed a significantly greater reduction in depressive symptoms and increases in HRV indices for individuals with respiratory sinus arrhythmia (Zucker et al., 2009). These breathing devices with feedback parameters have an advantage over visual or auditory instructions, without feedback (Brown et al., 2013), and have been widely applied in treatment for physical and mental disorders. Inviting a

senior breathing coach is an alternative solution for breathing practice and control (Drozd et al., 2016). In the present study, the coach strictly guided and monitored the entire process of diaphragmatic breathing during 20 sessions in a previous study. These two methods guaranteed that the participants breathed properly under two conditions under specific supervision during training. Having an appropriate monitor was another problem that was encountered in breathing practice studies.

Breathing Practice to Decrease Negative Affect

Emotional improvements have been reported to be the most obvious benefit of mind–body interventions (Stromberg et al., 2015). It has been suggested that the detrimental effects of stress and negative emotions could be counteracted by different forms of breathing techniques, meditation, and relaxation (Jerath et al., 2015), as well as by yoga and TCC (Benson, 1996; Telles et al., 2000; Oakley and Evans, 2014). A non-randomized study suggested that a 1-week breathing practice decreased the mean of Post-traumatic Checklist-17 (PCL-17), Beck Depression Inventory (BDI-21), General Health Questionnaire (GHQ-12) in survivors of the 2004 South-East Asia tsunami. Their results indicated an equivalent effect of breathing practice with a traumatic incident reduction exposure therapy. Moreover, the effects persisted for at least 24 weeks after the intervention had finished (Descilo et al., 2010). As a non-pharmaceutical treatment, breathing control therapy is now widely used in dealing with depression (Tsang et al., 2006), PTSD (Descilo et al., 2010), insomnia (Manjunath and Telles, 2005), and other relevant mental disorders (Brown and Gerbarg, 2005a). It is also applied as an adjuvant treatment for patients with physical disorders, including stroke (Marshall et al., 2014) and cancer (Hayama and Inoue, 2012). All these lines of evidence confirmed the efficacy of diaphragmatic breathing in clinical conditions, but we have shown its benefit for healthy individuals. A previous study has also reported that a better breathing technique was associated with greater reductions in anxiety (Sherlin et al., 2010). A 6-weeks' breathing training course was long enough to cause a significant decrease in anxiety levels in healthy adults (Chandla et al., 2013). Evidence from diaphragmatic breathing studies suggested a significant reduction in the state anxiety after an 8-weeks' intervention measured using the Beck Anxiety Inventory Assessment in adults (Chen et al., 2016), and a decrease in self-reported feelings of state anxiety and test performance in primary school students, by a pre-test/post-test, training-versus-control experiment (Khng, 2016). In the present study, our 20 sessions of diaphragmatic breathing practice significantly decreased the NA scores in the BIG. This is consistent with previous results and suggested a relief of basic NA in individuals' daily lives. Although we did not detect the time point at which the emotional benefit occurred, previous studies suggested that even a one-time intervention could reduce stress, disengaged coping (Arsenio and Loria, 2014), and could provide certain curative alleviation of job burnout, as well as other emotional disorders. In that case, it remains unknown whether the reduction of NA occurs after

the first intervention or after a significant reduction in respiration frequency. This could indicate that diaphragmatic breathing can provide an emotional improvement as a potential health care effect in healthy volunteers.

Breathing Practice to Enhance Sustained Attention

Sustained attention is critical for maintaining performance over a period of time. Deficits in sustained attention are major symptoms for several mental disorders (Winterer et al., 2000). In normal healthy participants, fatigue, work burnout, and task difficulty usually lead to poor performance in sustained attention (Aston-Jones et al., 1999). In the present study, we investigated the diaphragmatic breathing as a single intervention method for sustained attention improvement. The results suggested that 20 session's intervention provided an improvement in the NCT score. Indirect evidence supported these results, obtained from studies about breathing involving meditation (Lutz et al., 2009) and yoga training (Velikonja et al., 2010). In previous studies, both long-term intervention (MacLean et al., 2010), lasting for weeks, and short-term (Tang et al., 2007) intervention, for a few days, were effective. Consistent with previous studies, we have detected both long-term benefits after completing the intervention as a whole in the BIG and an immediate improvement in the CG. Notably, attention improvement was gained after 15 min of diaphragmatic breathing, which was markedly shorter than the 5 days' training reported in a previous study (Tang et al., 2007).

Previous studies have hypothesized that perceptual improvements (MacLean et al., 2010) and stress reduction (Jensen et al., 2012) are the mechanisms by which attentional improvement is gained. Combining the increase in the NCT score and the decrease in the NA observed in the present study, we propose that relaxation gained from diaphragmatic breathing improved the attention test performance (Amon and Campbell, 2008; Sonne and Jensen, 2016). From the point of view of neuroscience, adjusting the imbalances in the autonomic nervous system is the unique contribution provided by breathing intervention, and was directly supported by TCC research. It indicated that the HRV increased when diaphragmatic breathing was performed, which indicated an activity balance between the sympathetic and parasympathetic systems (Wei et al., 2016). Therefore, it is reasonable to infer that diaphragmatic breathing might modulate cognitive performance by predominantly exerting its influence on the autonomic nervous system. Although the neuro-mechanism remains to be clarified, it is likely that deep breathing could link mind and body together to regulate the information processing related to attention.

Breathing Practice and Physiological Responses

Cortisol is a reliable indicator of stress (Feder et al., 2009), because its concentration increases when having to cope with stressful events. We employed salivary cortisol as a measure of physiological response in the present study to estimate the neurophysiological benefit of diaphragmatic breathing. Its

concentration decreased significantly after the 20 sessions' intervention, which was consistent with previous results from parents of children and adolescents with diabetes type 1 (Tsiouli et al., 2014). This result was consistent with previous studies and indicated that breathing practice reduced the stress-related physiological response level in healthy volunteers.

Cortisol is also closely associated with the HPA axis (Clow et al., 2010), which can involuntarily control metabolism, immunity, and some mental processing, including memory and emotional appraisal (Pariante and Lightman, 2008), and can easily be affected by breathing (Argyropoulos et al., 2002). Its association with attention (van Honk et al., 1998), as well as breathing practice, cognitive processing, and emotional arousal is strong. It has been suggested that the sympatho-vagal stress response returns to an optimal balance at 4.5–5.5 bpm breathing in most adults (Lehrer et al., 2010), but no direct evidence has illuminated the potential mechanism of this physiological effect.

A hypothesis provided by Jerath et al. (2015) suggested that breathing stimulates vagal activation of GABA pathways from the prefrontal cortex and insula, to inhibit amygdala over-activity (Brown et al., 2013). A tuning function of the brain toward a parasympathetically driven mode and positive states was observed in the left insula and left orbitofrontal cortex with increasing yoga experience (Villemure et al., 2015). Evidence from brain imaging studies has supported this hypothesis. These results suggested that long and regular breathing-involving meditation practice significantly deactivated the limbic system (Kalyani et al., 2011) and rostral prefrontal cortex (Tomasino and Fabbro, 2016), and increased the activation of the right dorsolateral prefrontal cortex (DLPFC) while performing an attention-focused task. Moreover, specific changes, such as an increased cortical thickness (Wei et al., 2013), prefrontal-hippocampus functional connectivity (Tao et al., 2016), and a decreased regional homogeneity in the DLPFC (Wei et al., 2014), were also detected during the resting state among TCC senior practitioners. A structural change was proposed according to the autonomic nervous evidence, and was supported by an absence of age-related gray matter decline in senior yoga practitioners, as seen on structural MRI (Villemure et al., 2015). All these results suggested that the prefrontal cortex served as the predominant commander, supervising the activity from the limbic system, and modulating the activity of the autonomic nervous system.

Limitations

In the present study, we employed healthy volunteers as our target population. In that case, we estimated the improvement in mood and cognition and the physiological response of stress, rather than clinical symptoms, such as anxiety, depression, attentional impairment, or other stress-related pathological symptoms. In comparison to a previous study, our results investigated the application of diaphragmatic breathing as a daily health care for health population. Some of the limitations of the present study should be acknowledged. First, we discussed the diaphragmatic breathing benefits from the points of view of emotion, cognition, and physiology, respectively, rather than

by combining these three aspects together to obtain further mechanism-related results. Therefore, we have deduced a limited contribution to elucidating the underlying associations between cognitive progressing, affective improvement, and physiological change. Moreover, in terms of the neuroendocrine response, the cortisol levels were reduced after a 20 interventions, but no reference criteria were provided to demonstrate whether this decrease resulted in a real physiological benefit or was only statistically significant.

In future studies, it would be interesting to investigate the time-course between NA reduction and a sustained attention score increase. We expect that a time-course would elucidate the association between emotional and cognitive benefit, and allow testing of the hypothesis that the relaxation induced by diaphragmatic breathing relieves stress and thereby benefits cognitive possessing.

CONCLUSION

The present study illustrates the potential for diaphragmatic breathing practice to improve cognitive performance and reduce negative subjective and physiological consequences of stress in healthy adults. Despite the promise of diaphragmatic breathing practice in supporting function and health, further investigation is needed to delineate mechanisms that underlie these benefits.

ETHICS STATEMENT

The effect of diaphragmatic breathing exercise on attention, negative affect and stress level: a preliminary randomly controlled study on occupational population was approved by the ethical broad of school of psychology, Beijing Normal University. The method, experiment design, and safety of participants were strictly approved by the ethical broad of school of psychology, Beijing Normal University.

AUTHOR CONTRIBUTIONS

G-XW and Y-FL designed the work, drafted and finalized the manuscript. XM, Z-QY, HZ, N-YD, Y-TS, and Z-QG collected and analyzed the data, and revised the manuscript.

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Baduanjin Mind-Body Intervention Improves the Executive Control Function

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This study aims at comparing the effects of the Baduanjin mind-body (BMB) intervention with a conventional relaxation training program on enhancing the executive function. The study also attempts to explore the neural substrates underlying the cognitive effect of BMB intervention using near-infrared spectroscopy (NIRS) technique. Forty-two healthy college students were randomly allocated into either the Baduanjin intervention group or relaxation training (control) group. Training lasted for 8 weeks (90 min/day, 5 days/week). Each participant was administered the shortened Profile of Mood States to evaluate their mood status and the flanker task to evaluate executive function before and after training. While performing the flanker task, the NIRS data were collected from each participant. After training, individuals who have participated in BMB exercise showed a significant reduction in depressive mood compared with the same measure before the intervention. However, participants in the control group showed no such reduction. The before vs. after measurement difference in the flanker task incongruent trials was significant only for the Baduanjin intervention group. Interestingly, an increase in oxygenated hemoglobin in the left prefrontal cortex was observed during the Incongruent Trails test only after the BMB exercise intervention. These findings implicate that Baduanjin is an effective and easy-to-administering mind-body exercise for improving executive function and perhaps brain self-regulation in a young and healthy population.

Keywords: Baduanjin exercise, near-infrared spectroscopy, flanker task, mood state, executive function, left prefrontal cortex

INTRODUCTION

As an important concept of traditional Chinese medicine theories, mind-body training emphasizes the interaction between the brain, the mind, and the body (Chan et al., 2009a), with *qigong*, *tai chi*, and *yoga* being the most frequently used techniques. The fundamental assumption is that individuals can regulate breathing, heart, and body activities by their own thoughts, resulting in enhancement in physical and mental health (Cheng, 2015). A growing number of empirical studies have reported that doing mind-body exercise regularly has a positive impact on emotional and psychological processes in clinical and normal populations (Sandlund and Norlander, 2000; Chou et al., 2004; Wang et al., 2004). To take a simple example, Yoga practice was reported to lead to improvements in quality of life, psychological functioning, and symptom indices in

female cancer survivors. Interestingly, Yoga practice was associated with a linear increase in associative attention and positive affective valence (Mackenzie et al., 2014). Another randomized controlled study showed therapeutic benefits on reducing intake of antidepressants, improving depressive symptoms, and enhancing attentional abilities in patients with depression after a 10-session Chinese Chan-based Dejian mind-body intervention (DMBI) (Chan et al., 2013). Similar effects were also observed on primary school children after 4 months of DMBI (Brown et al., 1995), healthy adults after 1-month Shaolin Dan Tian Breathing (DTB) (Chan et al., 2011a), college students (Liu et al., 2008; Chen and Liu, 2013), and elderly individuals (Chen, 2013; Zhang and Ai, 2013) after 12 weeks of Eight-Brocade Exercise.

Apart from encouraging effects of mind-body training on emotional problems and psychological well-being, some empirical data have also suggested that mind-body training has a positive impact on cognitive function in clinical samples and healthy aging. In clinical practice, the DMBI helped the chronic epileptic patient enhance language, memory, attention, behavioral initiation, emotional control, and social functioning; and assisted low functioning patients with autism improve inhibitory control, cognitive flexibility, and memory functioning (Chan et al., 2009b, 2011b). A recent study (Chattha et al., 2008) reported that an 8-week integrated approach yoga therapy (IAYT) effectively elevated attention, concentration, mental balance, verbal retention, and recognition abilities in climacteric women compared with those participated in a conventional physical exercise program. For older adults, although age-related changes in cognitive function, such as declines in executive function, information processing speed, and attention are common, the benefits of mind-body exercise on these abilities in older adults are also well-documented. For instance, as a form of mind-body exercise, Tai Chi appears to help maintain executive function, language, learning and memory, and subjective memory in older adults (Miller and Taylor-Piliae, 2014).

Cognitive function refers to a person's ability to process thoughts, memory, learning new information, speech, and reading comprehension (Royall et al., 2002). Together they are key components of both "cognitive control" and "executive function." Executive function (EF) refers to the higher-order cognitive control process for the attainment of a specific goal (Moriguchi and Hiraki, 2013), and broadly encompasses a set of cognitive skills that are responsible for the planning, initiation, sequencing, and monitoring complex goal directed behavior (Royall et al., 2002). Components of executive function are measured by a variety of tests of abstraction and mental control, such as Stroop Test, Trail Making Test (TMT), Oral Reading Span Test (RST), and so on. Extensive neuroimaging studies have reported that better performance on these tests of executive functions was associated with larger prefrontal cortical volume and cortical thickness (Garavan et al., 2002; Moriguchi and Hiraki, 2013; Yasumura et al., 2014; Yuan and Raza, 2014). The unique structure and connectivity pattern of prefrontal cortex functionalized itself the only cortical region capable of integrating motivational, mnemonic, emotional, somatosensory, and external sensory information into unified, goal-directed

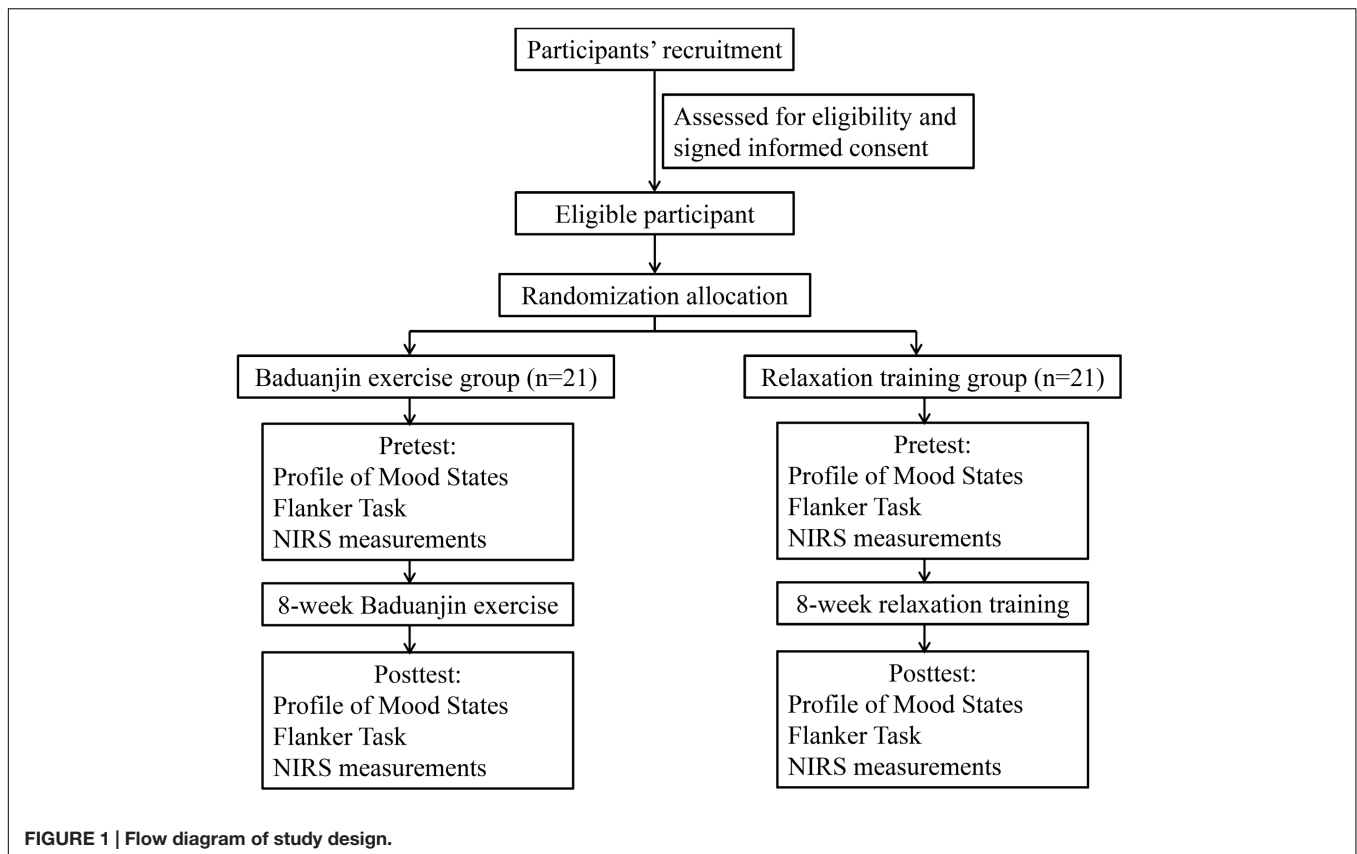
action (Royall et al., 2002). Individual's cognitive and neural development may be sensitive to physical activity (Yanagisawa et al., 2010; Davis et al., 2011; Verburch et al., 2014), but few studies have investigated the functional benefits and underlying training-induced neural plasticity contributing to the benefits.

The purpose of this study was to examine the effect of an 8-week Baduanjin mind-body (BMB), a traditional Chinese mind-body exercise, intervention on changing executive function and NIRS-measured prefrontal cortex activity in college students. NIRS is an emergent imaging technique for investigating cortical hemodynamic response. Since oxygenated hemoglobin (oxy-Hb) and deoxygenated hemoglobin (deoxy-Hb) have different absorption spectra in the infrared range, changes in oxy-Hb and deoxy-Hb can be calculated by detecting infrared light at different wavelengths on the skull. In general, enhanced oxy-Hb and reduced deoxy-Hb are associated with regional cortical activation (Yasumura et al., 2014). NIRS is noninvasive and robust against body movement, and has been validated as a suitable technique for investigating neural mechanisms in psychological experiments (Tsujii et al., 2012). Baduanjin, a form of *qigong*, can slow age-related memory decline (Wang, 2007) and has a positive effect on lowering blood pressure, blood lipid, and inflammatory factors, which are risk factors for cognitive impairment (Mei et al., 2012; Xiong et al., 2015). Zheng et al. (2016) have recently shown that the Baduanjin exercise is beneficial in maintaining or even improving both global cognitive function and specific domains of cognition including memory, processing speed, executive function, attention and verbal learning and memory in older adults with mild cognitive impairment. Consequently, we hypothesized that short-term Baduanjin training could be beneficial to executive function in normal people, and also be reflected in neural activity level.

MATERIALS AND METHODS

Participants

Forty-two right-handed undergraduate or graduate students (26 females; mean age 22.5 ± 2.0 years, range 19–26 years; body mass 52.2 ± 6.8 kg, height 165.3 ± 4.6 cm) took part in this experiment and were paid for their participation. Individuals were asked to complete a short questionnaire on their emotional and physical conditions. Individuals with any one of the following conditions were excluded from the study: (i) history of psychiatric, neurological, musculoskeletal disorders, or substance abuse; (ii) history of BMB or any other mind body training. The participants were randomly assigned into the BMB intervention group or relaxation exercise (control) group (see **Figure 1**). Paired *t*-test showed that no statistical differences were observed in any of the biographical variables indicated above between the two groups ($P_s > 0.05$). All subjects had normal or corrected-to-normal vision, and normal color vision. The Institutional Review Board at Capital University of Physical Education and Sports, where the experiments were performed, approved the study. All participants provided a written informed consent prior to their participation.



Training

The intervention group received 8 weeks of BMB exercise training under the guidance of an experienced coach, with a frequency of 5 days a week and 90 min a day including 10 min warm up, 70 min Baduanjin training and 10 min cool down. The BMB exercise consists of eight movements for limbs, body-trunk, and eye movements. These eight movements are “holding the hands high with palms up to regulate the internal organs,” “posing as an archer shooting both left and right-handed,” “holding one arm aloft to regulate the functions of the spleen and stomach,” “looking backwards to prevent sickness and strain,” “swinging the head lowering the body to relieve stress,” “moving the hands down the back and legs and touching the feet to strengthen the kidneys,” “thrusting the fists and making the eyes glare to enhance strength,” and “raising and lowering the heels to cure diseases”, respectively (Cheng, 2015). This movement set aims not only at strengthening musculoskeletal fitness (Koh, 1982) and circulation together with a qi breathing training, but also regulating emotions, representing body-mind effectiveness. On the other hand, the control group received 8 weeks of progressive muscle relaxation training with the same frequency and session duration, which helps the participants achieve physical and mental relaxation and calmness (Tang et al., 2007, 2010). Participants were taught in a standard procedure: beginning with some deep breaths with closed eyes, followed by relaxation exercises of different muscle groups over the face, head, shoulders, arms, legs, chest, back, and abdomen, guided by

an athletic trainer and compact disk. Participants were to inhale when tensing the muscles, exhale when relaxing, and concentrate on the sensation of relaxation, such as the feelings of warmth and happiness.

Measures

Each participant in the intervention and the control group was administered the baseline assessment within 2 weeks before the training, and the post-assessment within 3 days after the training. Participants were also administered the shortened Profile of Mood States (POMS) to evaluate their mood status. Executive function was measured using the flanker task. During completing the flanker task, the NIRS data were collected from each participant.

Profile of Mood States

The POMS (short version) is a checklist consisting of 40 adjectives that are rated on a scale from 0 (not at all) to 4 (extremely) according to how subjects feel. The items produce scores for seven subscales (score ranges in parentheses): Tension-Anxiety (six items), Anger-Hostility (seven items), Fatigue-Inertia (five items), Depression-Dejection (six items), Vigor-Activity (six items), Confusion-Bewilderment (five items), and Self-esteem (five items). Standard procedures were used to score the subscales of the original POMS (McNair et al., 1981). Total Mood Disturbance (TMD) scores were then computed using

the formula Depression-Dejection + Tension-Anxiety + Anger-Hostility + Fatigue-Inertia + Confusion-Bewilderment – (Vigor-Activity + Self-esteem) + 100 (Andrykowski et al., 1990; Andrykowski and Hunt, 1993). High TMD scores indicate negative affective states for all scales except vigor and self-esteem, which is a positive mood measure (Curran et al., 1995).

Flanker Task

Participants completed congruent and incongruent conditions of the flanker task (Eriksen and Eriksen, 1974). Congruent trials were those in which the target arrow was flanked by the same arrow (e.g., > > > > > or < < < < <). Incongruent trials were those in which the target arrow was flanked by the opposing response arrow (e.g., > > < > > or < < > < <). (White et al., 2011). An array of arrows were presented on a computer monitor from a distance of 1 m with visual angles of 1.7° and 3.7° in the vertical and horizontal directions, respectively. The stimuli were 7.62-cm-tall white arrows presented focally on a black background in a random order for 200 ms with an inter-stimulus interval of 1,000 ms from stimulus offset to onset.

Participants were asked to press “f” button when the central arrow in a display faced left, and “j” button when the central arrow faced right. The task was presented in a randomized block design consisting of four blocks congruent trials, four blocks of incongruent trials with 30 trials per block (a total of 240 trials). Each block lasted 30 s with a 30-s resting period separating the adjacent congruent and incongruent blocks.

NIRS Measurements

Each subject sat on a comfortable chair in a lighted room with eyes open throughout each measurement. NIRS measurements was completed immediately before and after the Eight Brocades. Changes in blood flow were measured by using a 44-channel NIRS system (ETG-4000: Hitachi Medical Corporation, Tokyo, Japan). Two equal probe sets with 3 × 5 arrays of light emitters and detectors were symmetrically placed on scalp over the prefrontal cortex. A measuring point of activation was defined as the region between one emitter and one detector. One array consisted of 22 channels and covered an area of 12 cm × 6 cm. This apparatus can measure the relative concentrations of oxy-Hb and deoxy-Hb at 44 measurement points in two areas of 15 cm × 6 cm each. The location of each shell was determined on the international 10–20 system. The most inferior medial channels of the left and right prefrontal lobes were located at Fp1 and Fp2, respectively. Concentration changes in oxy-Hb and deoxy-Hb were calculated by using the difference in absorbance based on a modified Beer-Lambert law (Shibuya-Tayoshi et al., 2007).

