Research Article

Gait-combined closed-loop brain stimulation can improve walking dynamics in Parkinsonian gait disturbances: A randomized-control trial

Ippei Nojima [1)], Mitsuya Horiba [2)], Kento Sahashi [2)], Satoko Koganemaru [3)], Satona Murakami [2)], Kiminori Aoyama [2)], Noriyuki Matsukawa [4)], Yumie Ono [5)], Tasuya Mima [6)], Yoshino Ueki [2)]

Authors' affiliations

1) Department of Physical Therapy, School of Health Science, Shinshu University, Japan

2) Department of Rehabilitation Medicine, Nagoya City University Graduate School of Medical Sciences, Japan

3) Department of Regenerative Systems Neuroscience, Human Brain Research Center, The Graduate School of Medicine, Kyoto University, Japan

4) Department of Neurology, Nagoya City University Graduate School of Medical Sciences, Japan

5) Department of Electronics and Bioinformatics, School of Science and Technology, Meiji University, Japan

6) The Graduate School of Core Ethics and Frontier Sciences, Ritsumeikan University, Japan

Corresponding author

The Graduate School of Core Ethics and Frontier Sciences, Ritsumeikan University, Japan

E-mail: t-mima@fc.ritsumei.ac.jp

Tasuya Mima

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Conflict of Interest

Notion to report.

Author Contributions

All authors contributed to the study concept and design; I.N, M.H, Y.U, and T.M contributed to the acquisition and analysis of data; all authors contributed to drafting the manuscript and preparing the figures.

ABSTRACT

Objective: Gait disturbance lowers activities of daily living in patients with Parkinson's disease (PD) and related disorders. However, the effectiveness of pharmacological, surgical, and rehabilitative treatments is limited. We recently developed a novel neuromodulation approach using gait-combined closed-loop transcranial electrical stimulation (tES) for healthy volunteers and post-stroke patients, and achieved significant entrainment of gait rhythm and an increase in gait speed. Here, we tested the efficacy of this intervention in patients with Parkinsonian gait disturbances.

Methods: Twenty-three patients were randomly assigned to a real intervention group using gait-combined closed-loop oscillatory tES over the cerebellum at the frequency of individualized comfortable gait rhythm, and to a sham control group.

Results: Ten intervention sessions were completed for all patients and showed that the gait speed ($F_{(1, 21)}$=13.0, p=0.002) and stride length ($F_{(1, 21)}$=8.9, p=0.007) were significantly increased after tES, but not after sham stimulation. Moreover, gait symmetry measured by swing phase time ($F_