RESULTS

Mood-Enhancing Effect

A 2 × 2 repeated measures ANOVA was performed on TMD score before and after treatment (time) between the two groups. A significant main effect of Time was found, [$F(1,40) = 14.23$, $p < 0.01$]. The interaction between time (before or after

TABLE 1 | Total Mood Disturbance (TMD) score with standard errors before and after treatment between the two groups (intervention vs. control).

	Intervention group	Control group
Before training	117.95 ± 3.22	110.86 ± 2.32
After training	98.43 ± 2.48	112.57 ± 3.32

treatment) and group (intervention or control group) was also significant [$F(1,40) = 20.24$, $p < 0.001$]. The two groups were comparable on the level of depressive mood at baseline as measured by their TMD score [$t(40) = 1.78$, $p = 0.08$]. For each group, statistical analysis was performed using paired t -test to compare the post-training mood states with the pre-training mood states within group. Within-group comparisons revealed that TMD score decreased significantly after BMB exercise training [$t(20) = 6.45$, $p < 0.001$] but the change was not significant in the control group [$t(20) = -0.47$, $p = 0.64$] (see **Table 1**).

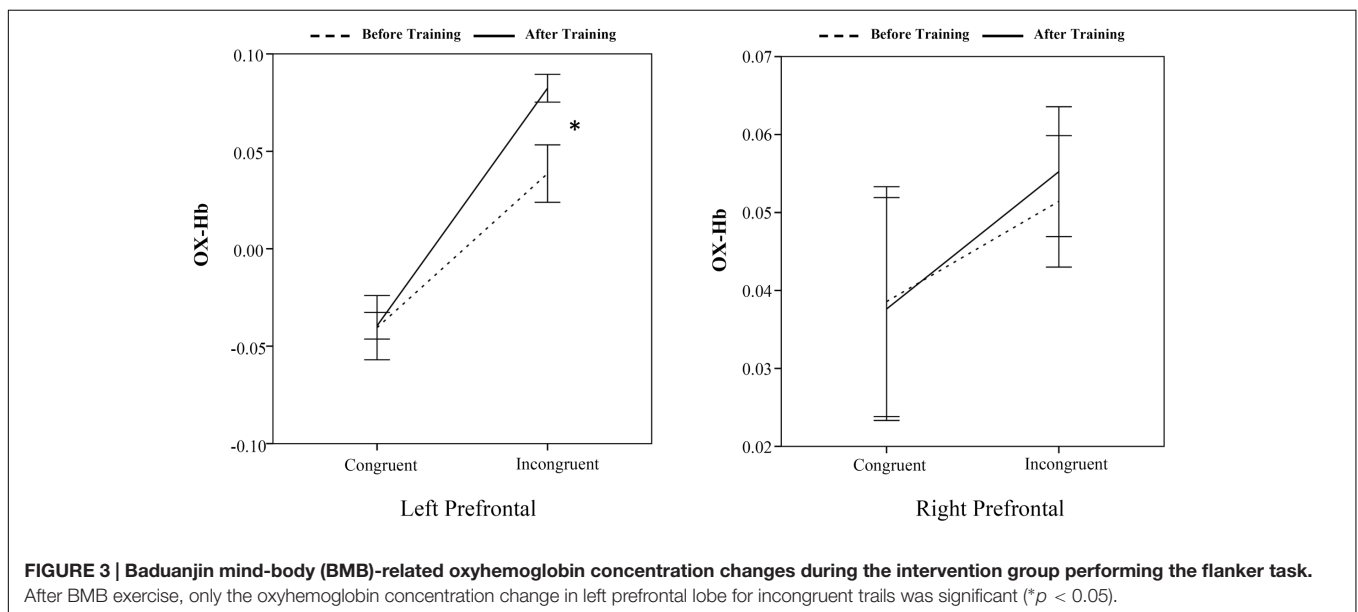
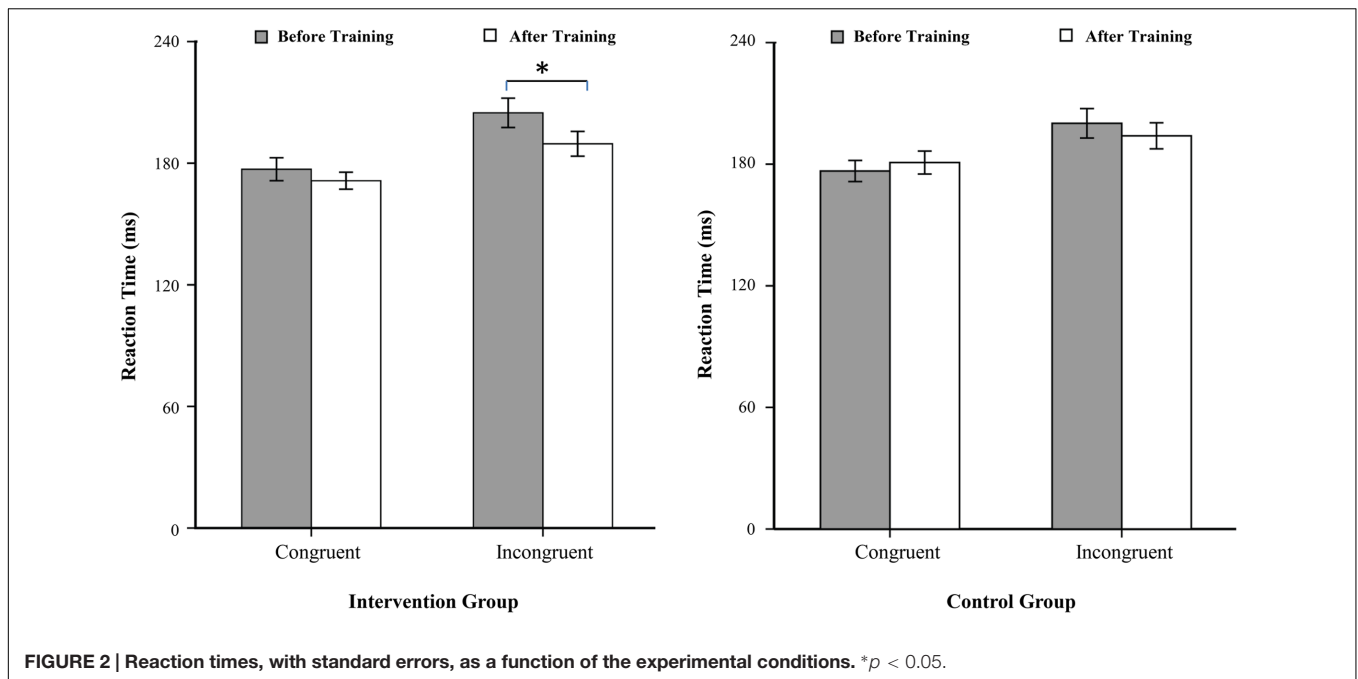
Flanker Task Performance

A 2 (time: before vs. after) × 2 (flanker congruency: congruent vs. incongruent) × 2 (group: intervention vs. control) analysis of variance (ANOVA) revealed the following significant main effects: time [$F(1,40) = 7.39$, $p < 0.01$] and flanker congruency [$F(1,40) = 108.31$, $p < 0.001$]. Importantly, the time × group and the time × flanker congruency interactions were also significant, $F(1,40) = 5.03$, $p < 0.05$, and $F(1,40) = 11.77$, $p < 0.001$. No other significant main effects or interactions were observed (**Figure 2**).

Separate ANOVAs, with the time (before vs. after) and the flanker congruency (congruent vs. incongruent) as two within-participant factors, were conducted for trials in the intervention group and for trials in the control group. For the control group, only the main effect of flanker congruency was significant [$F(1,20) = 40.94$, $p < 0.001$]. The main effect of time was not significant [$F(1,20) = 0.31$, $p = 0.59$], nor was the interaction between time and flanker congruency [$F(1,20) = 4.20$, $p = 0.055$]. Planned pairwise comparisons showed that no significant differences for reaction times were observed between the before and after measurement in the congruent trails [$t(20) = -1.51$, $p = 0.15$], as well as in the incongruent trails [$t(20) = 1.81$, $p = 0.085$]. For the intervention group, however, both the main effect of the time and the main effect of the flanker congruency were significant, $F(1,20) = 7.55$, $p < 0.05$, and $F(1,20) = 69.97$, $p < 0.001$, respectively. Importantly, the interaction between the two factors was also significant [$F(1,20) = 11.05$, $p < 0.005$]. Planned pairwise comparisons showed that reaction times did not differ in the congruent trails between the before and after training [$t(20) = 1.58$, $p = 0.13$], but they did differ in the incongruent trails between before and after training [$t(20) = 3.39$, $p < 0.005$]. Before BMB exercise training, no differences were found for executive networks in two groups ($P_s > 0.05$).

NIRS Response

A 2 (time: before vs. after) × 2 (flanker congruency: congruent vs. incongruent) × 2 (group: intervention vs. control) × 2 (frontal



lobe: left vs. right) analysis of variance (ANOVA) showed a significant main effect of time [before vs. after, $F(1,40) = 16.30$, $p < 0.001$]. The main effect of frontal lobe (left vs. right) was also significant [$F(1,40) = 80.13$, $p < 0.001$]. More importantly, the time \times group, frontal lobe \times group, and time \times frontal lobe \times group interactions were significant, [$F(1,40) = 15.44$, $p < 0.001$, $F(1,40) = 38.53$, $p < 0.001$, and $F(1,40) = 9.70$, $p < 0.005$]. No other main effects or interactions reached significance. Pairwise comparisons, with Bonferroni correction, were conducted. The increased oxy-Hb at the left prefrontal cortex was found only for the incongruent trails after the 8 weeks of BMB exercise [$t(20) = -2.42$, $p < 0.05$]. No other cross

(before vs. after)-training differences for the incongruent trails were found ($P_s > 0.05$) (Figure 3 for the intervention group and Figure 4 for the control group) (see Table 2).

DISCUSSION

This study revealed that BMD exercise may have a significant mood-enhancing effect on college students even with just 8 weeks of training. Specifically, individuals who participated in the BMB exercise showed significant reduction in depressive mood compared with those who participated in the relaxation exercise

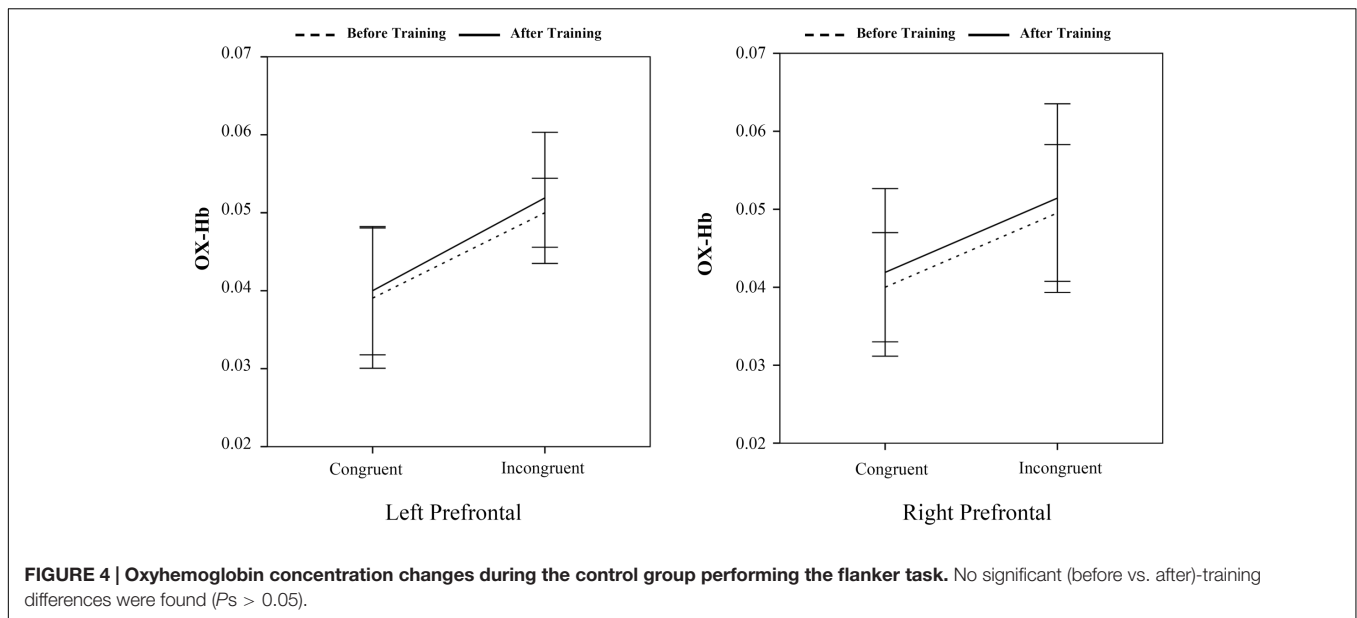


TABLE 2 | Statistical results of oxyhemoglobin concentration changes in prefrontal regions before and after treatment between the two groups (intervention vs. control), with standard errors.

	Intervention group				Control group			
	Left prefrontal cortex		Right prefrontal cortex		Left prefrontal cortex		Right prefrontal cortex	
	Congruent	Incongruent	Congruent	Incongruent	Congruent	Incongruent	Congruent	Incongruent
Before training	-0.041 ± 0.016	0.038 ± 0.014	0.038 ± 0.014	0.051 ± 0.008	0.039 ± 0.008	0.050 ± 0.004	0.040 ± 0.007	0.049 ± 0.008
After training	-0.039 ± 0.007	0.082 ± 0.007	0.037 ± 0.014	0.055 ± 0.008	0.040 ± 0.008	0.052 ± 0.008	0.041 ± 0.011	0.051 ± 0.012

training (control group). With a short-term intervention, the BMB exercise seemed to be more effective than a relaxation exercise program in improving executive control of college students. This was manifested by the observation that the before vs. after measurement difference in the incongruent trials was significant only for the group trained by the BMB exercise. In addition, the NIRs measures provided insights into the possible neural mechanism that may be associated with the improvement of executive functions. That is, the increased oxy-Hb in the left prefrontal cortex was found for the incongruent trials after 8 weeks of BMB exercise training.

There have been scientific and clinical studies which demonstrated positive effects of mind-body exercise on mood states in clinical and healthy populations (Liu et al., 2008; Chan et al., 2011a; Chen, 2013; Chen and Liu, 2013; Zhang and Ai, 2013). Consistent with these previous studies, no significant difference in mood was detected before training between the two groups. After training, however, the BMB exercise group showed significantly lower TMD scores in negative affect in comparison with the control group. These results suggest that a relatively short-term BMB exercise training can induce higher positive mood and lower negative mood states than a relaxation training. An important component of traditional Chinese *qigong*, *Baduanjin* exercise can help practitioners to reach coordination

between mind and body. When one can internally change his/her own thoughts and behaviors in a coordinated manner, enhancements in emotional and physical health are achieved (Cheng, 2015).

Executive function is a complex concept that includes working memory, overlaps with attention, and requires sensory selection, response selection, and vigilance (Miller and Taylor-Piliae, 2014). Previous studies that examined effects of mind-body interventions on cognitive function in community-dwelling older adults measured components of executive function using a variety of tests that included the Clock Drawing Test (CDT), Color Trails Test (CTT), Digit Span Tests (DS), Digit Symbol Tests (DSym), Color-Word Matching Stroop task (ST), and TMTs (Miller and Taylor-Piliae, 2014). These studies paid attention both to improving clinic treatments, and preventing cognitive impairment (World Health Organization and Calouste Gulbenkian Foundation, 2014) by means of creative measures, but not by focusing on how to enhance the executive function in healthy adults. In addition, in clinical populations, executive functions were often measured using the Functional Independence Measure (FIM), Functional Status Rating System (FSRS) (Chan et al., 2009b), and Behavior Rating Inventory of Executive Function (BRIEF) (Chan et al., 2011b). Self-reports of behaviors and attitudes are

strongly influenced by features of the research instrument, the data of which may weaken the results. Therefore, the present study used the flanker task as the standardized tool to explore the effects of BMB exercise on executive function in college students. Findings from the present study support the notion that the BMB exercise is an effective practice for improving executive function in the young and healthy population, indicated by a significant improvement in behavioral responses to the incongruent trials only in the BMB intervention group but not in the control group.

Extensive neuroimaging studies have reported that better performance on the tests of executive functions was associated with larger prefrontal cortical volume and cortical thickness (Garavan et al., 2002; Moriguchi and Hiraki, 2013; Yasumura et al., 2014; Yuan and Raza, 2014). Furthermore, Davis et al. (2011) found that aerobic exercise intervention contributed to a specific improvement on executive function and activity in prefrontal cortex circuitry in children. Similar findings were also observed in college students. Yanagisawa et al. (2010) suggested that the left dorsolateral prefrontal cortex is likely to be the neural substrate for the improved Stroop performance elicited by an acute bout of moderate exercise. In a recent study, participants exhibited a significant leftward shift of resting prefrontal activation asymmetry after receiving a mind-body exercise treatment (Yasumura et al., 2014). Therefore, besides studying the effect of the BMB intervention on the mood and executive function, we adopted the NIRS measure during the flanker task to investigate the prefrontal activation asymmetry during the congruent and incongruent trial performance induced by an 8-week BMB exercise intervention. Consistent with those previous studies, we found that the BMB training is beneficial for rectifying negative mood and improving executive function, and increasing the left prefrontal cortex activation for the incongruent flanker task, providing some evidence for neural substrate of the improved cognitive performance after a mind-body exercise.

We speculated that mind-body exercise improves executive function via effects on brain systems that underlie cognition and behavior. Executive functions are high-level cognitive functions that subservise and are a prerequisite for self-regulation (Barkley, 2001; Hofmann et al., 2012). Cognitive

neurosciences have shown that functioning of self-regulation and executive functions are both strongly but not exclusively dependent on the white matter integrity in the prefrontal cortex (Alvarez and Emory, 2006; Collette et al., 2006). As suggested by Marks et al. (2007), higher levels of aerobic fitness are associated with greater white matter integrity in prefrontal brain areas. The BMB exercise is an easy-to-do aerobic exercise that can improve executive function and brain self-regulation following a period of daily training. Thus, the greater left prefrontal cortex activation during the incongruent flanker task observed in this study may be due to that the mental process of the BMB exercise involves attention and self-regulation (or self-control of cognitive and emotional processes) as core cultivation, and the executive function shares with the brain circuits of self-regulation, mainly in the left prefrontal cortex.

A potential limitation of our study is that participants and exercise coaches cannot be blinded, possibly leading to the performance bias. In addition, the majority of participants recruited in the study were in a narrow age range (between 19 and 26 years), and were relatively well-educated and strong adaptability. Interpretation of the results should be made in light of this. Therefore, it will be very important that future studies are needed to replicate and extend the findings by adopting a larger sample from a wider age range and educational level.

AUTHOR CONTRIBUTIONS

CJ and GY designed experiments. TC and YT carried out experiments. TC and GY analyzed experimental results. TC analyzed experimental data and developed analysis tools. TC and CJ wrote the manuscript.

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Cardiorespiratory Fitness Is Associated with Executive Control in Late-Middle-Aged Adults: An Event-Related (De) Synchronization (ERD/ERS) Study

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The present study sought to determine whether cardiorespiratory fitness is associated with cognitive function in late-middle-aged adults from event-related desynchronization (ERD) and event-related synchronization (ERS) perspectives. Late-middle-aged adults were categorized into either the high-fitness group or the low-fitness group based on their estimated cardiorespiratory fitness values. The participants completed the Stroop Test, which is comprised of incongruent and neutral conditions, while the brain activities were recorded. The alpha ERD and ERS values based on the equation proposed by Pfurtscheller (1977) were further calculated. The results revealed that the adults with higher levels of cardiorespiratory fitness demonstrated superior Stroop performance, regardless of Stroop congruency. While these high-fitness adults had less positive upper alpha ERD values in the later epoch window compared to their lower-fitness counterparts, they had greater lower alpha ERD values in the early epoch window. Additionally, in the late epoch window, the high-fitness adults showed less positive lower alpha ERD values on neutral, but not incongruent condition, relative to their low-fitness counterparts. These findings suggest that cardiorespiratory fitness of the late-middle-aged adults is positively associated with cognitive functioning, especially the cognitive processes related to the inhibition of task-irrelevant information and those processes required the devotion of greater amounts of attentional resources to a given task.

Keywords: executive control, ERD, ERS, fitness, Stroop Test

INTRODUCTION

As life expectancies have continued to grow, the size of the aged population has surged dramatically over the last several decades. As of 2010, 8% of the world's entire population was over 60 years of age, and it is estimated that that number will rise to ~21% by 2050 (United Nations. Department of Economic Social Affairs: Population Division, 2013). This rapid projected rate of population aging will bring increased susceptibility to non-communicable diseases (U.S. Department of Health and Human Services, 2010). Age-related impairments have also contributed to a wide variety of cognitive deterioration, such as diminished processing speed, reduced working memory, and

poorer long-term memory (Park and Reuter-Lorenz, 2009). These aging-related disturbances lead to increased healthcare resource utilization, and as a result, there is increasing interest in exploring cost-effective strategies to deal with the relative issues (Williams and Kemper, 2010).

Fortunately, age-related cognitive decline is not entirely unavoidable. Higher cardiorespiratory fitness has been found to protect against such functional impairments in older populations (Angevaren et al., 2008; Boucard et al., 2012). Specifically, older adults with higher cardiorespiratory fitness have demonstrated better behavioral performances in terms of multiple cognitive tasks than those with lower fitness (Kramer et al., 2005; Prakash et al., 2011). The linkage between cardiorespiratory fitness and cognitive performance may involve a causal association, with enhanced cardiorespiratory fitness following exercise interventions having been found to lead to a variety of physiological structural changes that are related to cognitive functioning in late-middle-aged adults, suggesting that cardiorespiratory fitness is associated with enhanced cerebrovascular reserves for cognitive functioning (Angevaren et al., 2008; Erickson et al., 2008; Voss et al., 2013).

Interestingly, cardiorespiratory fitness is associated multiple aspect of cognitive processes, particularly cognitive processes involved executive control (Colcombe and Kramer, 2003; Colcombe et al., 2004). Executive control can be conceptualized as higher-order cognitive processes composed of a subset of processes, such as inhibition, planning, and switching (Miyake et al., 2000; Jurado and Rosselli, 2007). After analyzing 18 controlled clinical studies, Colcombe and Kramer (2003) reported that fitness training leads to improvements in all aspects of cognitive functioning in older adults; however, they found that tasks requiring executive control were more substantially improved ($ES = 0.68$) than those involving speed, visuospatial-awareness, and controlled cognitive functions. Colcombe et al. (2004) further examined the association between cardiorespiratory fitness levels, executive control, and volumes of prefrontal and parietal cortices through magnetic resonance imaging (MRI) approach. They observed that individuals with higher levels of cardiorespiratory fitness exhibited superior behavioral performance, in addition to exhibiting higher density in the brain regions such as the prefrontal and the parietal cortices involved in executive control (Colcombe et al., 2004). Similarly, another study employing functional MRI found that increased cardiorespiratory fitness following a 12-month exercise intervention was associated with improved executive control and more efficient brain functioning in the prefrontal, parietal, and sensorimotor cortical areas (Voelcker-Rehage et al., 2011). These findings suggest, from behavioral and neuroimaging perspectives, that cardiorespiratory fitness serves a protective function with regard to multiple aspects of age-related cognitive decline, including executive control.

Electroencephalography (EEG) has also been employed to explore cognitive functioning because it is noninvasive and can help shed light on the potential neural mechanisms underlying the executive control processes. EEG is believed to reflect spontaneous electrical activity in the brain that consists of diversified dynamic waveforms (Tong and

Thakor, 2009). Among the brain activities measured by EEG, much attention has been attracted by alpha frequency, an oscillation ranging between 8 and 14 Hz that are associated with sensorimotor, psycho-emotional, memory, and attentional control (Basar and Guntekin, 2012; Bazanova and Vernon, 2014). Such alpha frequency activity has been linked to the underlying neuronal activities, and it has been suggested that alpha frequency activity is inversely associated with the level of cortical activity (Bazanova and Vernon, 2014).

Nonetheless, EEG studies examining both cardiorespiratory fitness and cognitive functioning have only been limited and indirect. Gutmann et al. (2015) examined whether both acute and chronic exercise affect resting state alpha peak frequency (iAPF) in healthy young adults. Their findings indicated that iAPF values were increased after acute exercise but not after 4 weeks of cycling exercise training. Hogan et al. (2013) evaluated the cognitive performance of high-fit and low-fit adolescents after the cessation of acute exercise by utilizing the EEG coherence analysis. Their findings revealed that both the behavioral and EEG coherence assessed after the cessation of acute exercise were moderated by the participants' fitness levels. Specifically, while the adolescents with higher fitness levels demonstrated faster response time after the exercise than after the rest condition, the adolescents demonstrated lower levels of alpha coherence in the resting condition for tasks requiring executive control, suggesting increased efficacy of the attentional system after the acute exercise for those with higher cardiorespiratory fitness.

Event-related changes in alpha frequency activity (ER%) have also attracted interest, with well-known measures including both event-related desynchronization (ERD) and event-related synchronization (ERS), i.e., the suppression or expression activity in the alpha frequency triggered by stimuli or events, which reflect decreases or increases, respectively, in the harmonization of a given neuronal population (Pfurtscheller and Lopes Da Silva, 1999). ERD and ERS have also been suggested to reflect, respectively, the release from inhibition and inhibition (Bazanova and Vernon, 2014). To the best of our knowledge, only one study has utilized ER% to explore the association between exercise, fitness, and cognitive performance in older adults (Chang et al., 2015), finding that older adults with higher fitness levels benefited more from acute exercise. That is, those with higher fitness levels demonstrated significantly faster response speed after the cessation of acute exercise than their counterparts with lower fitness levels. Interestingly, a greater alpha ERD value after acute exercise compared to the control condition was also observed, suggesting that increases in neural resources for attentional investment and top-down processes were induced by acute exercise.

Accordingly, higher cardiorespiratory fitness has been linked to executive control in older adults, with substantial evidence from both behavioral and neuroimaging perspectives having been established. Despite the fact that event-related brain oscillations provide efficient and higher time resolution in terms of investigating cognitive processes, no research involving the use of ER% value has specifically explored how cardiorespiratory

fitness affects the cognitive functions in a late-middle-aged population. As such, the purpose of the current study was designed to examine the association between cardiorespiratory fitness and cognitive functioning as indicated by the alpha ER%. Specifically, cognitive functioning, as assessed by the Stroop Test that has been considered as susceptible measure for cognitive aging (Lague-Beauvais et al., 2013; Gauthier et al., 2015), cardiorespiratory fitness (Prakash et al., 2011; Gauthier et al., 2015), as well as neurophysiological indexes such as alpha ERD and ERS, were utilized. With respect to task performance, it would be expected that cardiorespiratory fitness might moderate the Stroop Test performance, such that late-middle-aged adults with higher cardiorespiratory fitness levels would demonstrate shorter response time or higher accuracy of the Stroop Test. With respect to neurocognitive processes indexed by the alpha ER%, it was predicted that late-middle-aged adults with higher cardiorespiratory fitness levels would demonstrated an increased alpha ERD or decreased ERS value compared to those with lower cardiorespiratory fitness levels.

METHODS

Participants

Healthy male adults with ages ranging from 50 to 65 years old were recruited through flyers posted in a community center and invited to a laboratory located at National Taiwan Sport University, Taoyuan County, Taiwan. Potential participants were included if they met the following criteria: (a) right-hand dominant; (b) free from neurological disease, cerebrovascular disease, or psychoactive medication use; (c) normal or corrected-to-normal vision based on the minimal 20/20 standard; (d) no color blindness; (e) free of recreational smoking habit; (f) intact cognitive functioning, as assessed by the Mini-Mental State Examination (MMSE, scores > 24); and (g) able to perform exercise of moderate intensity without any potential risk, as assessed by the Physical Activity Readiness Questionnaire (PAR-Q). Each participant's physical activity level and working memory functioning were measured by, respectively, the International Physical Activity Questionnaire (IPAQ; Bauman et al., 2009) and the Digit Span test of the Wechsler Adult Intelligence Scale-Third Edition (Wechsler, 1997).

Each participant was then instructed to complete the submaximal cardiorespiratory fitness assessment and was categorized into either a high-fitness or low-fitness group based on his/her estimated maximal oxygen consumption (VO_{2max}) value (i.e., higher or lower than the 40th percentile of the normative value of VO_{2max} for his/her age group; American College of Sports Medicine, 2013). Forty eligible participants ($n = 20$ for each group) were instructed individually to come to the laboratory again within a week, at which time they completed the behavioral and neuroelectric measures. The participants were each financially reimbursed \$30 for their participation. All the participants provided informed consent, with the study itself having been approved by the Institutional Review Board of the National Taiwan Sport University. The participants' demographic and cardiorespiratory fitness characteristics data are presented in **Table 1**.

TABLE 1 | Demographic and cardiorespiratory fitness characteristics of the high-fitness and low-fitness groups (means ± SD).

Variables	High-fitness ($n = 20$)	Low-fitness ($n = 20$)
Age (years)	60.20 ± 4.07	58.70 ± 3.53
Height (cm)	159.32 ± 0.12	161.12 ± 0.10
Weight	60.43 ± 5.58	65.33 ± 6.45
BMI	23.74 ± 1.65	25.24 ± 3.12
Digit Span test		
Forward	11.50 ± 1.79	10.60 ± 2.11
Backward	5.60 ± 1.60	5.50 ± 0.95
VO_{2max} (ml/kg/min)	49.50 ± 10.66 [*]	27.36 ± 4.50
IPAQ (MET/week)	3640.00 ± 2021.59 [*]	1638.00 ± 2504.32

BMI, body mass index; MMSE, Mini-Mental State Examination; VO_{2max} , Estimated VO_{2max} ; IPAQ, International Physical Activity Questionnaire (MET). ^{*}significant differences between groups, $p < 0.05$.

Cardiorespiratory Fitness Measure

Each participant's cardiorespiratory fitness level, as indicated by the VO_{2max} measured in milliliters per kilogram of body-mass per minute (ml/kg/min), was estimated using the YMCA cycling ergometer protocol. The YMCA cycling ergometer protocol is a widely used submaximal cardiorespiratory fitness assessment for adults with Class A risk stratification (American College of Sports Medicine, 2013). The fitness assessment was conducted during the first visit to the laboratory, with the participants having been asked not to perform any strenuous exercise 24h prior to the assessment. The given participant's heart rate was measured by a Polar Heart rate (HR) monitor fitted prior to the fitness assessment and was recorded at every 3 min throughout the fitness assessment.

The YMCA cycling ergometer protocol involves three or more 3-min stages of continuous cycling and is designed to increase the HR of participants to between 110 bpm and an HR that corresponds to 85% of the age-predicted maximal heart rate (i.e., 220 minus age). Briefly, participants were instructed to exercise at a pedaling rate and workload of 50 rpm and 25 W, respectively, in the initial stage. The HR during the last minute of the initial stage determines the workloads of succeeding stages. For instance, if a participant's HR in the last minute of the initial stage is between 80 and 89 beats/min, the workloads for second and third stages will be 100 and 125 W, respectively. The HR for the last 15 s of min 2 and min 3 at each stage were recorded. If these two recorded HRs differed by more than 5 bpm, an extra minute would be added until the participant's HR stabilized. The process was terminated when a participant's HR reached the steady-state, which was defined as being within 10 beats of 85% of the HR_{max} . Two HRs recorded from the second to last stage and from the last stage, along with the participant's body mass, age-predicted maximum heart rate, and workloads from the last two stages, were employed to calculate the estimated VO_{2max} (American College of Sports Medicine, 2013). Additionally, the Borg Rating of Perceived Exertion (RPE) on a scale of 6–20 (Borg, 1982) was recorded every 3 min.

Stroop Test

The item-by-item computerized Chinese version of the Stroop Test conducted in the current study was built and recorded using the Stim² software of the NeuroScan system (Neurosoft Labs, Inc. Sterling VA, USA). The Stroop task using Chinese characters has consistently induced a robust Stroop effect and has been utilized across a wide range of age groups (Liu, 2007; Wang, 2011; Wang et al., 2014). Two types of Stroop conditions were involved: the neutral and incongruent conditions. In the neutral condition, the stimuli consisted of colored squares printed in blue, red, or green color. The stimuli in the incongruent condition, on the other hand, consisted of three Chinese color-words [i.e., “紅”(red), “藍”(blue), and “綠”(green)] written in either of the two colors that did not match the semantic meaning of the given stimulus (e.g., the word “red” was printed in blue or green color). The stimuli proportions for each color of the colored square in the neutral condition were the same (e.g., 20 trials for each color of square), and so were the stimuli proportions for each possible color-word combination in the incongruent condition (e.g., 10 trials for each color-word combination). The stimuli in each block were presented in a randomized order.

All the stimuli were 2 cm in size and were presented in the center of white background displayed on a 15-inch computer screen with horizontal and vertical angles of 28.14° and 1.4°, respectively. Each stimulus was preceded by a fixation cross displayed in the center of the screen for 500 ms, after which the stimulus itself was shown for a maximum presentation duration of 500 ms.

Each participant practiced 12 trials prior to the formal experiment (i.e., 6 practice trials for the neutral condition and 6 practice trials for incongruent condition), having been instructed to respond, as quickly and accurately as possible, according to the chromatic nature of the stimuli by pressing, with the thumb of the left or right hand, one of three pre-specified buttons on the response box corresponding to each color. A response was accepted if it was made between 200 and 1000 ms after the onset of the stimulus. The response time (RT), defined as the time interval between the onset of a stimulus and a correct response made by the participant, and accuracy were subjected to further analysis.

EEG Recording and Processing

Participants were instructed to be seated comfortably in a chair at a distance of ~65 cm from a 14" computer screen in a sound and light-attenuated electrically shielded chamber. Throughout the recording of the EEG acquisition period, participants were instructed to keep their bodies as relaxed as possible. The EEG was recorded using the NeuroScan 4.5 system (NeuroScan Inc., El Paso, TX, USA). Prior to the administration of the Stroop Test, an electric cap (Neuroscan Quick-Cap, Neuroscan in. VA) with 32 Ag/AgCl electrodes arranged in the International 10–10 system (Chatrian, 1985) was mounted. Each electrode was referenced to the linked electrodes at the right and left mastoids with ground at FPz. The vertical and horizontal electro-oculographic (EOG) signals were simultaneously recorded from four electrodes placed below and above the left orbit and 1 cm lateral to the outer canthus of each eye, for the purpose of offline correction of ocular artifacts. For optimum signal transduction during the

recording process, it is essential that a low resistance current path between the skull skin surface and a given electrode is provided. By filling the electrodes with the Electro-GelTM (Electro-Cap International, Inc., Eaton, USA) and constantly monitoring to ensure maintenance of the impedance value during the recording process, the impedance of all of the electrodes was maintained below 5 k Ω . The analog signal of the EEG was converted to a digital signal, amplified at a sampling rate of 1000 Hz (12 dB/octave), digitally low-pass filtered with 70 Hz and digitally high-pass filtered with 0.05 Hz (60-Hz note filter) by the NeuroScan SymAmp² amplifier system (NeuroScan Inc., El Paso, TX, USA), and then stored to a hard disk on a continuous basis for off-line analysis.

The offline EEG data was further segmented into epochs ranging from –500 to 1000 ms relative to the onset of the stimulus using Editor 4.5 (NeuroScan Inc., El Paso, TX, USA) software. The trials with correct responses, but not those with incorrect responses or missing responses, were subjected to further analysis. After the data were recalculated based on the entire sweep serving as the baseline, as well as the EOG correction to remove the eye movement artifacts and eye blink activities, any trials for which exceeded $\pm 100 \mu\text{V}$ were automatically rejected. In order to reduce leakage, a Hamming window of 10% length was also utilized. Following the procedure conducted by Cooper et al. (2013), two bands of event-related EEG activities, namely, the upper alpha (11–13 Hz) and lower alpha (8–10 Hz) bands, were computed using the Event-Related Bandpower transform in Neuroscan Edit 4.5. The Event-Related Bandpower transform permits intricate demodulation and simultaneous bandpass filtering (zero phase filter, 24 dB/Oct roll-off, envelope computed). Filter warmup artifacts at each end were trimmed by 300 ms from each end of an epoch.

The percent change in alpha frequency (ER%) was computed based on the equation proposed by Pfurtscheller (1977): $\text{ER}\% = (B-E)/B \times 100$, where the B denotes the mean alpha power (i.e., of upper alpha or lower alpha frequencies) during the reference interval (i.e., the time period 200 ms prior to the onset of a cue) and E denotes the mean alpha power during the period 0–700 ms after the onset of a cue. Positive and negative values for the ER% indicate, respectively, ERD and ERS alpha activities during the time epoch of interest in comparison with the reference period. The mean ERD/ERS values recorded for each of the three epoch windows (T1: 0–250 ms, T2: 251–500 ms, T3: 501–750 ms post-stimulus onset) were computed.

Statistical Analysis

The assumption of normality of data was assessed with the Kolmogorov–Smirnov test prior to the further statistical analysis. An independent *t*-test or chi-square test was conducted initially to analyze the means of the demographic and characteristic differences between the two groups in order to determine the appropriate assignment of the two groups.

The response times of correct responses as well as accuracy levels were separately computed by a 2 (fitness: high-fitness, low-fitness) \times 2 (Stroop congruency: neutral, incongruent) mixed-model repeated ANOVA with Greenhouse-Geisser correction where appropriate.

The statistical analyses were confined to the Fz electrode location, with upper and lower alpha ER% values examined separately. Each of the three epoch windows (i.e., T1, T2, and T3) for the lower and upper alpha ERD measures were separately computed by a 2 (fitness) × 2 (Stroop congruency) mixed-model repeated ANOVA.

Findings of significant ANOVA effects were followed by multiple *t*-tests with Bonferroni adjustments. Greenhouse-Geisser corrections were utilized for the purpose of correcting for any violations of sphericity. Means and standard errors were presented. The family-wise alpha level was set at 0.05 for all statistical analyses.

RESULTS

Participant Characteristics

Descriptive data regarding the participants' demographic data are presented in **Table 1**.

No significant differences in age, height, weight, or BMI (*ts* > -1.2, *p* > 0.1) were observed between the two groups. Additionally, there were no significant differences between these two groups in terms of the raw scores of the Digit Span test and the MMSE (*ts* > -1.5, *p* > 0.1). With regard to cardiorespiratory fitness levels and physical activity levels, there were significant differences between the two groups for both VO_{2max} and IPAQ values (*ts* < -2, *p* < 0.01), suggesting that the participants were properly assigned into the two groups.

Behavioral Data

The two-way ANOVA of response time revealed a main effect of group, $F_{(1,38)} = 17.42, p < 0.001$, partial $\eta^2 = 0.31$, with shorter response time on high fitness (583.55 ± 15.89 ms) compared to low fitness group (677.34 ± 15.89 ms). A main effect of Stroop conditions was also revealed, $F_{(1,38)} = 238.71, p < 0.001$, partial $\eta^2 = 0.86$, with longer response time on incongruent condition (675.16 ± 12.46 ms) compared to control condition (585.73 ± 10.68 ms; **Figure 1**).

The analysis of accuracy revealed only a main effect of Stroop congruency, $F_{(1,38)} = 28.58, p < 0.001$, partial $\eta^2 = 0.43$, with lower accuracy exhibited for the incongruent condition (0.90 ± 0.02) compared to the neutral condition (0.95 ± 0.01).

ERD/ERS Data

Upper Alpha (11–13 Hz)

The analysis of upper alpha ER% values for the T1 and T2 epoch windows revealed no main effects or interaction effects. The analysis of values for the T3 epoch window revealed a main effect of fitness, $F_{(1,34)} = 5.16, p < 0.03$, partial $\eta^2 = 0.13$, with less positive upper alpha ERD values for the high-fitness participants (-45.40 ± 15.00) than for the low-fitness participants (0.30 ± 13.42). A main effect of Stroop congruency was also revealed, $F_{(1,34)} = 11.63, p < 0.002$, partial $\eta^2 = 0.26$, with more upper alpha ERS values exhibited for the incongruent condition (-2.90 ± 6.30) compared to the neutral condition (-42.20 ± 15.14 ; **Figure 2**).

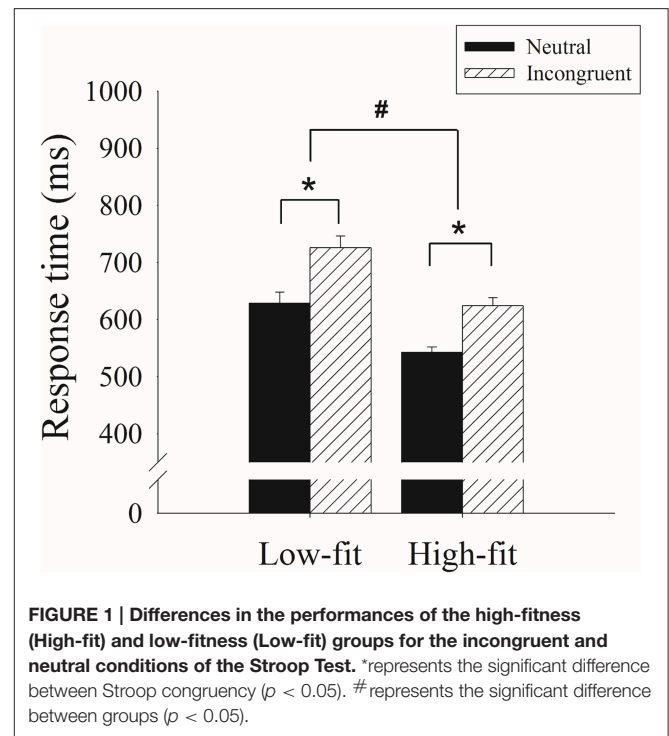


FIGURE 1 | Differences in the performances of the high-fitness (High-fit) and low-fitness (Low-fit) groups for the incongruent and neutral conditions of the Stroop Test. *represents the significant difference between Stroop congruency (*p* < 0.05). # represents the significant difference between groups (*p* < 0.05).

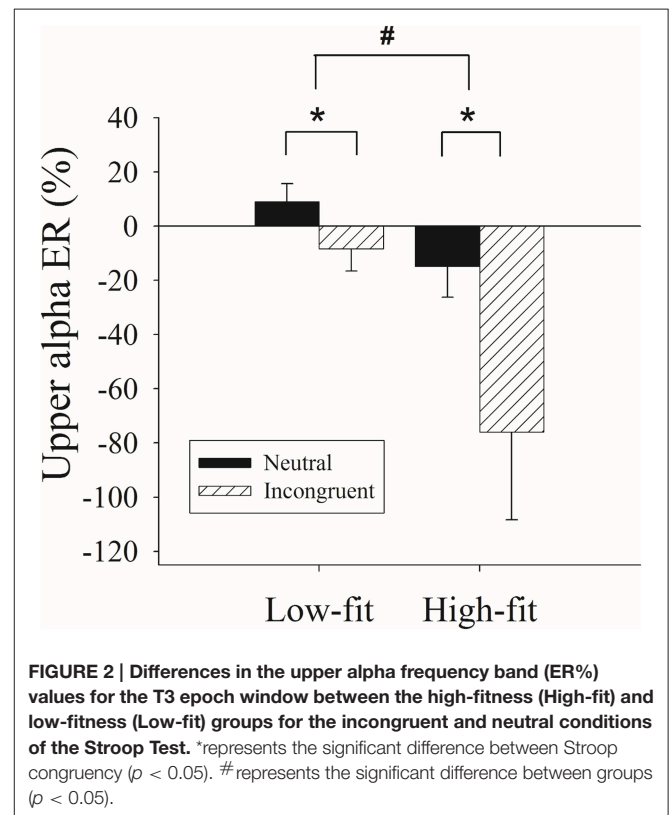


FIGURE 2 | Differences in the upper alpha frequency band (ER%) values for the T3 epoch window between the high-fitness (High-fit) and low-fitness (Low-fit) groups for the incongruent and neutral conditions of the Stroop Test. *represents the significant difference between Stroop congruency (*p* < 0.05). # represents the significant difference between groups (*p* < 0.05).

Lower Alpha (8–10 Hz)

The two-way ANOVA of lower alpha ER% values for the T1 epoch window revealed a main effect of fitness, $F_{(1,34)} = 4.60$,

$p < 0.04$, partial $\eta^2 = 0.12$, with greater positive lower alpha ERD values for high-fitness group (1.07 ± 3.38) compared to the low-fitness group (-8.65 ± 3.02). A main effect of Stroop congruency was also revealed, $F_{(1,34)} = 30.44$, $p < 0.001$, partial $\eta^2 = 0.46$, with less positive lower alpha ERD values exhibited for the incongruent condition (-13.06 ± 3.20) compared to the neutral condition (5.47 ± 2.38). However, the analysis of second time epoch revealed no any main and interaction effects.

Lower alpha ER% values for the T3 epoch window indicated significant effects of Stroop congruency [$F_{(1,34)} = 4.22$, $p < 0.05$, partial $\eta^2 = 0.11$], with the incongruent condition (18.73 ± 5.63) exhibiting greater positive lower alpha ERD values compared to the neutral condition (-2.39 ± 7.75). Additionally, a significant fitness \times Stroop congruency interaction was also observed [$F_{(1,34)} = 4.74$, $p < 0.04$, partial $\eta^2 = 0.12$]. The follow-up analysis revealed that the high-fitness group exhibited less positive lower alpha ERD values (-20.08 ± 11.55) than the low-fitness group (15.30 ± 10.33) in the neutral condition ($p < 0.03$), but not in the incongruent condition (23.42 ± 8.39 , 14.04 ± 7.50 , $p > 0.05$). Additionally, the high-fitness group exhibited more positive lower alpha ERD values in the incongruent condition (23.42 ± 8.39) compared to the neutral condition (-20.08 ± 11.55 ; $p < 0.01$), while the low-fitness group exhibited no such difference (14.04 ± 7.50 , 15.30 ± 10.33 , $p > 0.05$; **Figure 3**).

DISCUSSION

Despite the fact that an increasing number of studies have examined the relationship between cardiorespiratory fitness and cognitive functioning, no research has directly investigated that

relationship by utilizing ERD and ERS. Our primary behavioral findings revealed that late-middle-aged adults with higher levels of cardiorespiratory fitness demonstrated superior cognitive performance, as reflected by shorter reaction times in both congruency conditions of the Stroop Test, than those with lower cardiorespiratory fitness. Additionally, poor performances reflected by longer reaction time and less accuracy in incongruent condition were observed compared to those in neutral condition.

With regard to the upper alpha frequency, the high-fitness late-middle-aged adults exhibited less positive upper alpha ERD values in the later epoch window than their low-fitness counterparts. Additionally, the Stroop incongruent condition produced more upper alpha ERS values than the neutral condition. With regard to the lower alpha frequency, the high-fitness late-middle-aged adults exhibited greater lower alpha ERD values in the early epoch window. However, for the late epoch window, the high-fitness late-middle-aged adults showed less positive lower alpha ERD values in the neutral condition, but not the incongruent condition, relative to their low-fitness counterparts. Additionally, the high-fitness late-middle-aged adults showed more positive lower alpha ERD values in the incongruent condition compared to the neutral condition, but no such difference with regard to Stroop congruence was exhibited by the low-fitness group.

Behavioral Data

Our behavioral results showing worse performances in the incongruent condition relative to the neutral condition are consistent with those of previous studies (Qiu et al., 2006; Fan et al., 2014; Chang et al., 2015) and suggested a typical “Stroop effect” (Stroop, 1935). In contrast with the neutral

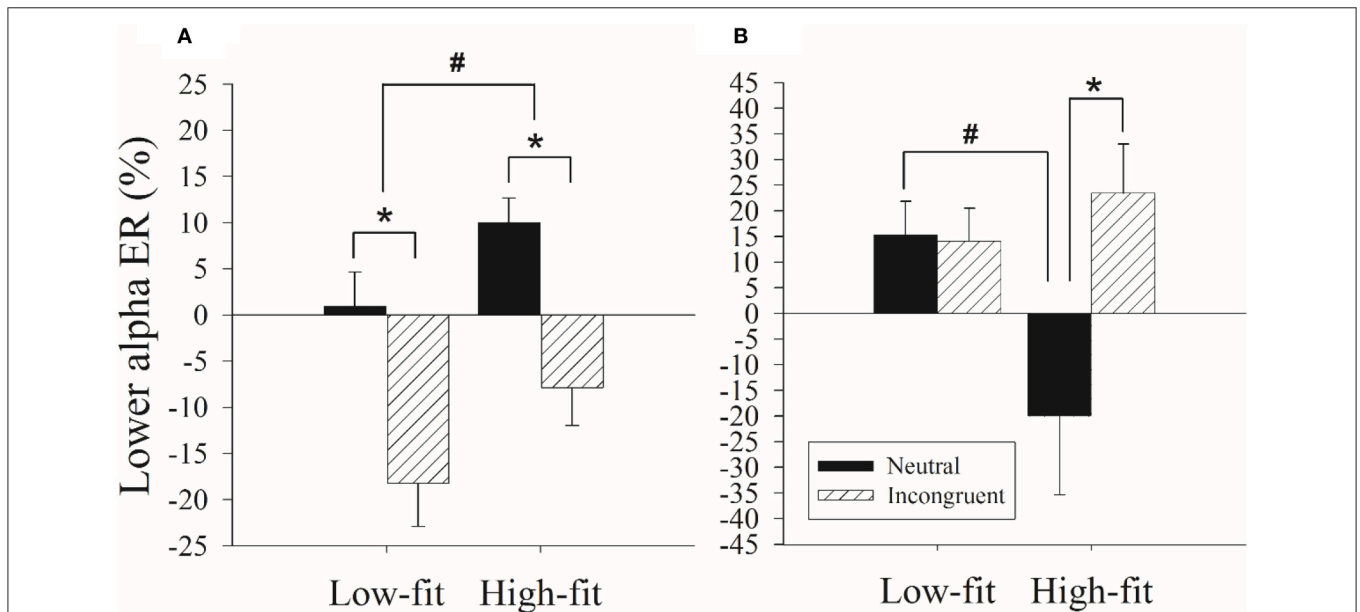


FIGURE 3 | Differences in the lower alpha frequency band (ER%) values for the (A) T1 epoch window and (B) T3 epoch window between the high-fitness (High-fit) and low-fitness (Low-fit) groups for the incongruent and neutral conditions of the Stroop Test. *represents the significant difference between Stroop congruency ($p < 0.05$). # represents the significant difference between groups ($p < 0.05$).

condition, which involves predominantly chromatic information, the incongruent condition involves conceptual discrepancies between the semantic meanings of the words and the chromatic dimensions (MacLeod and MacDonald, 2000). According to the parallel distributed processing of the Stroop Test (Cohen et al., 1990), conflict arises because the tendency of reading out the semantic meaning is stronger than that of naming the color. To perform the incongruent condition successfully, increasing attentional inhibitory processing engagement is required to inhibit the task-irrelevant information (MacLeod, 1991; Nigg, 2000); that is, greater cognitive resources are required of the participant in order for him or her to disengage their attention away from the task-irrelevant information (Guiney and Machado, 2013).

Our results indicating that the adults with higher levels of cardiorespiratory fitness demonstrated shorter response times across both the neutral and incongruent Stroop conditions than those with lower cardiorespiratory fitness were also in line with those of several previous studies (Prakash et al., 2011; Weinstein et al., 2012; Gauthier et al., 2015). These findings suggest that a positive association between fitness and general cognitive functions is reflected by the Stroop Test, at least in late-middle-aged adults. The beneficial effect of higher fitness was further confirmed by no difference in accuracy levels, indicating that the shorter response times were not attributable to a speed-accuracy trade-off. This better cognitive performance associated with higher fitness levels might be associated with increased brain function in older adults. For example, aged adults with higher cardiorespiratory fitness levels have demonstrated not only better behavioral performances in the Stroop Test, but also increased recruited activation of the prefrontal and parietal cortices (Prakash et al., 2011), greater preserved volume of the right inferior frontal gyrus and precentral gyrus (Weinstein et al., 2012), as well as aortic elasticity (Gauthier et al., 2015). Our study also expands upon previous studies that typically employed the Erikson flanker task (Colcombe et al., 2004; Hillman et al., 2006). Taken together, our study again confirms that cardiorespiratory fitness is positively linked to cognitive functioning in the aged adults.

ERD/ERS Data

The novelty of the current study was its exploration of the association between cardiorespiratory fitness and cognitive performance through variations in alpha frequency band activity (Pfurtscheller and Lopes Da Silva, 1999). EEG activities, such as ERD and ERS, have been suggested to be reflections of the uncoupling and coupling of functional networks in the cortex (Bastiaansen and Hagoort, 2006). Additionally, upper and lower alpha frequencies have been linked primarily to attention and task-related processing, respectively (Klimesch et al., 2007). Examining the upper and lower alpha ERD/ERS of neuronal activity for a given function might thus provide alternative explanations for the influence of cardiorespiratory fitness on cognitive performance.

Our results revealed that the adults with high-fitness exhibit less upper alpha ERD, reflecting increased alpha synchronization, which has typically reflected reduced information processing

in the “idling” state (Klimesch et al., 2007). According to the inhibition-timing hypothesis (Klimesch et al., 2007), however, enhanced cognitive performance is associated with increased upper alpha ERS through greater upper alpha activity or alpha oscillations, suggesting higher efficiency in inhibiting non-essential or task-irrelevant processing (Klimesch et al., 2007; Buschman et al., 2012; Bazanova and Vernon, 2014). Increased upper alpha frequency has also been observed during the encoding and retention interval periods, implying that inhibition of the retrieval of interference information was involved (Klimesch et al., 2005). That is, alpha synchronization could be considered to be a specific type of inhibition, in which the cognitive process is focused on the task-relevant information and interference from the non-task-relevant information is prevented (Klimesch et al., 2007). Accordingly, along with the positive association between fitness and Stroop Test performance, the results regarding higher upper alpha ERS might suggest that late-middle-aged adults with high levels of fitness have superior ability to inhibit or deselect the current task-irrelevant processing during the performance of the given task. Moreover, since no difference was observed in the upper alpha ERD values for the early epoch window between the high- and low-fitness groups, it is possible that a stronger influence of cardiorespiratory fitness occurs during the later stage of semantic encoding processes.

Lower alpha frequency desynchronization has been linked to attentional engagement for task-independent and non-stimulus factors (Klimesch et al., 1998; Klimesch, 1999). For instance, a decrease of lower alpha frequency activity, representing increased ERD, after a warning signal was observed, implies an association between lower alpha ERD and attentional demands such as alertness or expectance (Klimesch et al., 1998). In line with the interpretation of attentional processes, research involving dyslexic individuals observed significantly larger lower alpha desynchronization during the reading of words and pseudowords in such individuals than in non-dyslexic counterparts, suggesting that more attentional resources are engaged by dyslexic individuals during the encoding of words and pseudowords (Klimesch et al., 2001). Furthermore, during a combined Stroop-Task-Switching paradigm study, a larger degree of lower alpha ERD during the color-to-word condition compared to the word-to-word condition also suggested the involvement of attentional resources (Wu et al., 2015). That is, the larger positive lower alpha ERD values observed during the early epoch window following the stimulus onset observed in high-fitness individuals might suggest their superior ability, compared to those with lower cardiovascular fitness levels, to engage a greater amount of attentional resources in response to a stimulus.

In line with prior findings in which more alpha power for the incongruent condition compared to the neutral condition was observed (Hanslmayr et al., 2008), a larger lower alpha ERD in the incongruent condition than in the neutral condition in high-fitness individuals was observed in this study. However, it should be noted that during the later epoch window, no lower alpha ERD difference between the incongruent and neutral conditions was observed in the low-fitness participants. Based on the parallel distributed processing account of the Stroop effect (Cohen et al., 1990), conflict arises when participants attempt to

inhibit the highly activated, yet task-irrelevant response tendency (i.e., reading the word), and selectively attend to the relevant, but less activated response tendency (i.e., naming the color) in the incongruent condition. That is, increasing attentional inhibitory processing engagement is required to inhibit the task-irrelevant information (MacLeod, 1991; Nigg, 2000). Considering the superior behavioral performance of the high-fitness individuals compared to their low-fitness counterparts, the current findings might imply that high-fitness late-middle-aged adults are able to utilize more attentional resources for conflict resolution than their low-fitness older counterparts.

Limitations and Future Directions

Despite novel findings associated with cardiorespiratory fitness levels and cognitive functioning from the perspective of alpha ER% values, some limitations of the present study should be considered. First, a previous study reported differences in neuroelectric activities between males and females (Shen, 2005). Given that only male participants were recruited, whether or not our findings can be generalized to both genders requires further examination. Second, the association between cardiorespiratory fitness and executive control might be affected by other factors, such as education level and social economic status, in which these confounding factors are warranted to control. Additionally, while the Stroop Test, being a classical neuropsychological assessment, reflects inhibition appropriately, it should be noted that several different processes associated with inhibition can be involved during various inhibitory tasks, such as motor response inhibition tasks (e.g., the stop-signal task and go/no-go task) and the interference aspect of inhibition tasks (e.g., the Erickson flanker task and Stroop Test; Wostmann et al., 2013). Examining the alpha frequencies induced by utilizing various inhibitory tasks might provide a more comprehensive map for the relationship between fitness and inhibition. Moreover, the current alpha ERD/ERS study focused solely on the variation of alpha frequencies on the Fz electrode location. While it is reasonable as a preliminary study because of the high association between the prefrontal cortex and executive control, including more electrodes as well as more EEG frequency bands might

provide more comprehensive information related to ERD/ERS and clarify the associations between various cortical regions during cognitive processing. Future study is also encouraged to utilize longitudinal design to establish causal relationship.

CONCLUSION

The findings reported herein add imperative knowledge to the fitness-cognition literature by exploring differences in alpha ERD/ERS activity between high-fitness and low-fitness middle-aged adults. Middle-aged adults with greater cardiorespiratory fitness were showed superior cognitive function, as reflected by the Stroop Test, in comparison to those with lower fitness. Additionally, the results of neuro-electrophysiological index of the alpha ERD/ERS suggested that cardiorespiratory fitness is also related to the efficiency of inhibiting task-irrelevant information processing as well as the ability to engage greater attentional resources to given cognitive demands. Future research is encouraged to consider that generalization with regard to gender, types of inhibition cognitive tasks, and EEG frequencies to further the understanding of the relationships between cardiorespiratory fitness and cognitive functioning in different populations.

AUTHOR CONTRIBUTIONS

YC, TH, and CC made substantial contribution to conceive and design the experiment protocol, organize the experiment, as well as critically revise manuscript; KY, TS, and JL involved in conducting the experiment, statistical analysis processes, and development of overall research plan; all authors have involved in part of writing and approved the final manuscript.

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Obesity, Cardiovascular Fitness, and Inhibition Function: An Electrophysiological Study

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The purpose of the present study was to examine how obesity and cardiovascular fitness are associated with the inhibition aspect of executive function from behavioral and electrophysiological perspectives. One hundred college students, aged 18–25 years, were categorized into four groups of equal size on the basis of body mass index and cardiovascular fitness: a normal-weight and high-fitness (NH) group, an obese-weight and high-fitness (OH) group, a normal-weight and low-fitness (NL) group, and an obese-weight and low-fitness (OL) group. Behavioral measures of response time and number of errors, as well as event-related potential measures of P3 and N1, were assessed during the Stroop Task. The results revealed that, in general, the NH group exhibited shorter response times and larger P3 amplitudes relative to the NL and OL groups, wherein the OL group exhibited the longest response time in the incongruent condition. No group differences in N1 indices were also revealed. These findings suggest that the status of being both normal weight and having high cardiovascular fitness is associated with better behavioral and later stages of electrophysiological indices of cognitive function.

Keywords: obese, fitness, Stroop test, P3, N1

INTRODUCTION

Obesity, an emerging global pandemic, has not only been found to be associated with various health problems, including cardiovascular disease, diabetes, stroke, and high blood pressure (Nilsson and Nilsson, 2009; Ng et al., 2014), but has also been found to be associated with psychological and social issues such as a distorted body image and poorer interpersonal relationships (Edmunds et al., 2001). Moreover, harmful obesity-related outcomes have also been reported to extend to cognitive function. Specifically, obesity has been found to be associated with an elevated risk of impaired cognitive function (Gunstad et al., 2010; Hsu et al., 2015), and this negative relationship between obesity and cognitive function has been demonstrated in people of various ages, including children, adolescents (Liang et al., 2014; Martin et al., 2014), and even older adults (Siervo et al., 2011).

Executive function, one aspect of cognitive profiles that has been found to be linked to obesity, has received a substantial amount of research attention (Fitzpatrick et al., 2013; Reinert et al., 2013). Executive function is believed to reflect higher-order and effortful cognitive control that serves to manage and supervise multiple basic cognitive processes in order to achieve purposeful or

goal-directed behaviors (Alvarez and Emory, 2006; Etnier and Chang, 2009), with the frontal lobe believed to be heavily involved (Alvarez and Emory, 2006; Anderson et al., 2008). Interestingly, obese status has been observed to be associated with neurostructural deficits in the prefrontal and orbitofrontal cortices (Reinert et al., 2013) and in the frontal-subcortical activation of cognitive function (Stanek et al., 2013), suggesting a physiological linkage between obesity and executive dysfunction.

Rather than being a singular concept, executive function is thought to involve multiple subdomains, including inhibition, updating, and shifting (Miyake et al., 2000), with the inhibition aspect of executive function having been examined most often in obesity-related studies (Reinert et al., 2013). Inhibition refers to the ability to focus limited attention on relevant information or to resolve conflicting responses from unrelated or distracting stimuli (Heinemann et al., 2009; Wostmann et al., 2013). Kamijo et al. (2012a) indicated that higher body weight and fat mass, as assessed, respectively, by body mass index (BMI) and dual-energy X-ray absorptiometry, are negatively associated with performance on cognitive tasks requiring substantial inhibitory control, but not tasks requiring only limited degrees of inhibition, a finding which suggests that obesity is particularly relevant to inhibition. Similar findings indicating an association between obesity and impaired inhibition have also been reported (Kamijo et al., 2012b; Nederkoorn et al., 2012; Wirt et al., 2014).

One factor that has been found to have a protective effect on executive function is cardiovascular fitness; the positive association between the fitness and cognitive function has been reported by several meta-analytic reviews (Sibley and Etnier, 2003; Angevaren et al., 2008; Smith et al., 2010; Scherder et al., 2014). Additionally, high levels of cardiovascular fitness have been observed to have benefits with regard to multiple cognitive functions, with larger effects having been observed on executive function (Colcombe and Kramer, 2003). Research has also shown a robust relationship between cardiovascular fitness and inhibition. For example, Dupuy et al. (2015) found that highly fit females, regardless of whether they are young or old, exhibit better inhibition performance on the Stroop Task than those with low-fitness status. Other studies employing longitudinal designs also demonstrated better Stroop Task performances following an aerobic exercise training program lasting either 10 months (Smiley-Oyen et al., 2008) or 16 weeks (Vaughan et al., 2014) compared to non-exercise control interventions. While a positive effect of cardiovascular fitness on inhibition has thus been suggested, it should be noted that these studies generally examined normal-weight, older populations. As such, whether cardiovascular fitness can moderate the relationship between obesity and inhibition remains unknown.

The relationship between fitness and executive function has been investigated through the measurement of event-related potentials (ERPs). More specifically, the term "ERP" refers both to the electrical activity of the brain connected to a specific task event or response and the neuroelectrical or electrophysiological technique that offers precise temporal resolution of such neural processes underlying task performance (Fabiani et al., 2009). ERP studies investigating fitness and executive function have generally reported a relationship between

higher levels of cardiovascular fitness and increased P3 amplitude during the performance of executive function tasks (Hillman et al., 2004, 2006; Pontifex et al., 2011; Chang et al., 2013). This association between P3 amplitudes and fitness may be of particular importance to the obese population. Tascilar et al. (2011) examined cognitive function and related P3 alterations among children with and without obesity and found that obese children demonstrated decreased P3 amplitudes compared to their non-obese counterparts. When they further categorized those obese children into sub-groups consisting of those with or without insulin resistance, greater decreases in P3 amplitudes were observed in the obese children with insulin resistance than among the children without such resistance, suggesting that obesity is associated with impaired electrophysiological activity.

Studies of the relationship between fitness and ERPs have typically placed an emphasis on the P3 component. However, studies of other, less-examined ERP components, such as the N1 component, have reported mixed results. For example, Chang et al. (2013) observed that highly active older adults demonstrated not only larger P3 amplitudes but also larger N1 amplitudes during an update task relative to those with low activity levels. However, in a study of younger adults with high or low fitness levels, only P3 (but not N1) amplitudes during a shifting task were found to be significantly different between the two groups (Scisco et al., 2008). These inconsistent findings may be attributable to the heterogeneous methods employed by the different studies, so further investigations of fitness and its relationship, if any, to the N1 are required. Relatedly, to our knowledge, no previous study has examined the relationships between fitness, inhibition, and N1 in both normal- and obese-weight populations.

Taken together, obesity and cardiovascular fitness have been associated, respectively, with weakened and strengthened inhibitory capacity in executive function. However, few studies have explored the relationships among cardiovascular fitness, obesity, and inhibition simultaneously. Furthermore, how cardiovascular fitness and obesity are associated with multiple electrophysiological activities during inhibition is not yet well-understood. The purpose of the present study, then, was to examine the relationship between cardiovascular fitness and inhibition in both obese and non-obese adults, with both behavioral and electrophysiological activity in terms of the P3 and N1 components of ERPs being investigated. It was expected that obese individuals would exhibit worse inhibition-related performances in terms of both behavioral and electrophysiological measures than the normal-weight individuals. It was also expected that the relationship between obesity and inhibition would be moderated by cardiovascular fitness in those obese individuals who had high cardiovascular fitness.

MATERIALS AND METHODS

Participants

Male university students between 18 and 25 years of age were recruited via internet advertisements and flyers posted in communities and health centers in cities in northern Taiwan.

These potential participants were required to meet the following initial inclusion criteria: (a) normal or corrected-to-normal vision, (b) no color blindness, (c) no history of neurological disorders or cardiovascular disease, and (d) the capacity to complete a cardiovascular fitness assessment measured by the Physical Activity Readiness Questionnaire (PAR-Q). Potential participants meeting those requirements were also required to meet the additional criterion of being either normal-weight (BMI = 18.5–24 kg/m²) or obese (BMI > 27 kg/m²). The determination of an individual participant's weight status was based on the BMI norms for adults published by the Ministry of Health and Welfare, Taiwan. Finally, potential participants also had to meet the criterion of having either a high (maximal oxygen uptake measures, VO_{2max} > 65th percentile) or low (VO_{2max} < 35th) fitness level. The cardiovascular fitness status of the participants was determined according to normative data provided by the American College of Sports Medicine (2013). One hundred participants met the above inclusion criteria and were then placed into one of the following four groups: the normal-weight and high-fitness (NH) group, the obese-weight and high-fitness (OH) group, the normal-weight and low-fitness (NL) group, or the obese-weight and low-fitness (OL) group. **Table 1** summarizes the participants' demographic information. All the participants completed a written informed consent form in accordance with the requirements of the Institutional Review Board of National Taiwan University.

Cardiovascular Fitness Assessment

The cardiovascular fitness (indexed as estimated VO_{2max}) of each participant was estimated using a YMCA cycling ergometer submaximal exercise test (Golding et al., 1989). The protocol was designed for adults with a Class A risk stratification (Fletcher et al., 2001) and consists of a series of 2–4 consecutive 3-min cycling stages. Through the whole testing process, the participant's heart rate (HR) was continuously monitored by a Polar heart rate monitor (Sport Tester PE 3000, Polar Electro Oy, Kempele, Finland). Participants were instructed to sit and pedal at a constant speed of 50 rpm with a workload of 150 kpm/min (25 W) on an electronically braked cycle ergometer (Ergoselect 100/200 Ergoline GmbH, Germany) for the first 3-min stage. After the initial 3-min stage, the workload was increased in accordance with the given participant's steady-state HR recorded during the last 15–30 s of the first 3-min stage. For instance, if the participant's HR was less than 80 bpm, then the workloads for the second and third 3-min stages would be set to 125 W (750 kpm/min) and 150 W (900 kpm/min), respectively. An additional 3-min cycling stage (at 150 kpm/min or 25 W) would be added if the participant's target HR, which was 85% of the participant's age-predicted HR_{max}, was not achieved. A numerical rating of perceived exertion (RPE) from the Borg 6 to 20 (Borg, 1982) scale was provided by the participant every 2 min during the cardiovascular fitness assessment.

Stroop Task

The study also employed a computer-based modified Stroop Color-Word test (Stroop, 1935), a widely utilized neuropsychological test of both basic information processing

and executive function. Two Stroop conditions, the incongruent and neutral conditions, were employed. For the incongruent condition, the visual stimuli were Chinese words (e.g., 紅 [Red], 綠 [Green], and 藍 [Blue]). However, the semantic meaning of the given word and the actual pixel color of it as it appeared on the screen were inconsistent in the incongruent condition (e.g., the word 紅 [Red] was shown in blue color). For the neutral condition, colored squares of red, green, or blue were used as the stimuli. A single test block consisted of 36 incongruent and 36 neutral stimuli with a mixed and random order presentation. Participants were required to complete six blocks (432 trials in total), with a 2-min rest between each block. Each individual trial began with a fixation cross being displayed for 600 ms, followed by the presentation of the given 2-cm stimulus for 500 ms in the center of a 17-inch computer screen. Participants were instructed to respond in accordance with the color of the stimulus as quickly and accurately as possible by pressing one of the three color response buttons on a response box (10 cm × 8 cm × 2 cm box). Each trial was completed once a response was made between 200 and 1000 ms after the display of the stimulus or if no response was made within 1000 ms. The response time and number of errors of each correct response were further identified and recorded as behavioral indices.

Electrophysiological Recording and Analysis

Continuous scalp electroencephalographic (EEG) activity measurements were collected using the Neuroscan SynAmps² system Scan (Scan 4.0, Compumedics Neuroscan) via an elastic cap with 32 Ag/AgCl electrodes arranged according to the international standard 10–10 system (Quick-cap, Neuroscan, Inc., Lexington, VA, USA). A suitable elastic cap was chosen to fit each individual participant's head size and each electrode was filled with electro-gel to reduce the impedance of each electrode to 5 kΩ. The recording was re-referenced to the averaged mastoids, with AFz serving as the ground electrode. Adhesive electrodes were placed below and above the left orbit and on the outer canthus of each eye to monitor bipolar electrooculography (EOG) activity. With a low cutoff value (70 Hz) and high cutoff value (0.05 Hz), the data were digitized at an A/D rate of 500 Hz with a 60 Hz notch filter.

Offline EEG data were initially processed to remove the ocular artifact and segmented into epochs for time-locked ERP components. The epoch was set to last from 200 ms before and 1000 ms after the onset of the stimulus. The baseline correction was conducted using the 200 ms pre-stimulus period. Data were then filtered using a low-pass shift 30 Hz (12 dB/octave). Any trials with a response error or artifact exceeding ±100 μV were rejected before averaging. The remaining effective data were then averaged. The P3 and N1 components from each correct response were identified and recording as electrophysiological indices. The time windows for peak detection of the P3 and N1 components were 300–700 ms (positive-going peak) and 100–150 ms (negative-going peak), respectively. The ERP amplitude and latency from Fz, Cz, and Pz were collected for further statistical analysis (Hillman et al., 2005; Themanson and Hillman,

2006; Chang et al., 2013; Dai et al., 2013). Scalp topographies based upon 32 electrodes for N1 and P3 components across the four groups were also provided.

Experimental procedure

Each participant was required to visit the laboratory located on the National Taiwan Sport University campus individually on two occasions within a 7-days interval. On the first visit, a brief introduction regarding the experimental procedure was provided and the participant then filled out the informed consent form. Next, the participant's demographic information, BMI, and working memory aspect of the intelligence quotient assessed by the Digit Span test of the Wechsler Adult Intelligence Scale-Third Edition (WAIS-III; Wechsler, 1997) were acquired. The participant was then fitted with a Polar heart rate monitor and instructed to perform the YMCA cycling ergometer submaximal exercise test. The participants were only required to visit the laboratory twice if they met the cardiovascular fitness and obesity status criteria.

On their second visit, the qualified participants were instructed to be comfortably seated in a chair in a dimly lit, sound-proof, electrically shielded room. Prior to the official test, each participant was instructed to practice the Stroop Task until a correct response rate of 85% was achieved, during which time the electrophysiological recording settings were prepared. The official Stroop Task was then administered and EEG recordings were taken throughout the task performance period. Each participant was compensated with a payment of approximately US \$30 dollars for participating in the experiment.

Statistical Analysis

Statistical analyses were performed using SPSS version 21.0 (IBM, Corp., Armonk, NY, USA). Demographic data were analyzed using the one-way analysis of variance (ANOVA) to examine any potential demographic differences among the four groups. For behavioral data, a 4 (Group: NH, OH, NL, OL) \times 2 (Stroop condition: neutral, incongruent) mixed-model ANOVA was employed for response time and number of errors, respectively. For the ERP components, a 4 (Group) \times 2 (Stroop condition) \times 3

(Site: Fz, Cz, Pz) mixed-model ANOVA was employed to analyze P3 amplitude, P3 latency, N1 amplitude, and N1 latency, respectively. The Greenhouse-Geisser method that was utilized to correct for violations of the sphericity assumption. *Post hoc* Student-Newman-Keuls and multiple *t*-test comparisons with Bonferroni correction were conducted where appropriate. The partial eta squared (η^2) was reported for significant effects, as determined by $p < 0.05$, before any statistical adjustment.

RESULTS

Participant Characteristics

One-way ANOVAs revealed no significant differences among the four groups in age, height, or Digit Span test scores. However, the analysis revealed significant differences among the four groups in weight [$F(3,96) = 53.50, p < 0.001$], BMI [$F(3,96) = 86.00, p < 0.001$], waist/hip ratio [$F(3,96) = 43.02, p < 0.001$], body fat mass [$F(3,96) = 94.59, p < 0.001$] and VO_{2max} [$F(3,96) = 182.24, p < 0.001$]. *Post hoc* Student-Newman-Keuls tests revealed that the OH and OL groups had higher values of weight, BMI, waist/hip ratio, and body fat mass than the NH and NL groups ($ps < 0.05$), while the NH and OH groups had higher estimated VO_{2max} values than the NL and OL groups ($ps < 0.05$; **Table 1**).

Behavioral Data

Response Time

A two-way ANOVA revealed a main effect of the group [$F(3,96) = 9.25, p < 0.001, \eta_p^2 = 0.22$]. A *post hoc* Newman-Keuls test revealed that shortest response time was observed for the NH group (474.46 ± 16.76 ms), followed by the OH group (526.53 ± 16.76 ms) and the NL group (542.79 ± 16.76 ms), while the longest response time was observed for the OL group (598.25 ± 16.76 ms, $ps < 0.05$).

The analysis also revealed a main effect of the Stroop condition [$F(1,96) = 250.69, p < 0.001, \eta_p^2 = 0.72$]. The follow-up comparison revealed that a shorter response time was observed for the neutral (492.57 ± 7.53 ms) condition as compared to the incongruent condition (578.45 ± 9.92 ms, $p < 0.001$).

TABLE 1 | Demographic and cardiovascular fitness characteristics among four groups (mean \pm SD).

Variables	NH (n = 25)	OH (n = 25)	NL (n = 25)	OL (n = 25)
Age (years)	21.00 \pm 1.80	20.84 \pm 2.30	21.96 \pm 2.19	21.16 \pm 2.23
Height (cm)	174.00 \pm 4.06	178.36 \pm 8.43	175.84 \pm 6.66	173.88 \pm 5.99
Digit Span test				
Forward	14.88 \pm 0.83	14.24 \pm 1.17	14.52 \pm 1.26	14.12 \pm 1.17
Backward	10.00 \pm 3.11	7.84 \pm 2.78	8.44 \pm 2.79	8.36 \pm 3.09
Weight				
Weight (kg)	64.64 \pm 5.28 ^a	95.44 \pm 17.18 ^b	66.92 \pm 7.42 ^a	101.20 \pm 17.14 ^b
BMI (kg/m ²)	21.34 \pm 1.42 ^a	29.80 \pm 3.32 ^b	21.59 \pm 1.44 ^a	33.42 \pm 5.23 ^b
Waist/hip ratio	0.78 \pm 0.04 ^a	0.87 \pm 0.05 ^b	0.82 \pm 0.04 ^a	0.91 \pm 0.04 ^b
Body fat mass (%)	13.97 \pm 3.70 ^a	24.28 \pm 3.87 ^b	16.01 \pm 3.80 ^a	30.10 \pm 4.01 ^b
VO_{2max} (ml/kg/min)	54.66 \pm 4.81 ^a	51.96 \pm 3.44 ^a	35.77 \pm 2.89 ^b	36.23 \pm 3.49 ^b

NH, normal-weight and high-fit; OH, obese-weight and high-fit; NL, normal-weight and low-fit; OL, obese-weight and low-fit; VO_{2max} , estimated VO_{2max} . ^{a,b}Significant differences between groups, $p < 0.05$.

An interaction of group and Stroop condition was also observed [$F(3,96) = 4.83, p < 0.01, \eta_p^2 = 0.13$]. The follow-up comparison revealed a shorter response time for the NH group (440.15 ± 15.06 ms) relative to NL and OL groups (504.70 ± 15.06 and 537.68 ± 15.06 ms, $ps < 0.05$) for the neutral condition. For the incongruent condition, the OL group had the longest response time (658.81 ± 19.85 ms), followed by the NL, OH, and NH groups ($580.89 \pm 19.85, 565.29 \pm 19.85,$ and 508.78 ± 19.85 ms, $ps < 0.05$), but no significant differences were observed among the latter three groups. In addition, all four groups had a shorter response time for the neutral as compared to the incongruent condition ($ps < 0.001$; **Figure 1**).

Number of Errors

The analysis revealed no main effect of group, but did reveal a main effect of Stroop condition [$F(1,96) = 58.72, p < 0.001, \eta_p^2 = 0.38$]. The follow-up comparison revealed that there were fewer errors for the neutral condition ($91.40 \pm 0.01\%$) than for the incongruent condition ($86.10 \pm 0.01\%$). No significant interaction of group and Stroop condition was observed ($F = 1.93, p > 0.05$).

ERP Data

Figure 2 illustrates the grand-averaged ERP waveforms and **Figure 3** illustrates the topographic distributions of the P3 voltages across the four groups.

N1 Amplitude

The three-way ANOVA revealed no main effect of group, but it did reveal main effects of Stroop condition [$F(1,96) = 53.26, p < 0.001, \eta_p^2 = 0.36$] and site [$F(2,192) = 30.09, p < 0.001,$

$\eta_p^2 = 0.24$]. The follow-up comparison revealed that a larger N1 amplitude was observed for the incongruent condition ($-2.56 \pm 0.29 \mu\text{V}$) than for the neutral condition ($-1.55 \pm 0.30 \mu\text{V}, p < 0.001$). In addition, larger N1 amplitudes were observed for the Fz ($-2.43 \pm 0.31 \mu\text{V}$) and Cz ($-2.31 \pm 0.31 \mu\text{V}$) sites as compared to the Pz site ($-1.43 \pm 0.27 \mu\text{V}$). No other significant effects were observed.

N1 Latency

The analysis revealed no main effect of group, Stroop condition, or site, in addition to revealing no two- or three-way interaction effects ($ps > 0.05$).

P3 Amplitude

The analysis revealed a main effect of the group [$F(3,96) = 6.67, p < 0.001, \eta_p^2 = 0.17$]. *Post hoc* Student-Newman-Keuls tests revealed a larger P3 amplitude for the NH group ($12.69 \pm 0.82 \mu\text{V}$) than for the NL ($9.06 \pm 0.82 \mu\text{V}$), OH ($8.60 \pm 0.82 \mu\text{V}$), and OL groups ($8.02 \pm 0.82 \mu\text{V}, p < 0.05$).

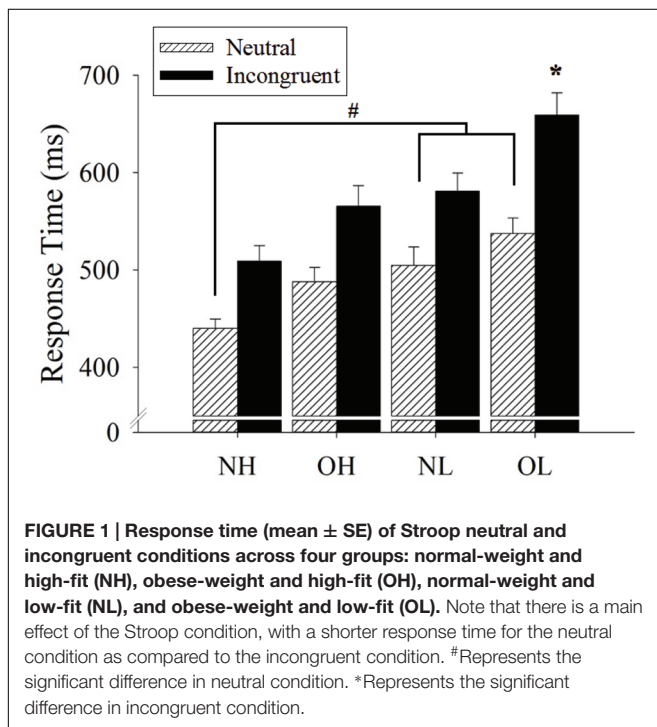
The analysis also revealed significant main effects of Stroop condition [$F(1,96) = 22.80, p < 0.001, \eta_p^2 = 0.19$] and site [$F(2,192) = 63.64, p < 0.001, \eta_p^2 = 0.40$]. The follow-up comparison revealed that a larger P3 amplitude was observed for the incongruent condition ($10.08 \pm 0.44 \mu\text{V}$) than for the neutral condition ($9.11 \pm 0.40 \mu\text{V}, ps < 0.001$). In addition, the largest P3 amplitude was observed for the Cz site ($10.99 \pm 0.49 \mu\text{V}$), followed by the Pz site ($10.13 \pm 0.41 \mu\text{V}$), and the Fz site ($7.65 \pm 0.43 \mu\text{V}, ps < 0.01$).

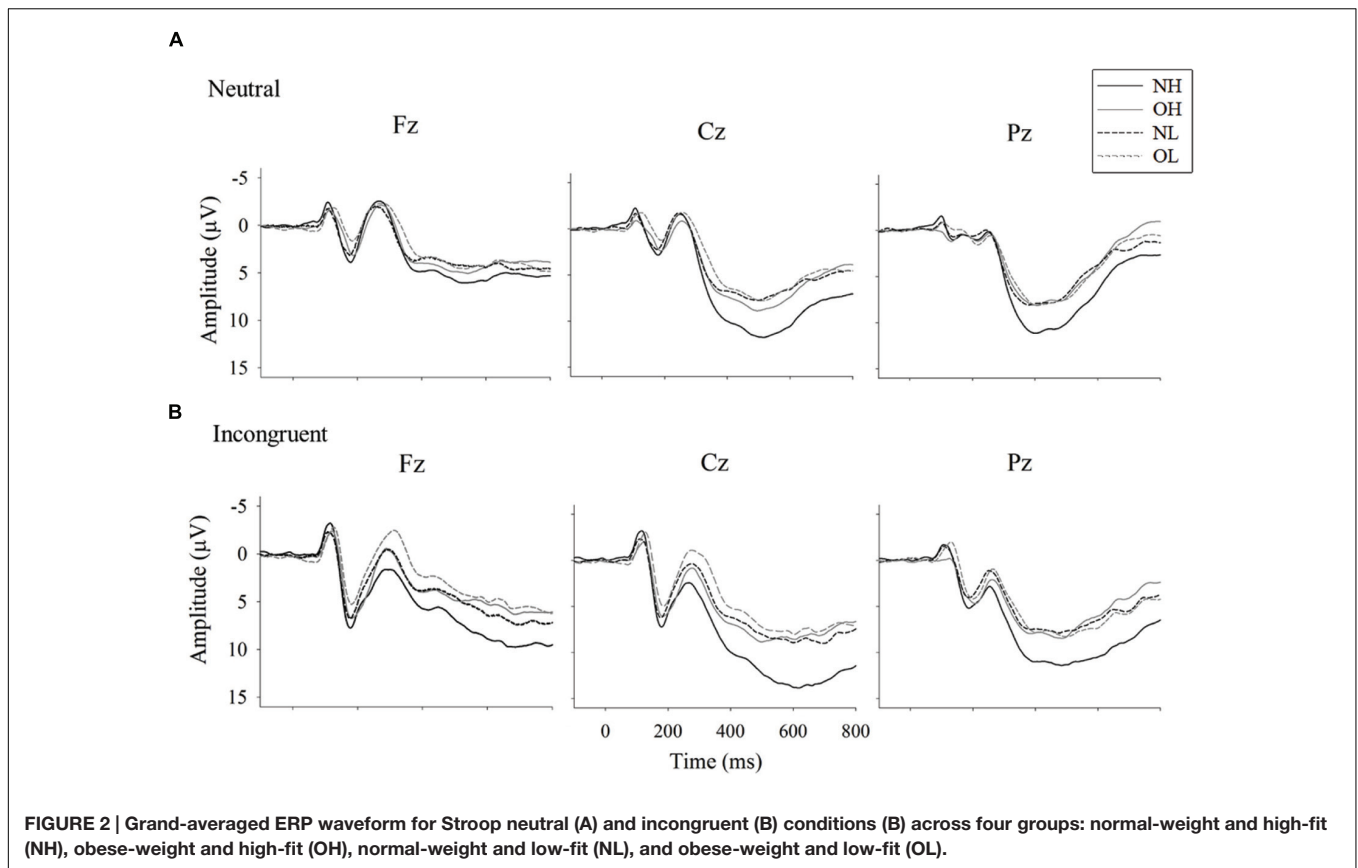
An interaction of group and Stroop condition was observed [$F(3,96) = 2.72, p < 0.05, \eta_p^2 = 0.08$]. The follow-up comparison showed that the NH group had a larger P3 amplitude than the NL, OH, and OL groups in both the neutral (11.78 ± 0.80 vs. $8.46 \pm 0.80, 8.35 \pm 0.80$ and $7.84 \pm 0.80 \mu\text{V}, ps < 0.05$) and incongruent conditions (13.60 ± 0.88 vs. $9.66 \pm 0.88, 8.86 \pm 0.88$ and $8.20 \pm 0.88 \mu\text{V}, ps < 0.05$), with no significant differences among the latter three groups. Both the NH and NL groups had a larger P3 amplitude in the incongruent condition than in the neutral condition ($p < 0.01$), whereas the same difference was not observed in the OH and OL groups.

An interaction between Stroop condition and site was also observed [$F(2,192) = 10.93, p < 0.001, \eta_p^2 = 0.10$]. The follow-up comparisons revealed that the incongruent condition had larger P3 amplitudes than the neutral condition at the Fz (8.35 ± 0.48 vs. $6.96 \pm 0.41 \mu\text{V}, p < 0.001$), Cz (11.49 ± 0.53 vs. $10.49 \pm 0.48 \mu\text{V}, p < 0.001$), and Pz sites (10.39 ± 0.44 vs. $9.87 \pm 0.41 \mu\text{V}, p < 0.05$). The Fz site had a smaller P3 amplitude than the Cz and Pz sites ($ps < 0.001$) for the incongruent condition, whereas the Fz site had the smallest P3 amplitude, followed by the Pz and Cz sites, for the neutral condition ($ps < 0.01$).

P3 Latency

The analysis revealed only significant main effects of Stroop condition [$F(1,96) = 78.82, p < 0.001, \eta_p^2 = 0.45$] and site [$F(2,192) = 96.59, p < 0.001, \eta_p^2 = 0.50$]. The follow-up comparison revealed a longer P3 latency for the incongruent condition (546.19 ± 10.64 ms) than for the neutral condition (477.94 ± 9.64 ms, $p < 0.001$). In addition, the longest P3 latency





was observed for the Fz site (552.72 ± 11.17 ms), followed by the Cz site (528.13 ± 10.29 ms), and the Pz site (455.36 ± 9.35 ; $p < 0.001$).

DISCUSSION

The current study is among the first to examine how obesity and cardiovascular fitness affect the inhibition aspect of executive function from behavioral and electrophysiological perspectives. Our primary behavioral findings generally revealed that the NH and OL group demonstrated the shortest and longest response time for the Stroop Task, respectively. In addition, the OL group demonstrated the longer response time relative to the NH in the neutral condition and longest response time compared to other three groups (i.e., NL, OH, and NH) in the incongruent condition. In terms of ERP measurements, the NH group demonstrated a larger Stroop Task-induced P3 amplitude than the other three groups, while there were no significant differences among the other three groups. However, no group differences were observed for the N1 indices across the four groups.

Obesity, Cardiovascular Fitness, and Behavioral Measures

Our results showing longer response times and more errors for the incongruent condition than for the neutral condition indicated a typical phenomenon known as the “Stroop Effect”

(Chang et al., 2015a,b). That is, the incongruent Stroop condition required more time from the involvement of executive function to inhibit or over-ride prepotent responses than did the neutral condition, a finding that reflects basic information processes (Friedman et al., 2009). The presentation of the Stroop Effect also suggested that our modified Stroop Task was appropriately manipulated.

Shorter response times in the neutral condition for high cardiovascular fitness relative to low fitness were observed regardless of whether the individuals were normal-weight (i.e., the NH and NL groups) or obese (i.e., the OH and OL groups). Although, OL and NL demonstrated longer response time compared to NH, no differences were observed in individuals in high cardiovascular fitness (i.e., the NH and OH groups) or low cardiovascular fitness (i.e., the NL and OL groups). These results suggested that while obesity may be associated with slower basic information processing and fitness has larger influence in the processes. Fitness is related to increase the processing speed replicated those of previous obesity studies that examined other cognitive demands such as complex attention, verbal memory, and visual memory (Prickett et al., 2014), as well as a fitness study that examined a normal-weight population (Buck et al., 2008). At the same time, the results of the present study extend the knowledge base regarding associations between obesity, cardiovascular fitness, and Stroop task-related basic information processing.

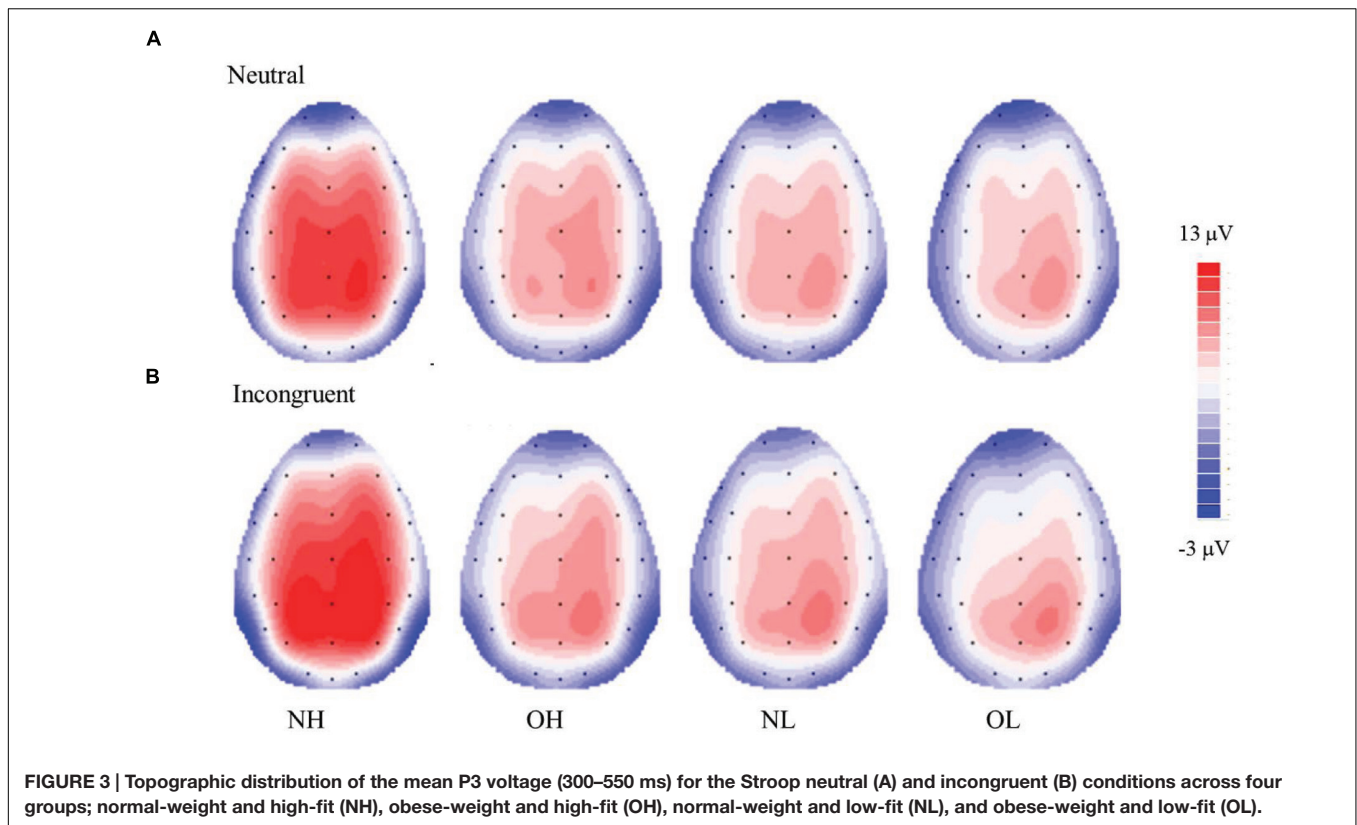


FIGURE 3 | Topographic distribution of the mean P3 voltage (300–550 ms) for the Stroop neutral (A) and incongruent (B) conditions across four groups; normal-weight and high-fit (NH), obese-weight and high-fit (OH), normal-weight and low-fit (NL), and obese-weight and low-fit (OL).

In terms of the incongruent condition, OL group had worse performance compared to other three groups (i.e., the OH, NL, and OL groups), with no significant difference being observed among the groups. This fact suggests that individual with both more obese and less fitness has a crucial influence on inhibition. These results are partly consistent with those of previous studies that reported a negative relationship between obesity and executive function (Fitzpatrick et al., 2013; Reinert et al., 2013; Miller et al., 2015). A notable study that utilized both cross-sectional and longitudinal perspectives previously demonstrated the positive association between cardiovascular fitness and executive function (Colcombe and Kramer, 2003; Chaddock et al., 2011). The beneficial effects of higher cardiovascular fitness have been linked to the greater brain volumes associated with better executive function (Erickson et al., 2009; Weinstein et al., 2012; Voss et al., 2013) and to the proliferation of new neurons or related molecules (e.g., *N*-acetylaspartate levels) associated with cognitive function (Erickson et al., 2012). Notably, our exploration extended the current understanding regarding how both obesity and cardiovascular fitness affects executive function.

Obesity, Cardiovascular Fitness, and Electrophysiological Measures

The ERP measures were partly consistent with the behavioral performances; that is, the group with normal weight and high cardiovascular fitness showed the largest P3 amplitude compared to the other three groups. However, this largest P3 amplitude was found in the NH group for both the neutral and incongruent

conditions of the Stroop Task. Our finding of a larger P3 amplitude in individuals with higher fitness compared to those with lower fitness was in line with other studies of ERP associated with fitness. For example, previous studies have found that individuals with higher fitness or who regularly engage in exercise demonstrated a larger P3 amplitude during a visual oddball task (Hillman et al., 2005) and an executive function-related task (Pontifex et al., 2011; Dai et al., 2013; Fong et al., 2014) than their less fit or less active counterparts. P3 amplitude is believed to reflect proportionally the amount of resources allocated toward the suppression of extraneous neuronal activity in order to facilitate attention (Polich, 2007). As such, it is plausible that the fitness level is associated with greater engaged attention to multiple aspects of cognitive domains.

While individuals with normal weight and high cardiovascular fitness showed a larger P3 amplitude compared to the obese individuals, it seems that cardiovascular fitness itself had a limited effect on the obese individuals, given that no P3 amplitude difference was observed between the OH and OL groups. These findings also suggest that obesity itself has a prominent effect in terms of impairing cognitive function as indicated by electrophysiological measurements. Given the previous evidence indicating that obesity is associated with lower P3 amplitudes (Tascilar et al., 2011), it is important to consider reducing the degree of obesity suffered by individuals in order to help them achieve higher levels of cognitive functioning. Moreover, our findings of different P3 amplitudes for the incongruent and neutral conditions in the normal-weight groups but not the obese

groups are similar to those of a study conducted by Kamiyo et al. (2012b), who reported that only healthy weight individuals, but not their obese counterparts, exhibited greater P3 topographic distributions for tasks involving or not involving inhibition (i.e., the Go/NoGo task). These findings again implied that obesity is associated with inferior inhibition. Furthermore, similar to the findings for the behavioral measures, no P3 amplitude differences were found for the NL group compared with OH and OL groups, suggesting that normal weight individuals with low fitness have lower levels of inhibition similar to those of obese populations. Taken together from an electrophysiological perspective, both reducing the level of obesity and increasing the level of cardiovascular fitness seem to be necessary for higher levels of cognitive function, regardless of the nature of cognitive functions.

Given that the majority of ERP studies related to fitness have placed an emphasis on the P3 component, our goal in this study was to further explore the N1 component. Unlike the P3 component, which is regarded as a late positive and endogenous component that represents the allocation of attentional resources, the N1 component is an early negative and exogenous component that reflects the initial extraction of visual discrimination processes (Fabiani et al., 2009). Contrary to our hypothesis based upon previous ERP studies associated with fitness (Chang et al., 2013) and obesity (Key and Dykens, 2008), we found that neither obesity nor cardiovascular fitness was associated with N1 amplitude and latency. It should be noted, however, that the Chang et al.'s study examined the level of physical activity in an older population, while the Key and Dykens (2008) study examined adults with Prader–Willi syndrome, a genetic disorder. Our results were consistent with a study by Scisco et al. (2008) that emphasized cardiovascular fitness in young adults. Accordingly, it is speculated that cardiovascular fitness and obesity may have less of an influence on the early stage of sensory information processing in healthy younger adults whose specific cognitive processing ability may have reached the ceiling level.

Limitations and Future Directions

The virtue of the present study is that it provides an initial step in identifying the association between cardiovascular fitness and obesity in terms of their effects on inhibition by examining both behavioral and electrophysiological results; however, it should be considered in light of several limitations. First limitation is related to the current cross-sectional design of the study, which meant that only associations among cardiovascular fitness, obesity, and inhibition, rather than cause-effect relationships, could be determined. It is suggested that further research be

conducted to examine the effect of fitness on inhibition among an obese population by conducting a long-term exercise program. It is also important to consider factors that may be confounded with obesity. For example, factors such as environment related to intake food and nutrition history have been shown to influence neurocognitive performance in obese populations (Duchesne et al., 2010) and therefore should be taken into consideration. Given that our scope is to examine the inhibition, we were unable to draw conclusions in regard to other aspects of executive function, such as updating, shifting, and planning. The further examination of these aspects specifically would offer a more focused understanding.

CONCLUSION

The present study provides evidence regarding the associations between obesity and inhibition as well as cardiovascular fitness and inhibition. Additionally, individuals with both normal weight and high cardiovascular fitness were found to exhibit significantly better behavioral results and greater electrophysiological measurements of the later stage of cognitive function, wherein those with both obesity and low cardiovascular fitness exhibited the poorest performances in executive function. These findings may support a primary practical suggestion that in order to achieve the optimal level of cognitive function, both a normal weight and high level of cardiovascular fitness should simultaneously be maintained.

AUTHOR CONTRIBUTIONS

Y-KC and CZ designed the study and oversaw the data collection. T-FS, Y-KC, and CZ analyzed the data and wrote-up the initial manuscript. LC, C-HC, and F-TS assisted with analysis of the data and organize the manuscript. All authors played a part in the preparation of the manuscript at each stage of its development. All authors have read and approved the final version of the manuscript.

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Motivation toward Physical Exercise and Subjective Wellbeing: The Mediating Role of Trait Self-Control

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Motivation toward physical exercise (MPE) and trait self-control (TSC) were identified as key predictors of subjective wellbeing (SWB). However, there has not been any research designed to examine the mediating role of TSC in the relationship between MPE and SWB. The present study utilizes self-determination theory, control-process theory of self-regulation, and theory of multiple pathways of TSC in order to examine whether TSC mediates the relationships of autonomous MPE (A-MPE), controlled MPE (C-MPE), and impersonal MPE (NO-MPE) with SWB using structural equation modeling (XLSTAT PLS). Three hundred seventeen adult American individuals ($M_{\text{age}} = 32.97$, $SD_{\text{age}} = 11.30$), who reported to be regular exercisers, voluntarily answered questionnaires assessing MPE, TSC, and SWB. Correlational analyses revealed positive relationships between A-MPE, TSC, and SWB, and negative relationships of C-MPE and NO-MPE with TSC and SWB. Mediation analyses revealed that TSC mediated the relationships of A-MPE (partial mediation) and C-MPE (full mediation) with SWB, but did not mediate the relationship between NO-MPE and SWB. The estimates of the quality of the hypothesized model were acceptable (outer model GoF = 0.935; absolute GoF = 0.330; relative GoF = 0.942; inner model GoF = 1.008; $R^2 = 36.947\%$). Finally, this study supports the view that MPE can influence SWB through TSC, and incites to pursue the examination of the relationships between self-determined motivation, self-regulation mechanisms, and health-related outcomes.

Keywords: physical activity, self-determined motivation, self-regulation, self-control, psychological health

INTRODUCTION

Subjective wellbeing (SWB), which can be defined as “. . . people’s evaluations of their lives – the degree to which their thoughtful appraisals and affective reactions indicate that their lives are desirable and proceeding well” (Diener et al., 2015, p. 234), represents a growing subject of interest around the world for psychologists, economists, philosophers, and politicians (e.g., Diener, 2000; Diener et al., 2015). Such a phenomenon would reflect the tendency of most societies to recognize the value of the human being and the importance of taking into account self-perceptions for evaluating individual’s life (e.g., Diener et al., 2015). Furthermore, the study of SWB lies in positive psychology that corresponds to the scientific study of positive psychological outcomes, psychological health, human capacities, and processes leading to fostering optimal functioning and development (e.g., Peterson and Park, 2003; Peterson and Seligman, 2004). In that perspective, research has found that SWB was a key predictor of health, longevity, moral behavior, and

performance (e.g., De Neve et al., 2013). For that reason, examining the predictive factors of SWB is of great importance. In addition, such a subject may represent a considerable interest to a worldwide audience.

Research has found that performing physical exercise regularly could promote health and SWB (e.g., Scully et al., 1998; Melzer et al., 2004; Puetz et al., 2006; Sjögren et al., 2006; Meyers, 2008; Biddle and Asare, 2012). Physical exercise represents a leisure-time physical activity, and can be defined as "...cumulative, acute bouts of physical activity that are planned, structured, and repeated and result in improvement or maintenance of one or more components of physical fitness, including cardiorespiratory capacity, muscle strength, body composition, and flexibility" (Puetz et al., 2006). Exercise psychologists have recently examined the relationship between the different types of commitment to exercise – operationalized through the notion of motivation toward physical exercise (MPE) – and SWB (e.g., Sebire et al., 2009; Gillison et al., 2011). Globally, they evidenced that the type of MPE (autonomous vs. controlled) could influence differently SWB. However, the mechanisms underlying such effects are still unclear and require additional investigations (e.g., Standage and Ryan, 2012). Self-control, which can be conceived as the effortful control and effortless (automatic) forms of goal-directed behavior (e.g., Hagger, 2013, 2014), would represent one of the most adaptive variables of the human psyche (e.g., Carver and Scheier, 1998; Tangney et al., 2004) and was found to play a key role in the development of SWB (e.g., Hofmann et al., 2014). Additionally, the type of motivation for goal pursuit appeared to affect the effectiveness of self-control processes (e.g., Milyavskaya et al., 2015). Therefore, the present article aims at examining *whether* the type of MPE might influence SWB through self-control.

Self-Determined Motivation and Subjective Wellbeing

Research focused on MPE frequently used the conceptual framework of self-determination theory (e.g., Deci and Ryan, 1985, 2000, 2008a,b). This theory is grounded in an organismic approach that considers that individuals are, by nature, active, curious, self-motivated, vital, and enthusiastic, even if they may display passivity, laziness, anxiety, or depression symptoms. Interestingly, the theory posits that individuals' positive or negative functioning would result from their interaction with the social environment that either supports or thwarts their deep nature, respectively. Specifically, the theory posits that the human psyche is characterized by three innate needs – i.e., needs for competence (i.e., need of being able to perform well something), relatedness (i.e., need of being connected with others), and autonomy (i.e., need of being at the origin of one's own behavior) – that serve the function of developing optimal functioning, performance, and wellbeing. The theory distinguishes three types of functioning: Autonomous, controlled, and impersonal regulations (or motivations).

Autonomous motivation reflects "...a motivational state in which self-initiation and coordination of personally endorsed

behaviors predominate" (Weinstein et al., 2011, p. 527). A social environment capable of satisfying the three innate needs is presumed to develop a strong sense of autonomous motivation, which is supposed to be related to optimal functioning, performance, and wellbeing. Intrinsic regulation (i.e., action is based on personal interest and satisfaction), integrated regulation (i.e., action is consistent with different aspects of the self), and identified regulation (i.e., action is personally valued and important) represent forms of autonomous motivation (e.g., Standage and Ryan, 2012). Controlled motivation reflects a "...functioning driven by externally imposed and introjected contingencies, eliciting pressure to conform to perceived expectations" (Weinstein et al., 2011, p. 527). It is assumed that a social environment that is capable of satisfying the needs for competence and relatedness but that thwarts the need for autonomy is presumed to develop a strong sense of controlled motivation. This type of motivation is supposed to be associated with a rigid functioning and a decreased wellbeing. Introjected regulation (i.e., action responds to the desire to avoid guilt and shame or to develop feelings of worth) and external regulation (i.e., action responds to the desire to obtain reward, to avoid punishment, or to meet external expectations) refer to controlled forms of motivation (e.g., Standage and Ryan, 2012). Impersonal motivation (or amotivation), which is associated with poor functioning, passivity and depressed symptoms, develops when the social environment thwarts the three needs (e.g., Standage and Ryan, 2012).

Grounded in the self-determination theory, empirical investigations have reported associations between the different types of motivation and constructs related to SWB (e.g., Gillison et al., 2006; Sebire et al., 2009). Specifically, it was found that autonomous forms of MPE (A-MPE) were positively related to: (a) SWB (measured through a composite construct combining subjective vitality and happiness, $r = 0.29$, Sebire et al., 2009), (b) quality of life (measured through a composite construct combining 10 health-related variables, $\beta = 0.37$, Gillison et al., 2006), (c) body satisfaction ($r_s = 0.28-0.34$, Gillison et al., 2011), and (d) physical self-worth ($r_s = 0.24-0.36$, Thøgersen-Ntoumani and Ntoumanis, 2006; Sebire et al., 2009). Authors also revealed that A-MPE was negatively related to exercise anxiety ($r = -0.33$, Sebire et al., 2009) and social physique anxiety ($r_s = -0.22$ to -0.11 , Thøgersen-Ntoumani and Ntoumanis, 2006; Gillison et al., 2011). In contrast, controlled forms of MPE (C-MPE) and impersonal MPE (NO-MPE) were associated with maladaptive psychological outcomes (e.g., Thøgersen-Ntoumani and Ntoumanis, 2006). Specifically, forms of C-MPE appeared to be positively related or unrelated to social physique anxiety ($r_s = 0.10-0.26$, Thøgersen-Ntoumani and Ntoumanis, 2006; Gillison et al., 2011). On the other hand, they appeared to be negatively related to body satisfaction ($r = -0.16$, Gillison et al., 2011) and SWB (measured through subjective vitality, $\beta = -0.81$, Edmunds et al., 2006), and negatively related or unrelated to physical self-worth ($r_s = -0.19$ to 0.01 , Thøgersen-Ntoumani and Ntoumanis, 2006). NO-MPE was positively related to social physique anxiety ($r_s = 0.18-0.21$, Thøgersen-Ntoumani and Ntoumanis, 2006; Gillison et al., 2011) and negatively related to

body satisfaction ($r = -0.19$, Gillison et al., 2011) and physical self-worth ($r = -0.18$, Thøgersen-Ntoumani and Ntoumanis, 2006).

Finally, there is strong evidence that A-MPE (or NO-MPE) can promote (or hinder) positive feelings. However, the detrimental effect of C-MPE on positive feelings lacks of consistent evidence in the literature. In the present study, we present some reasons to argue that the relationship between self-determined motivation and positive feelings might be explained, at least partly, by the intervention of self-regulation mechanisms, such as trait self-control (TSC).

Self-Determined Motivation and Trait Self-Control

Self-regulation reflects self-corrective adjustments that occur to be on track to attain the desired goal (e.g., Carver and Scheier, 1998), and self-control represents a crucial component of this broader phenomenon. Generally, self-control reflects the ability to operate changes in the self so as to develop an optimal adjustment between the self and the world (Tangney et al., 2004). More specifically, self-control can be defined as the capacity of the self to override prepotent responses and to regulate affects, cognitions, and behaviors (e.g., Baumeister and Heatherton, 1996; Tangney et al., 2004; Baumeister et al., 2007). For example, people with high TSC would be better at suppressing one goal (e.g., eating a palatable food) than people with low TSC to pursue another one (e.g., controlling one's weight) that is viewed to have greater importance or utility (e.g., Stroebe et al., 2013). Additionally, authors agree that self-control corresponds to a reservoir of limited resources designed to promote helpful responses (e.g., making plans) and inhibit unhelpful responses (e.g., inhibiting temptations) (e.g., Baumeister and Heatherton, 1996; Muraven, 2008; Hagger, 2013). For example, people with high TSC would have more available resources to self-regulate than people with low TSC.

According to the control-process theory of self-regulation (Carver and Scheier, 1990, 1998), the effectiveness of self-regulation mainly depends upon one's capacity to pursue clear goals. A strong adherence to a specific activity leads to operate a goal selection depending on the degree of *relevance* and *importance* of goals for the self. For example, in the case of physical exercise, such a goal selection could consist, on the one hand, in adopting certain goals – such as going to bed early, eating a balanced diet, etc., – and, on the other hand, in eschewing other goals – such as drinking alcohol, smoking, etc. Deci and Ryan (2000) considered that goal-directed activities might differ in the degree to which they are autonomous (i.e., enacted with a full sense of volition and choice) or controlled (i.e., enacted with a full sense of being pressured and controlled). Autonomous activities are consistent with one's integrated sense of self, whereas controlled activities have not been assimilated to the self and, thus, remain external to the self. Accordingly, people who pursue autonomous activities, relative to those who pursue controlled activities, would be

more likely to operate a goal selection in accordance with their activity. In that regard, autonomous motivation would be associated with better self-regulation processes than controlled motivation.

Research has revealed that exerting self-control for autonomous reasons was less depleting than exerting self-control for controlled reasons, allowing people to perform better on a subsequent task (Muraven et al., 2007, 2008; Muraven, 2008). Similarly, Moller et al. (2006) found that making choices for autonomous reasons were less depleting and led to better self-control performance than making choices for controlled reasons. This pattern of results suggest that autonomous motivation, relative to controlled motivation, would save more resources for subsequent tasks, thereby leading to maintaining focus on relevant goals. An important implication of such results is that autonomous self-control refers to the identification and integration of values, goals, and behavioral regulations, thereby leading to reinforcing self-control (Deci and Ryan, 1987).

Focusing on mechanisms of self-control, studies have found that autonomous motivation was globally associated with enhanced self-control. More specifically, autonomous motivation appeared to be positively related to TSC ($r = 0.22$, Briki et al., 2015) and implementation planning ($r_s = 0.15-0.31$, Koestner et al., 2008), and positively related or unrelated to automatic attraction toward helpful goals (e.g., healthy foods; $\beta_s = -0.05$ to 0.33 , Milyavskaya et al., 2015). However, it appeared to be negatively related to automatic attraction toward temptations (e.g., highly palatable, but unhealthy foods; $\beta_s = -0.25$ to -0.17 , Milyavskaya et al., 2015) and perception of encountering obstacles ($\beta_s = -0.25$ to -0.22 , Milyavskaya et al., 2015). Autonomous motivation appeared to be unrelated to controlled attraction toward helpful and unhelpful goals ($\beta_s = 0.01-0.09$, Milyavskaya et al., 2015) and perception of effort ($\beta = 0.03$, Milyavskaya et al., 2015). Using functional magnetic resonance imaging, Lopez et al. (2016) observed positive associations between autonomous motivation, positive mood, and the activity of inferior frontal gyrus – a brain zone reputed to be associated with inhibitory control –, suggesting that autonomous motivation would particularly involve the inhibition of goal-disruptive temptations. Furthermore, controlled motivation was globally unrelated to self-control (e.g., Briki et al., 2015; Milyavskaya et al., 2015). More specifically, it appeared to be unrelated to TSC ($r_s = 0.09-0.14$, Briki et al., 2015), implementation planning ($r_s = 0.01-0.05$, Koestner et al., 2008), automatic attraction toward helpful and unhelpful goals ($\beta_s = -0.05$ to 0.19 , Milyavskaya et al., 2015), and controlled attraction toward helpful goals ($\beta = -0.05$, Milyavskaya et al., 2015). However, controlled motivation was found to be positively related to perception of encountering obstacles ($\beta = 0.28$, Milyavskaya et al., 2015).

Finally, the set of these studies suggest that autonomous motivation would entail a sense of self-control (e.g., TSC) by inhibiting automatically goal-disruptive impulses and implementing helpful strategies. However, the results reveal unclear relationships between controlled motivation and mechanisms of self-control.

Trait Self-Control and Subjective Wellbeing

Does TSC make people happy? A set of studies have provided evidence about the beneficial effects of TSC on positive affect and SWB. The results of De Ridder et al.'s (2012) meta-analysis revealed a positive relationship between TSC and SWB [measured through different types of construct, such as self-esteem, happiness, or absence of depression; $r = 0.33$ (number of used studies = 16)]. In the same vein, authors reported that TSC was positively related to SWB (measured through satisfaction with one's life or happiness, $r_s = 0.24$ – 0.50 , Cheung et al., 2014; Hofmann et al., 2014; Briki et al., 2015), general self-worth ($r = 0.25$, Briki et al., 2015), and positive affect ($r_s = 0.27$ – 0.35 , Hofmann et al., 2014; Briki et al., 2015). With a particular focus on the exercise setting, authors showed a positive relationship ($r = 0.20$) between conscientiousness (reflecting the notion of self-control) and positive affective attitude (measured through different constructs, such as enjoyment, interest, and calmness; Rhodes et al., 2002).

Why does TSC make people happy? To answer that question, it is necessary to understand how TSC can predict goal-directed behavior. Drawing from empirical and theoretical work, Hagger (2013, 2014) proposed a theory delineating three types of pathway by which TSC influences goal-directed behavior (i.e., direct, indirect, and interactive effects). The first pathway (called "P1") corresponds to a direct relationship between TSC and behavior. For example, people with high TSC are more likely to display goal-directed behavior than people with low TSC. The second and third pathways (called "P2" and "P3") correspond to indirect pathways mediated by motivational components. Specifically, P2 reflects an effect on goal-directed behavior mediated by intention, in the sense that people are more likely to develop intentions to reach a goal that promote, in turn, subsequent goal-directed behavior. P3 corresponds to an effect on goal-directed behavior mediated by impulsive motives, indicating that people are more likely to borrow impulsive route to action in which behavior is controlled by more spontaneous processes. The fourth and fifth pathways (called "P4" and "P5") correspond to two interactive pathways in which TSC (and, especially, available resources) moderates the relationship between motivational components and behavior. Specifically, P4 and P5 represent the processes by which self-control resources influence the conversion of intentions and impulsive motives into action, respectively. In sum, people with high TSC would be not only more likely to develop plans for action in order to attain a self-relevant goal (P2), but also more likely to convert such reasoned decisions into action (P4). In addition, they would be less likely to fall under the control of impulses (P3) and more likely to suppress such impulses (P5).

In line with that theory that predicts that TSC stimulates plans for action and their conversion into action (Hagger, 2013, 2014), authors have demonstrated that TSC could foster happiness and SWB by stimulating effective strategies (Cheung et al., 2014). More specifically, Cheung et al. (2014) revealed a positive relationship between TSC and promotion focus (i.e., motivational orientation concerned with growth, advancement, and accomplishment; $r = 0.21$), and a negative relationship

between TSC and prevention focus (i.e., motivational orientation concerned with vigilance, responsibility, and ought; $r = -0.48$). Additionally, they found that both motivational orientations mediated partially the relationship between TSC and happiness, indicating that TSC positively predicted happiness directly and indirectly through increased promotion focus and decreased prevention focus. Furthermore, and consistent with the view that TSC can inhibit goal-disruptive impulses (Hagger, 2013, 2014), authors have revealed that the beneficial effects of TSC on positive affect and SWB might be due to the capability of TSC to manage conflict between competing goals and desires (Hofmann et al., 2012, 2014). More specifically, Hofmann et al. (2014) revealed that TSC was negatively related to problematic desires ($r_s = -0.18$ to -0.20 ; Hofmann et al., 2012, 2014) and that people with high TSC experienced less frequently problematic desires than people with low TSC. Interestingly, Hofmann et al. (2014) also revealed that positive affect (reflecting a momentary psychological state) positively predicted SWB (reflecting satisfaction with life in general), and the authors interpreted this result as the reflection of the tendency of TSC to reinforce consistency within the self, leading to promote positive experiences.

In sum, the set of results suggest that TSC would promote happiness and SWB by activating helpful plans for action and inhibiting problematic desires, leading to increase satisfaction and the likelihood of goal attainment. In that regard, happiness and SWB would be the product of the combination of increased consistency within the self and successful experiences.

The Mediating Role of Trait Self-Control

McCullough and Willoughby (2009) suppose that TSC represents not only the key variable of self-regulation processes, but also one of the most important mediating variables to account for the relationship between motivation and psychosocial outcomes. Several studies conducted in different domains, such as health, education, and leisure, have evidenced such a hypothesis (e.g., Koestner et al., 2008; Briki et al., 2015; Milyavskaya et al., 2015). More specifically, authors examined the mediating role of self-control components (e.g., implementation planning, automatic attraction toward temptations) in the relationship between self-determined motivation and perceptions of obstacles or goal progress (Koestner et al., 2008; Milyavskaya et al., 2015). They observed that autonomous motivation positively predicted goal progress through increased implementation planning, and that the implementation of approach-oriented plans (i.e., intentions to move toward desired outcomes) strengthened that relationship (moderating effect; Koestner et al., 2008). They also observed that autonomous motivation negatively predicted perception of obstacles through decreased attraction toward goal-disruptive temptations (Milyavskaya et al., 2015), and that controlled motivation negatively predicted goal process through increased perception of obstacles (Milyavskaya et al., 2015). Focusing on wellbeing, Briki et al. (2015) have recently examined the relationships between identified religiosity (reflecting autonomous motivation), introjected religiosity (reflecting controlled motivation), TSC, and SWB. The authors showed that autonomous motivation positively predicted SWB directly

and indirectly through enhanced TSC. By contrast, TSC did not mediate the relationship between controlled motivation and SWB, supporting previous results showing an independence relationship between controlled motivation and self-control (Koestner et al., 2008; Milyavskaya et al., 2015).

Research Overview

The present study attempted to examine how MPE, TSC, and SWB might be interrelated, and whether TSC might mediate the relationships of A-MPE and C-MPE with SWB (assessed through the happiness and vitality subscales) in adult regular exercisers. To do so, we built and examined a model using the structural equation model analysis (Figure 1). To assess its quality, different sorts of index were used, such as the goodness-of-fit (GoF) index (e.g., absolute GoF, relative GoF) and the coefficient of determination of the endogenous latent variables (R^2) (e.g., Henseler et al., 2009; Vinzi et al., 2010). The higher the values of GoF indexes and R^2 , the better the model (see the “Analysis” section below). Based on the tenets of the self-determination theory (e.g., Deci and Ryan, 2000, 2008a,b), the control-process theory of self-regulation (e.g., Carver and Scheier, 1990, 1998; McCullough and Willoughby, 2009), and the theory of multiple pathways of TSC (Hagger, 2013, 2014), and consistent with the above-mentioned studies’ results, the present study proposes four sets of hypothesis:

Relationships between MPE and SWB

In line with previous studies showing that A-MPE (or NO-MPE) was positively (or negatively) related to positive psychological outcomes (e.g., SWB) (e.g., Gillison et al., 2006, 2011; Thøgersen-Ntoumani and Ntoumanis, 2006; Sebire et al., 2009), we expect A-MPE (or NO-MPE) to be positively (or negatively) related to SWB. However, because research has found that C-MPE was negatively related or unrelated to positive psychological outcomes (e.g., Thøgersen-Ntoumani and Ntoumanis, 2006), we do not

have any prediction concerning the relationship between C-MPE and SWB.

Relationships between MPE and TSC

Consistent with previous studies showing positive relationships between autonomous motivation and self-control (e.g., Koestner et al., 2008; Briki et al., 2015), we expect A-MPE to be positively related to TSC. However, no clear or consistent evidence allow us to formulate any expectations regarding the relationships between C-MPE and TSC and between NO-MPE and TSC.

Relationship between TSC and SWB

Because research has found that self-control fostered positive affect and SWB (e.g., Cheung et al., 2014; Hofmann et al., 2014), we expect TSC to be positively related to SWB.

Mediations

Consistent with previous studies indicating that self-control-related-variables (e.g., TSC, implementation planning) might mediate the relationship between autonomous motivation and positive perceptions or feelings (Koestner et al., 2008; Briki et al., 2015), we expect TSC to mediate the relationship between A-MPE and SWB. No consistent evidence allow us to formulate any predictions about the mediating role of TSC in the relationships between C-MPE and SWB and between NO-MPE and SWB.

MATERIALS AND METHODS

Participants

Three hundred seventeen adult American volunteers (223 females, 70.3%, and 94 males, 29.7%; $M_{age} = 32.97$, $SD_{age} = 11.30$; $M_{size} = 1.64$ m, $SD_{size} = 0.18$; $M_{weight} = 81.24$ kg, $SD_{weight} = 29.14$), who reported to perform physical exercise regularly (e.g., walking, swimming, biking), were recruited from a popular crowdsourcing on-line platform (ClickWorker¹). On average, the participants reported to perform physical exercise 3.84 times a week ($SD = 1.53$) since 5.70 years ($SD = 7.73$). This sample included participants who were heterogeneous on several qualitative variables, such as socio-demographic variables, (i.e., ethnicity, professional status, familial status), medical variables (i.e., chronic mental and physical disease), and exercise-related variables (i.e., intensity, duration, and mode; Table 1).

Study Design and Procedure

The present study design was submitted and approved by the ethics committee of Qatar University. Each participant provided his/her informed written consent. The setup of this study included a form that was accessible to participants via a specific web address. This form included general information about the study, a consent form, and questionnaires (see the “Measures” section below). Before answering questions, participants were told that: (a) the survey was designed to examine relationships between physical exercise and feelings, (b) they had to perform physical exercise regularly to participate

¹www.clickworker.com

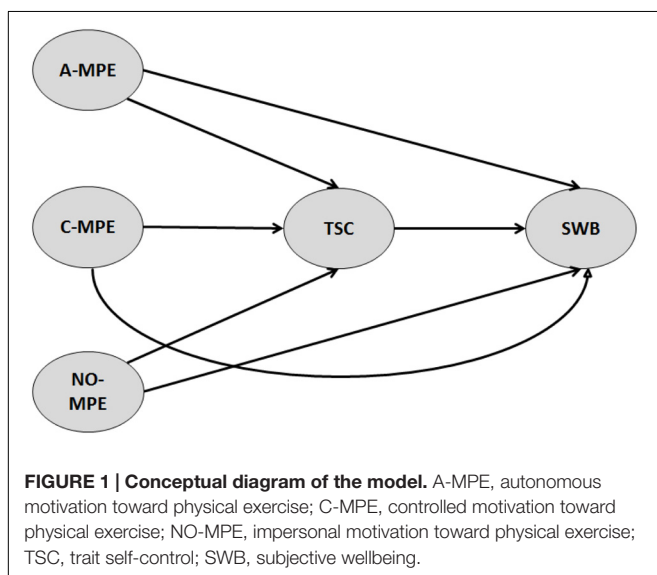


TABLE 1 | Socio-demography and medical situation of participants, as well as characteristics of their physical exercise.

	<i>n</i>	%
Socio-demography:		
Ethnicity:		
African American	58	18.3
Asian American	22	6.9
Caucasian American	203	64
Hispanic American	22	6.9
Other	12	3.8
Familial status:		
Living in family	274	86.4
Professional status:		
Working	104	32.8
Medical situation:		
Physical disease:		
Having a chronic disease	40	12.6
Psychological disease:		
Having a chronic disease	42	13.2
Living in wheelchairs:		
No	317	100
Exercise characteristics:		
Duration:		
0–30°min	101	31.9
31–60°min	167	52.7
61–90°min	39	12.3
91–120°min	10	3.2
Mode:		
Alone	199	62.8
With friends/partner/family	81	25.6
Within a guided program	37	11.7
Intensity:		
Aerobic exercise	236	74.4
Anaerobic exercise	81	25.6

in that survey, and (c) their responses to the survey would be completely anonymous. Thus, the participants should not hesitate to report their honest thoughts and feelings. Then, after approving the consent form and answering questions, participants received a compensation of 0.30 \$ in exchange of their participation in the survey.

Measures

The Behavioral Regulation Exercise Questionnaire-2 (BREQ-2, Markland and Tobin, 2004) was used to measure MPE through the 4-item intrinsic regulation (e.g., “Because I think exercise is fun,” $\alpha = 0.92$), the 3-item identified regulation (e.g., “Because I value the benefits of exercise,” $\alpha = 0.81$), the 4-item introjected regulation (e.g., “Because I feel guilty when I don’t exercise,” $\alpha = 0.75$), the 4-item external regulation (e.g., “Because other people say I should,” $\alpha = 0.85$), and the 4-item amotivation subscales (e.g., “I don’t see why I should have to exercise,” $\alpha = 0.88$). A-MPE was assessed through different subscales, such as intrinsic and identified regulations, while C-MPE was assessed through the subscales of introjected regulation and

external regulation. Impersonal MPE was assessed through the amotivation subscale. The items of BREQ-2 were scored from 1 (“Not true for me”) to 5 (“Very true for me”).

The 13-item questionnaire provided by Tangney et al. (2004) was used to assess TSC (e.g., “I am good at resisting temptation;” $\alpha = 0.76$) on a 7-Lickert scale ranging from “Not at all” (“1”) to 7 “Very much so” (“7”). SWB was assessed through two scales: Happiness and vitality. The 8-item Oxford Happiness Questionnaire (OHQ; Hills and Argyle, 2002) was employed to measure happiness (e.g., “I am well satisfied about everything in my life;” $\alpha = 0.81$; 1 = “Strongly disagree,” 6 = “Strongly agree”), while vitality was measured using the 6-item vitality scale of Bostic et al. (2000), which was a revised version of the vitality scale developed by Ryan and Frederick (1997) (e.g., “I feel alive and vital”; $\alpha = 0.94$; 1 = “Not at all;” 7 = “Very true”).

Analysis

To examine our hypotheses, a two-step analysis was carried out in order to assess the quality of model: Measurement model analysis and structural model analysis. Those assessments were carried out using the PLS (Partial Least Square) structural equation method (XLSTAT-PLS, Addinsoft, version 2016.02.29253). A bootstrapping with 1000 iterations of resampling was conducted.

Measurement Model and Correlation Analyses

A measurement model includes latent and manifest variables. According to Vinzi et al. (2010), a set of manifest variables can be considered as a latent variable when, at least, one of the following conditions is satisfied: (a) the principal component analysis reveals that the first eigenvalue of the correlation matrix is higher than 1, while the other eigenvalues are smaller; (b) the Cronbach’s alpha index (determining the internal consistency) is larger than 0.700; and (c) the Dillon-Goldstein’s rho index (determining the composite reliability of the latent variables) is larger 0.700 (Vinzi et al., 2010). According to the self-determination theory (e.g., Deci and Ryan, 2008a,b), human functioning can be conceptualized through the concepts of autonomous and controlled motivations, which might both endorse the status of latent variable. As a result, the measurement model considered intrinsic and identified regulations as the manifest variables of the latent variable “A-MPE,” while it considered introjected and external regulations as the manifest variables of the latent variable “C-MPE.” Moreover, the model gathered the constructs of happiness and vitality to form an index of SWB. Furthermore, based on the latent variables coming from confirmatory factor analysis, non-parametric (Spearman’s rho) correlations were carried out.

Structural Model and Mediation Analyses

A structural model provides standardized path coefficients (estimated through ordinary least squares regressions) that indicated the strength of the causal relationships. Mediation might be shown when the following criteria are satisfied: (a) the direct relationship between independent and dependent variables (excluding the interaction of the mediator) is significant; (b) the mediator establishes significant relationships with the

independent and dependent variables; and (c) the indirect and total effects (including the interaction of the mediator) are significant. Additionally, the strength of mediation is assessed through variance accounted for (VAF): $VAF = (\text{indirect effect}/\text{total effect})$. The VAF values above 80%, between 20 and 80%, or below 20% indicate that the mediation is full, partial, or non-significant, respectively (Hair et al., 2014). The data were controlled for socio-demography, medical status, and characteristics of exercise as potential confounders (Figure 2).

Estimation of Model Quality

Two sorts of index were used to assess the quality of measurement and structural models: GoF indexes and the coefficient of determination of the endogenous latent variables, R^2 . Firstly, different GoF indexes can be distinguished: Absolute GoF (assessing the overall quality of the measurement and structural models), relative GoF (corresponding to a transformation of the absolute GoF), outer model GoF (assessing the quality of the measurement model), and inner model GoF (assessing the quality of the structural model). A GoF index varies between 0 (model rejection) and 1 (model validation). More specifically, the critical value for the relative GoF, outer model GoF, and

inner model GoF indexes is 0.900. In other words, a value equal to or higher than that threshold for those indexes is considered as satisfying (e.g., Tenenhaus et al., 2005; Vinzi et al., 2010). In addition, a value of the absolute GoF equal to or higher than 0.010, 0.250, or 0.360 reflects a small, moderate, or large overall quality of the both measurement and structural models (Antiocho et al., 2008). Secondly, a structural model can be also assessed through R^2 (e.g., Henseler et al., 2009). More specifically, the R^2 values of 0.19, 0.33, and 0.67 are considered weak, moderate, and substantial, respectively (Chin, 1998).

RESULTS

Factor Analysis and Correlations

The factor analysis yielded three factors: A-MPE, C-MPE, and SWB (Table 2; Figure 2). Non-parametric correlations revealed that the three types of motivation (i.e., A-MPE, C-MPE, and NO-MPE) were all related to each other ($\rho_s = -0.513$ to 0.511 ; Table 3). A-MPE was negatively related to C-MPE ($\rho = -0.233$) and NO-MPE ($\rho = -0.513$), while C-MPE and NO-MPE were

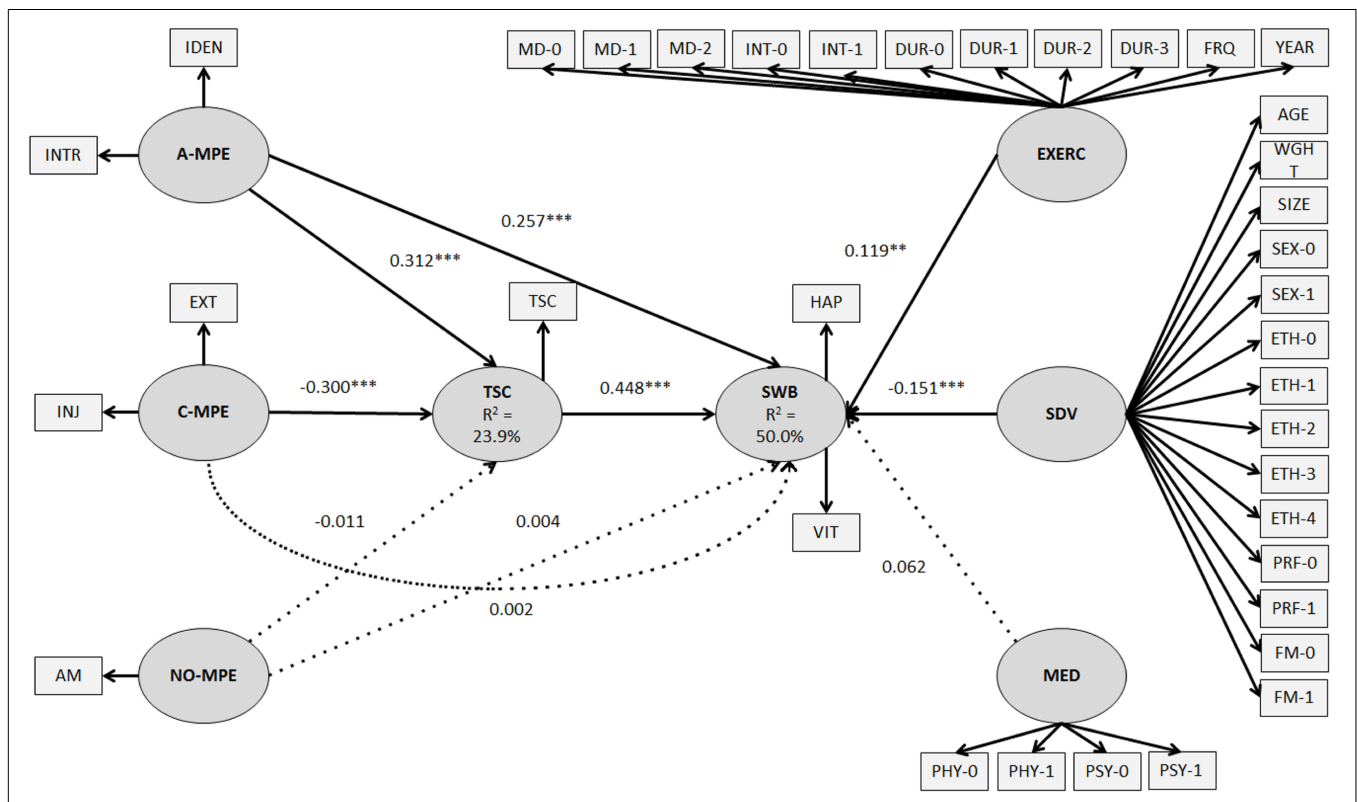


FIGURE 2 | Structural equation model of the relationships among MPE, TSC (as mediator), and SWB (while controlling for socio-demographic, medical, and exercise variables). All coefficients are standardized and solid lines indicate statistical significance. Significance thresholds for a two-tailed test are *** $p < 0.001$, ** $p < 0.01$. A-MPE, autonomous motivation toward physical exercise; INTR, intrinsic regulation; IDEN, identified regulation; C-MPE, controlled motivation toward physical exercise; INJ, introjected regulation; EXT, external regulation; NO-MPE, impersonal motivation toward physical exercise; AM, amotivation; TSC, trait self-control; SWB, subjective wellbeing; HAP, happiness; VIT, vitality; EXERC, exercise-related variables; MD, mode of exercise; INT, intensity of exercise; DUR, duration of exercise; FRQ, frequency of exercise per week; YEAR, number of years of performing exercise regularly; SDV, socio-demographic variables; WGH, weight; ETH, ethnicity; PRF, professional status; FM, familial status; MED, medical variables; PHY, chronic physical disease; PSY, chronic psychological disease.

TABLE 2 | Unidimensionality of manifest variables blocks.

LV Name	# of MVs	Cronbach's α	D.G.'s ρ	PCA eigenvalues
A-MPE	2	0.700	0.869	1.538 0.462
C-MPE	2	0.462	0.788	1.300 0.700
SWB	2	0.866	0.937	1.764 0.236

LV, latent variable; MV, manifest variable; D.G.'s ρ , Dillon-Goldstein's rho; PCA, principal component analysis; A-MPE, autonomous motivation toward physical exercise; C-MPE, controlled motivation toward physical exercise; SWB, subjective wellbeing.

positively related ($\rho = 0.511$; **Table 3**). Furthermore, A-MPE, TSC, and SWB appeared to be all positively related to each other ($\rho_s = 0.382$ – 0.603 ; **Table 3**). C-MPE and NO-MPE were both negatively related to TSC and SWB ($\rho_s = -0.426$ to -0.292 ; **Table 3**).

Structural Equation Model and Mediations

The R^2 value associated with the endogenous latent variables was moderate, being equal to 36.947% (absolute GoF = 0.330; relative GoF = 0.942; outer model GoF = 0.935; inner model GoF = 1.008; **Table 4**; **Figure 2**). Two significant mediations were found: TSC appeared to mediate the relationships between A-MPE and SWB (partial mediation), and between C-MPE and SWB (full mediation; **Table 5**; **Figure 2**). However, TSC did not mediate the relationship between NO-MPE and SWB (**Table 5**; **Figure 2**).

DISCUSSION

The present study utilized the self-determination theory (e.g., Deci and Ryan, 2008a), the control-process theory of self-regulation (e.g., Carver and Scheier, 1998; McCullough and Willoughby, 2009), and the theory of multiple pathways of TSC (e.g., Hagger, 2014) in order to examine how MPE, TSC, and SWB might be interrelated, and whether TSC might mediate the relationships between MPE and SWB.

TABLE 3 | Non-parametric (Spearman's rho) correlations for all latent variables.

Latent variable	1	2	3	4
1. A-MPE	–			
2. C-MPE	–0.233***	–		
3. NO-MPE	–0.513***	0.511***	–	
4. TSC	0.382***	–0.426***	–0.390***	–
5. SWB	0.516***	–0.292***	–0.358***	0.603***

The data of all latent variables come from confirmatory factor analysis. *** $p < 0.001$ for a two-tailed test.

A-MPE, autonomous motivation toward physical exercise; C-MPE, controlled motivation toward physical exercise; NO-MPE, no motivation toward physical exercise; TSC, trait self-control; SWB, subjective wellbeing.

Relationships between Motivation toward Physical Exercise, Trait Self-Control, and Subjective Wellbeing

The analyses revealed that A-MPE was positively related to SWB, whereas C-MPE and NO-MPE were both negatively related to SWB. These results support the predictions of the self-determination theory according to which A-MPE (or C-MPE and NO-MPE) may promote (or hinder) wellbeing (Deci and Ryan, 2008a,b). They also support previous studies that showed that autonomous (or controlled and impersonal) forms of MPE were positively associated with positive (or negative) psychological outcomes (e.g., Edmunds et al., 2006; Gillison et al., 2006, 2011; Sebire et al., 2009). According to the self-determination theory, people may deeply experience wellbeing when their social environment supports their innate needs for competence, relatedness, and especially autonomy. By contrast, when the social environment thwarts their need for autonomy, they are more likely to experience a decreased sense of wellbeing or even depression symptoms.

The analyses revealed that A-MPE was positively related to TSC, supporting the results of previous studies that have revealed positive associations between autonomous motivation and indicators of high self-control, such as TSC (Briki et al., 2015), implementation planning (Koestner et al., 2008), and automatic attraction toward helpful goals (Milyavskaya et al., 2015). This result is also compatible with the studies that have shown that autonomous motivation was negatively associated with indicators of low self-control, such as automatic attraction toward temptations and perception of encountering obstacles (Milyavskaya et al., 2015). Finally, our result supports the general view that autonomous activity would improve the effectiveness of self-regulation processes (e.g., goal selection) because such an engagement in the activity would lie in a strong sense of volition and willingness (Carver and Scheier, 1998; Deci and Ryan, 2000). Furthermore, the analyses revealed that C-MPE was negatively related to TSC, running counter most of the studies that have shown independent relationships between controlled motivation and indicators of high self-control (i.e., TSC, implementation planning, automatic attraction toward helpful goals; Koestner et al., 2008; Briki et al., 2015; Milyavskaya et al., 2015). However, our result is consistent with Milyavskaya et al.'s (2015) study that has revealed a positive association between controlled motivation and low self-control (i.e., perception of encountering obstacles). More generally, our result is compatible with the view that controlled activity would lead to a decreased sense of self-regulation because imposed contingencies driving such a psychological functioning would stay away from the self (Carver and Scheier, 1998; Deci and Ryan, 2000). Taken together, our results support the studies that have revealed that autonomous motivation saved more resources and enabled people to perform better on subsequent tasks than controlled motivation (Moller et al., 2006; Muraven et al., 2007, 2008; Muraven, 2008). Furthermore, NO-MPE was negatively associated with TSC, and this result supports the view of the self-determination theory that NO-MPE would be associated with passivity (the opposite of self-control), anxiety, and depression, caused by the

TABLE 4 | Path estimates of the PLS model.

Effects	Path	β	SE	t-values	p-values	f^2
Direct	A-MPE → SWB	0.453	0.058	7.830	0.000	0.196
	C-MPE → SWB	0.151	0.054	2.804	0.005	0.025
	NO-MPE → SWB	0.021	0.063	0.331	0.741	0.000
Mediating	A-MPE → TSC	0.312	0.060	5.244	0.000	0.088
	C-MPE → TSC	0.300	0.055	5.464	0.000	0.095
	NO-MPE → TSC	0.011	0.064	0.169	0.866	0.000
	TSC → SWB	0.448	0.047	9.477	0.000	0.291
	A-MPE → SWB	0.257	0.052	4.961	0.000	0.080
	C-MPE → SWB	-0.002	0.047	-0.033	0.974	0.000
	NO-MPE → SWB	0.004	0.053	0.082	0.935	0.000

A-MPE, autonomous motivation toward physical exercise; C-MPE, controlled motivation toward physical exercise; NO-MPE, impersonal motivation toward physical exercise; TSC, trait self-control; SWB, subjective wellbeing.

TABLE 5 | Mediation analysis.

Effects	Path	Mediator	IV → Mediator	Mediator → DV	Direct effect	Indirect effect	Total effect	VAF	Mediation strength
Direct without mediator	A-MPE → SWB	N/A	N/A	N/A	0.453***	N/A	N/A	N/A	N/A
	C-MPE → SWB	N/A	N/A	N/A	0.151**	N/A	N/A	N/A	N/A
	NO-MPE → SWB	N/A	N/A	N/A	0.021	N/A	N/A	N/A	N/A
Indirect with mediator	A-MPE → SWB	TSC	0.312***	0.448***	0.257***	0.140***	0.396***	35.2%	Partial
	C-MPE → SWB	TSC	-0.300***		-0.002	0.134***	0.133*	101.2%	Full
	NO-MPE → SWB	TSC	-0.011		0.004	0.005	0.009	N/A	N/A

Significance thresholds for a two-tailed test are *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$. VAF > 80% = Full mediation, 20% ≤ VAF ≤ 80% = Partial mediation, and VAF < 20% = No mediation (none).

IV, independent variable; DV, dependent variable; VAF, variance accounted for; N/A, not applicable; A-MPE, autonomous motivation toward physical exercise; C-MPE, controlled motivation toward physical exercise; NO-MPE, impersonal motivation toward physical exercise; TSC, trait self-control; SWB, subjective wellbeing.

dissatisfaction of the three innate needs (e.g., Deci and Ryan, 2008a,b).

The analyses revealed that TSC was positively related to SWB, supporting the results of previous studies that have reported positive associations between self-control, goal attainment, and satisfaction (e.g., Rhodes et al., 2002; De Ridder et al., 2012; Cheung et al., 2014; Hofmann et al., 2014; Briki et al., 2015). More specifically, research has found that TSC: (a) increased positive affect and SWB (e.g., De Ridder et al., 2012; Hofmann et al., 2014; Briki et al., 2015); (b) inhibited conflict among goals and goal-disruptive impulses (e.g., Hagger, 2014; Hofmann et al., 2014); and (c) stimulated and inhibited helpful (e.g., promotion focus) and unhelpful (e.g., prevention focus) plans for action, respectively (e.g., Cheung et al., 2014; Hagger, 2014). Taken together, those studies suggest the view that TSC would develop a strong sense of SWB by optimizing self-regulation processes and increasing the frequency of positive emotions.

The Mediating Effects of Trait-Self-Control

The analyses revealed two significant mediations. Firstly, TSC appeared to mediate partially the relationship between A-MPE and SWB, supporting the results of Briki et al.'s (2015)

study showing a partial mediation of TSC in the relationship between autonomous religious motivation and SWB. More specifically, our result indicates that A-MPE would increase directly SWB, supporting the view that the satisfaction of people's innate needs would be, by nature, a deep source of wellbeing (e.g., Deci and Ryan, 2008a,b). Our result also indicates that A-MPE would increase indirectly SWB via TSC. This suggests that A-MPE would improve self-regulation processes, thereby leading to facilitate the movement toward the desired goals and to increase the frequency of positive experiences (Koestner et al., 2008; Hofmann et al., 2014; Milyavskaya et al., 2015). Secondly, TSC appeared to mediate fully the relationship between C-MPE and SWB, suggesting that C-MPE would influence SWB only through TSC. In other words, C-MPE would decrease SWB by decreasing the effectiveness of self-regulation processes while pursuing goals (e.g., Deci and Ryan, 2000). Our result runs counter the results of Briki et al.'s (2015) study showing that TSC did not mediate the relationship between controlled religious motivation and SWB, but is compatible with the study of Milyavskaya et al. (2015) showing that self-control mediated the relationship between controlled motivation and goal process. Such inconsistencies should incite psychologists to pay more attention to the link between controlled forms of motivation and the mechanisms of

self-control. Furthermore, TSC did not mediate the relationship between NO-MPE and SWB, and NO-MPE did not predict TSC and SWB. These results suggest that NO-MPE would not activate any regulatory processes. Indeed, NO-MPE reflects a psychological state characterized by the absence of intentionality to behave in the exercise context, thus standing in contrast to A-MPE and C-MPE. More generally, this study supports the views that TSC reflects an effective mediating variable to account for the relationship between commitment to an activity and wellbeing (McCullough and Willoughby, 2009), and that TSC corresponds to a crucial determinant of psychological health (e.g., Tangney et al., 2004).

CONCLUSION AND PERSPECTIVES

The results of the present study are the first to support the hypothesis that MPE can influence SWB through TSC. However, this study is not without limitations. Chief among them is its correlational nature, and thus further studies should use stronger causal tests. To do so, experimental studies should examine the effects of stimuli related to A-MPE (e.g., autonomy-supportive context in exercise settings) and C-MPE (e.g., controlling context in exercise settings) on self-control and SWB. Furthermore, by considering motivation, self-control, and feelings as dynamical processes, time-based designs should be used to examine how MPE, TSC, and SWB may fluctuate over time. In that regard, the use of the ambulatory assessment

methodology (i.e., computerized devices designed to collect self-reported, physiological, and behavioral data in natural contexts) might be helpful to collect data over time within natural contexts (e.g., Kanning et al., 2012). Importantly, such research directions should shed the light on the mediating role of self-control in the MPE-SWB relationships. From an applied standpoint, in order to promote the development of SWB in exercisers, exercise instructors should promote the development of A-MPE since A-MPE appeared to predict SWB (directly as well as indirectly through TSC). To do so, exercise instructors should enhance exercisers' perceptions of autonomy by offering them the opportunity of choice, encouraging them to explore new tasks and techniques, listening to them, limiting controlling self-talk, etc. (Standage and Ryan, 2012). In addition, exercise instructors should always associate exercise with the notion of pleasure.

AUTHOR CONTRIBUTIONS

WB conceived the study, collected and analysed the data, and wrote and revised the article.

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Uncovering the Framework of Brain-Mind-Body in Creative Insight

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THE INTERACTION OF BRAIN-MIND-BODY: THE FUNDAMENTAL IDEA AND SUPPORTIVE EVIDENCE

The TIBMB is rooted in the brain-mind principle in psychology and can be traced back to the central issue of the relation between mind and body in philosophy (Gallagher, 2015). This theory argues that CI can be achieved by the TIBMB and that CI also conversely exerts critical influences on these three agents. Interfering with one of the three subsystems may also partly change the CI process.

The term “mind” *commonly* refers to a collection of mental processes rather than a substance or spirit (Fischbach, 1992), similar to the elements that are manifested when understanding a joke, solving a difficult problem, or experiencing happiness. Of note, rather than being the process of CI itself, the mind *here* refers to the psychological mechanisms underlying CI. One of these mechanisms is restructuring (Ohlsson, 1984; Luo and Knoblich, 2007; Weisberg, 2015). According to representational change theory, CI is achieved by restructuring because of the biased initial problem representation. Consequently, increasing studies demonstrate the importance of restructuring in insight. For example, Öllinger et al. (2014) adopted three versions of the nine-dot problem that can evoke different representations by removing particular sources of difficulty from the standard problem, which demonstrated that three experimental versions of the problem resulted in a higher solution rate than did the standard problem. Representational restructuring/change remains necessary even when the “insight problem” doesn’t involve any spatial structure change, such as brainteaser (Luo and Niki, 2003) or riddles (Fleck and Braun, 2015). Durso et al. (1994) examined restructuring in CI by having participants attempt to solve a riddle that involves a missing piece of information and observed that less misdirected representation easily resulted in CI by shortening the distance between the connections critical to restructuring. Additionally, studies on representational change training also support this role of representational change or restructuring. Patrick and Ahmed (2014), for example, documented that solution rates improved substantially with restructuring training for three different categories of insight problems, and their

facilitation rates were also similar. Other mental mechanisms were also reported to manage insight (see Dietrich and Kanso, 2010); however, these mechanisms are all manifestations of the mind in CI regardless of whether the specific mechanism is representation restructuring.

Early psychologists separated the neural machinery responsible for the mind—the brain (the head)—from the body in the question of mind-body. In psychology, the mind and the brain are two independent variables; however, the mind is a complex phenomenon built on the physical scaffolding of the brain (Libet, 2006), which is a complex, temporally and spatially multi-scale structure that engenders molecular, cellular, and neuronal phenomena that together constitute the neurobiological basis of the mind (Bassett and Gazzaniga, 2011). Consequently, the term *brain* in the TIBMB primarily refers to the brain mechanisms of CI. In the past 20s years, the association between the brain and the mind has been greatly expanded. Many studies indicate that CI is related to the activations in a network of brain regions such as the bilateral prefrontal cortex, the anterior cingulate cortex, the bilateral temporal cortices, the precuneus, the insula, the amygdala, the medial temporal lobe, and the cerebellum (Shen et al., 2013). Although the precise roles of those brain regions in insight have not been entirely determined, substantial evidence across various levels reveals the complexity of insight. These levels include brain lesion investigations and neuroscience studies on healthy subjects using electrophysiological/neuroimaging (see Dietrich and Kanso, 2010) or brain stimulation techniques (Cerruti and Schlaug, 2009; Wei et al., 2014). All of the studies have documented the robust activations in those regions of insight across various tasks, indicating that CI recruits a distributed network encompassing many distant but highly connected brain regions rather than one localized area or a few specific brain regions.

Another entity that may be related to the mind in the question of mind-body is the body “below the head” (hereinafter, the term *body* refers to the body below the head), which in the human species generally comprises a neck, trunk, arms and hands, legs and feet. Beginning in the late 1980s, however, some scientists challenged the view that the body is merely an input-output facility for the brain (Libet, 2006). These scientists argued that instead, higher mental processes are grounded in bodily experience and in the neural systems that govern the body (Carpenter, 2011). The likely link between the body and CI was not recognized until recently, with mounting evidence (Grant and Spivey, 2003; Werner and Raab, 2014) that reveals the embodiment of CI. By contrast to the view of the body as an anchor or understructure of the head, increasingly more studies (Lipnicki and Byrne, 2005; Thomas and Lleras, 2007, 2009a,b) have recently demonstrated that the body may play an equal role, if not a more important role, than the brain in CI. Accumulating evidence indicates that gesturing can hinder or advance creative cognition (Garber and Goldin-Meadow, 2002), with several findings that reliably document the influence of body posture (Friedman and Förster, 2000), gestures (Lipnicki and Byrne, 2005), or bodily movement (Thomas and Lleras, 2009a) on CI. Another line of evidence for this role of body in CI is that attention guiding (Grant and Spivey, 2003; Thomas

and Lleras, 2007, 2009b) or flexible eye shifting (Fleck and Braun, 2015) can facilitate insight. One common method of manipulating attention is requiring participants to perform a secondary task that can guide attentions or elicit various attention patterns by presenting visual stimuli of this secondary task in different display positions (Thomas and Lleras, 2009b). Furthermore, the automatic system acts on cardiac muscles and glands, carrying afferent signals from the vegetative organs to the brain and spinal cord, which regulate what are loosely called the body’s “innards.” Kinds of critical functions like breathing are all controlled by this system (Başar, 2008). This effect of the body on CI has been replicated by studies (e.g., Jausovec and Bakracevic, 1995; Whitehurst et al., 2016) using physiological measurements on skin conduction or cardiovascular responses like heart rate variability. Overall, these lines of evidence indicate the involvement of the body in insight.

Rather than the unidirectional view that the bodily experience as a cause or a result of the insight, it seems more appropriate to take the cycled view that the insight and bodily experience both can be a cause or result of each other. If the bodily response seems to be organized and regular, or in an unchanging status that can promote self-reflective thought, this response is likely an antecedent of the insight; By contrast, if the bodily experience is less organized or only peripheroneural responses like cardiovascular activity, such experience may be a consequence of the insight. Specifically, the solvers’ bodily experience of a swinging is likely an antecedent of an insight to the two-string problem (e.g., Thomas and Lleras, 2009a), which may lead to new bodily responses as a result of this insight, e.g., an increasing heart rate accompanying insight solutions (Jausovec and Bakracevic, 1995). This cycled view stresses the importance of pre-insight bodily experience/status in the insight.

Although the three key elements in the TIBMB have been discussed separately, these elements are not independent influences and generally function as an interactive system because cognitive processes are grounded in bodily experience and in the central neural systems that govern the body (Carpenter, 2011). One example of the interaction of mind and body is that individuals use their bodies to think and that cognitive processes can also result from the manner in which our bodies interact with our immediate surroundings and how “directed actions can guide thought” (Thomas and Lleras, 2009a,b). In addition, some evolutionary evidence implies the interaction of mind and body (Carpenter, 2011). In fact, the brain can be understood as a complex system that is responsible for processes or that modulates the functions of the body. The brain also modulates the cardiac muscles and glands that carry signals from the vegetative organs to the brain and spinal cord (Başar, 2008), in which mental states emerge from interactions on multiple physical and functional levels (Bassett and Gazzaniga, 2011). Meditation, for example, is known to be one method of integrative body-mind training that can improve CI performance (Ding et al., 2015) and that can be linked to the interaction of the central and autonomic nervous systems (Tang et al., 2009), and activity in the default mode network and brain connectivity (Brewer et al., 2011), supporting the TIBMB for CI. Additional support comes from Shen (2014), in which the insight experience,

known to be one defining characteristic of CI (Shen et al., 2016), was revealed to be associated with the triadic interaction of brain-mind-body.

Importantly, the bodily experience should not be only regarded as a proxy of the brain. Although the bodily experience seems difficult to be entirely free of the brain due to the function of central nervous system, reliable evidence was documented that not all bodily responses were generated by these neural activities. In some situations, the bodily experience can still appear even if its corresponding central nervous system is inactive or damaged (see Bechara and Damasio, 2005), implicating the necessity of placing the body in the same position of the brain within the TIBMB. Given that similar interaction frameworks or the brain-mind-body agents might involve in some non-creative/non-insightful cognition, the specificity of the triadic interaction of them in CI should be addressed, which remains open as this field is emerging. Nonetheless, not all cognition involves the body (Irwin, 2000) and the brain and mind agents of the TIBMB are specific to CI, referring to the insight-related brain or psychological mechanisms rather than the brain or mind itself in a broader sense, suggesting the agents are specific to CI.

THE NECESSITY OF SEPARATING THE BRAIN FROM THE ENTIRE BODY

In the traditional view of the mind-body relation, the brain is not separated from but is an essential component of the body (Skinner, 1990) because the brain is incorporated into the body and is essential for the consciousness or the mind. However, mounting evidence has revealed that the brain is the most important substrate of psychological processes. The special significance of the brain renders it necessary for the brain to isolate itself from the entire body, resulting in “the head” and “the body below the head.” The brain-body separation not only stresses the importance of the brain in mind or consciousness

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but is also helpful in illustrating the relation between the central and automatic nervous systems and their specific functions in psychological processes and some physiological phenomena. This isolation is conducive to determining precise neural correlates of types of psychological processes and facilitating CI in a variety of situations and relevant health promotion programs. With increasing interest in embodied cognition, such separation can benefit identifying the role of body components in different psychobiological/biopsychological activities and also manifest the zeitgeist of the embodied approach. Additionally, some neuroscientists have planned to conduct human head transplant experiments involving the brain-body separation (Osborne, 2016). From this perspective, the TIBMB can be considered as a theoretical response to such separation and may offer some implications into one’s post-surgery behavioral/psychological change.

Collectively, we believe that the TIBMB likely play critical roles in triggering CI and studying insight experience is a key avenue validating the framework TIBMB in insight.

AUTHOR CONTRIBUTIONS

WS and YY designed the study and wrote the manuscript. CL and JL provided intellectual input and participated in the discussion. JL, YY, and CL critically revised the manuscript. All authors have read and approved the final manuscript.

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How Mind-Body Practice Works – Integration or Separation?

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In this *Opinion* piece, we take mindfulness as an example of mind-body practice to explore the question of how mind-body practice works based on recent neuroimaging evidence. Mind-body practice encompasses a family of complex practices such as mindfulness meditation, Tai Chi, Yoga, and Qi Gong. Of these practices, mindfulness meditation has received the most attention in the field of psychology and neuroscience over the past two decades. In our recent review, we summarize that mindfulness meditation includes three components that interact closely to constitute a process of enhanced self-regulation: enhanced attention control, improved emotion regulation and altered self-awareness. We also pointed out many people use the term “mindfulness,” but often refer to completely different things or ideas (Tang et al., 2015). In the following sections, we provide examples to briefly clarify the fundamental understanding of the mindfulness concept and how mind-body practice works.

THE CLARIFICATION OF MINDFULNESS

One recent perspective divides mindfulness meditation or intervention into (1) “mindfulness-based stress reduction (MBSR) and related group-based mindfulness interventions such as mindfulness-based cognitive therapy (MBCT), and (2) mindfulness-related interventions such as acceptance and commitment therapy (ACT), dialectical behavior therapy (DBT), cognitive behavioral stress management, and integrative body-mind training (IBMT)” (Creswell, 2017). The confusion is that the phrase “related group-based mindfulness interventions” and the term “mindfulness-related interventions” seem exactly the same. However, even if the author suggests the difference resides in the “group-based” aspect, many other interventions including MBSR and MBCT are also group-based. Furthermore, interventions in (1) are defined as training that foster mindfulness, whereas interventions in (2) are characterized as training that incorporate mindfulness as one component of the program. In reality, this is far from accurate, since MBSR and MBCT also involve multiple components including mindfulness. Therefore, after careful examination of the distinction made by the author, the major difference between (1) and (2) seems to be that the former has the term “mindfulness” in the name of the intervention, hence categorized as mindfulness-based interventions, but the latter does not. To provide a more thorough understanding, we outline below some of the similarities and differences in these interventions as discussed by leading researchers in the field (Kabat-Zinn, 1990; Segal et al., 2002; Davidson and Kabat-Zinn, 2004; Smith, 2004; Linehan, 2014; Tang et al., 2015; Hayes et al., 2016; Tang, 2017).

MBSR has multiple components including mindfulness, yoga exercise, body stretching, group discussion and other components in the program, just like (2) the mindfulness-related interventions mentioned above. For instance, MBSR was described as a “*program that focuses on learning how to mindfully attend to body sensations through the use of body scans, gentle stretching, and yoga mindfulness exercises, along with discussions and practices geared toward*

applying mindful awareness to daily life experiences, including dealing with stress” (Creswell, 2017, p. 495). Therefore, it does not make sense to only characterize MBSR or MBCT as mindfulness intervention, but exclude other mindfulness interventions that simply do not have the term “mindfulness” in their names. Consistent with MBSR developer Kabat-Zinn’s clarification in his book and later articles, there isn’t a pure mindfulness program, and mindfulness intervention such as MBSR incorporates other techniques (Kabat-Zinn, 1990; Davidson et al., 2003; Davidson and Kabat-Zinn, 2004). Smith (2004) also pointed out that “MBSR system is an amalgam of mindfulness meditation, concentrative meditation, passive breathing exercises, yoga stretching, and other components.” Therefore, mindfulness intervention or training works through an integration of several techniques and components rather than a single technique - mindfulness. In the same vein, MBCT developers described the training as a program that draws from cognitive behavior therapy (CBT) and traditional mindfulness practices such as MBSR. By definition, MBCT is a psychological intervention for individuals at risk of depressive relapse (Segal et al., 2002). Clearly, MBCT also incorporates other trainings such as CBT into its program, and it does not make sense to suggest MBCT as a mindfulness intervention, but other similar programs (e.g., ACT, DBT, IBMT) without the term “mindfulness” are not. This clarification is crucial since the misunderstanding of what mindfulness interventions are will mislead the research community and general public on mindfulness and its application, and may create confusion or even bias for people who are interested in research and applied work in this field.

In reality, mindfulness is NOT just a concept or a term, instead it is a direct experience prior to one’s conceptualization *per se*. Without any personal experience of mindfulness, one can only get partial reflection of that experience—perhaps like a blind man touching an elephant. Therefore, the name of a mindfulness meditation or intervention with or without the term “mindfulness” should not define the nature of the program. Instead, the exact components and instructions of mindfulness practice are the key to define the program. Moreover, we need to understand that mindfulness methods always include several components and there is no pure “mindfulness” with only a mindfulness component (Davidson and Kabat-Zinn, 2004; Smith, 2004; Tang, 2017). In the following section, we will provide examples to demonstrate two key components - body-based exercise and mind-based practice in mindfulness program (Tang, 2017).

HOW MIND-BODY PRACTICE WORKS

Speaking more broadly, mind-body practices such as mindfulness meditation shares key components of body-based exercise and mind-based practice. Body-based exercise emphasizes specific posture and body control with a state of relaxed stillness or gentle movement, whereas mind-based practice focuses on self-control of attention, emotion, and thought in a mindful way (Tang et al., 2007, 2015; Hölzel et al., 2011; Tang, 2017). Most studies suggest that mind-based

practice such as mindfulness plays a major role in enhancing the beneficial effects of training. However, recent evidence has shown that body-based exercise also has positive effects on cardiovascular system and central nervous system (CNS), and has been applied to promote health and alleviate chronic diseases (Kerr et al., 2013; Tang and Tang, 2015; Tang, 2017). Taken together, it remains unknown which components or ingredients of mind-body practice play a key role in producing training benefits.

In our series of randomized studies employing one form of mindfulness training - Integrative Body-Mind Training (IBMT), we selected relaxation training (RT) as an active control for two reasons: (1) our goal is not to compare which mindfulness training is the best, instead, we focus on IBMT as an intervention that influences the activity and connectivity of brain areas such as the anterior cingulate cortex which are parts of a self-control network. We assume that other forms of mindfulness training influencing similar brain regions will obtain similar results (Hölzel et al., 2011; Tang et al., 2015); (2) RT control has been widely used as an effective behavioral therapy. Research has shown that RT targets a series of somewhat different brain regions than IBMT. Brain regions affected by RT include sensory motor areas, prefrontal and parietal regions than IBMT (Tang et al., 2007, 2009; Tang, 2017). Since IBMT primarily involves 3 components—body relaxation, mental imagery and mindfulness training, whereas RT mostly involves 2 components—body relaxation (relaxing different muscle groups) and mental imagery (with eyes closed and in a sequential pattern, one concentrates on the sensation and feelings of warmth and heaviness of body), we assume that we could differentiate the effects of mindfulness component by subtracting RT from IBMT. Our previous results indicate that both IBMT and RT produce effects on brain and physiology even after a short-term practice (Tang et al., 2007, 2009, 2010, 2012, 2013, 2015), but they involve different brain areas and thus distinct mechanisms.

In general, the positive effects on behavior, physiology and brain following IBMT are greater than that of RT. At first, these results might seem to suggest that the mindfulness component alone is responsible for these significant differences. However, during our teaching experience when we worked with IBMT participants and applied only the mindfulness component, we could not achieve the similar effects as shown in our series of randomized studies (Tang, 2009), which suggests that IBMT combines different ingredients from the mind-body practices and it is the integration that produces the beneficial effects.

SEVERAL POSSIBLE REASONS OF WHY BRIEF IBMT WORKS EFFECTIVELY

First, IBMT integrates components of both body (including autonomic nervous system) and mind (including central nervous system), which together have shown to have overall positive effects on attention, emotion, and social behaviors (Tang et al., 2015). This combination and integration may amplify the training effects over the use of only one of these components.

Second, everybody can experience mindfulness moment in own life. A qualified coach or trainer can help each practitioner increase this experience and thus ensure that every practice session can achieve a mindfulness state (Tang et al., 2007; Tang, 2017). We view the role of the coach as extremely critical. The coach knows how to interact with the practitioners to achieve the desired mindfulness state. The trainers could well be a part of the effective ingredient in IBMT, and their role requires additional research.

Third, a soft music background integrates the instruction and occupies the practitioner's wandering mind through continuous sensory input, facilitating, and maintaining the mindfulness state. Many training techniques use music background to help beginners. Based on our series of research, we believe that the integration of various techniques into one easy-to-use training package may explain why IBMT is effective at such low dosage although we do not know which of these features is of greatest importance in obtaining changes.

In summary, in this *Opinion* piece, we clarify the misunderstanding of mindfulness-based interventions and

summarize recent mindfulness findings to explore the question of how mind-body practice works. We propose that mind-body practice is a holistic system and works through the integration of different ingredients rather than separated components to achieve the beneficial effects.

AUTHOR CONTRIBUTIONS

All authors listed, have made substantial, direct and intellectual contribution to the work, and approved it for publication.

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Mechanisms of Mind-Body Interaction and Optimal Performance

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Keywords: mind-body interaction, optimal performance, mindfulness meditation, integrative body-mind training (IBMT), anterior cingulate cortex, striatum, central nervous system (CNS), autonomic nervous system (ANS)

Studies have long indicated that effort increases focus on the attentional target and increases distraction inhibition and this type of cognitive control enhances performance (Kahneman, 1973). However, more evidence also shows that a reduction in effortful control can also improve performance, such as in creativity, implicit learning and sensorimotor skills, consistent with the multi-action plan (MAP) model in sport performance (Beilock et al., 2002; Bortoli et al., 2012; Ding et al., 2014a; Stillman et al., 2014; Bertollo et al., 2015, 2016; Amer et al., 2016). We use one form of mindfulness meditation - integrative body-mind training (IBMT) in our series of randomized studies. IBMT emphasizes no effort or less effort to control mind and opening awareness to internal and external stimuli with an attitude of acceptance and equanimity. Our results show that as few as 5 sessions of IBMT (20–30 min per session) can improve attention, positive emotion and diverse cognitive performance including creativity, working memory, conflict resolution and learning (Tang et al., 2007, 2014, 2015; Posner et al., 2010; Ding et al., 2014a,b; Fan et al., 2014, 2015; Tang, 2017). This raises the possibility that less effortful attention or effortless attention can contribute to performance in activities involving creativity, sensorimotor skills or implicit learning.

Based on recent findings, we propose a framework for a relationship among attention, effort and optimal performance, as shown in **Figure 1**. Optimal performance often refers to an effortless and automatic, flow-like state of performance. Mindfulness (mindful acceptance) regulates the focus of attention to optimal focus (balanced attention) on the core component of the action, avoiding too much attention that could be detrimental for elite performance (Bertollo et al., 2016). Balanced attention is a trained state that can optimize any particular attentional activity on the dual-process spectrum. One can exert minimal effort to maintain balanced attention, resulting in a large impact on performance in cognition, positive emotion, health and quality of life. To optimize tasks that require high effort and explicit processing such as working memory, one can reallocate attentional resources, resulting in more efficiently focused attention and less effort. To optimize tasks that require low effort and implicit processing such as creativity or sensorimotor skills, one can bring diffused attention to the task, resulting in more control and monitoring. Through balanced attention, different activities with different cognitive demands can be optimized with a balance of implicit and explicit processing, the appropriate level of attention and effort. Balanced attention has also been called the “being” state (Tang and Posner, 2009, 2014; Tang et al., 2015; Tang, 2017).

What are the underlying mechanisms supporting these distinct processes? Neuroimaging research has suggested that explicit processing with more effort, such as working memory tasks, often recruits the frontoparietal network (Takeuchi et al., 2010; Tang and Posner, 2009, 2014; Ekman et al., 2016; Nissim et al., 2017). The frontoparietal network mainly includes the lateral frontal and parietal cortex and supports continuous effort. It should be noted that it is impossible to maintain a steady and continuous effort because attention states are in constant fluctuation regardless of ongoing task demands (Petersen and Posner, 2012; Tang, 2017; Tang et al., 2017). Studies have shown that attentional lapses lead to poor performance on the task and are associated with midline frontal areas such as anterior cingulate cortex (ACC)

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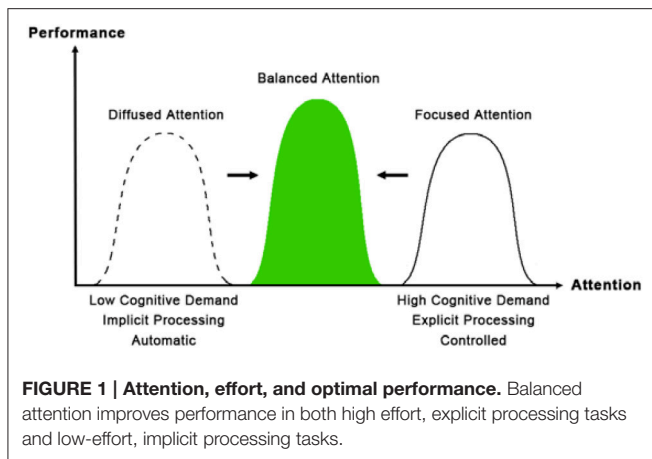
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(Adam et al., 2015; Chang et al., 2015). In contrast, implicit processing with less effort, such as creativity and sensorimotor tasks, also involves the ACC, insula and striatum (Tang and Posner, 2009, 2014; Ding et al., 2014a,b). The ACC is involved in monitoring and maintaining a state by reducing conflict with other states; the insula involves switching between states; and the striatum is associated with the reward experience and habitual responses to make state maintenance easier (Tang et al., 2012, 2015; Tang and Tang, 2013). Meanwhile, the ACC and insula also collaborate to support the role of the autonomic nervous system (ANS) in maintaining the effortless state, which has parasympathetic dominance indexed by lower skin conductance response (SCR) and greater high frequency heart rate variability (HRV) (Tang et al., 2009; Tang, 2017). In contrast, sympathetic dominance more often occurs in effortful processing that requires alertness and activation of the frontoparietal network (Tang et al., 2012, 2009; Tang and Posner, 2014). These findings are consistent with the results of optimal and suboptimal performance in sports (Bertollo et al., 2013, 2015).

Studies have elucidated the interaction and dynamics between the central nervous system (CNS) and ANS (Critchley et al., 2003; Tang and Posner, 2009; Tang et al., 2009; Critchley and Harrison, 2013; Tang, 2017). For example, we examined the brain and physiological changes at rest before, during, and after 5 sessions of IBMT and active control—relaxation training. During and after training, compared to the relaxation control, the IBMT group showed significantly greater parasympathetic activity in each of these measures including heart rate, respiratory amplitude and rate, HRV and SCR. During and after IBMT, differences in HRV and EEG power suggested greater involvement of the ANS. Imaging data showed greater ACC, striatum and insula activity in the IBMT group. Most

importantly, frontal midline ACC theta was also correlated with high-frequency HRV, suggesting control by the ACC over parasympathetic activity (Tang et al., 2009; Tang, 2017). These results indicate that brief IBMT induces better regulation of the ANS by a midline ACC brain system, suggesting the interaction and coordination of body and mind following IBMT, a form of mindfulness that optimizes activities for maximal self-control, attention and efficiency with minimal effort (Tang, 2017). Other studies have shown that parasympathetic activity is associated with the flow state (de Manzano et al., 2010; Keller et al., 2011; Thomson and Jaque, 2011; Jacobs, 2014), a prime example of balanced attention in which high control is achieved with low subjective mental effort (Bruya, 2010). We call this mechanism “parasympathetic-attentional interaction” or “PA mind-body interaction.”

In summary, growing empirical evidence indicates that PA mind-body interaction often triggers optimal performance and is one possible mechanism for optimizing performance (Tang and Posner, 2009, 2014; Bruya, 2010; Tang et al., 2012; Tang, 2017). PA mind-body interaction can also have a large impact on positive emotion, health benefits, quality of life and self-growth. The field of body-mind practice is rapidly growing. However the majority of research focuses on health and behavior effects (and related brain changes) from training (Lutz et al., 2008; Tang et al., 2015). There has been less effort to scientifically investigate the underlying mechanisms (e.g., key biomarkers) of mind-body interaction and optimal performance when practitioners engage and maintain an effortless state. The current perspective aims to address this research gap. By integrating evidence from neuroimaging with evidence from physiology we propose the key brain markers in the ACC-insula-striatum network and the key physiological markers in the parasympathetic regulation of HRV and SCR. This effort will also shed light on how humans learn and practice physical and mental training effectively. Future studies can examine the relationship between PA mind-body interaction and short-term or long-term training such as mindfulness and its underlying mechanisms, using psychosocial, physiological, multimodal neuroimaging, and genetic methods.

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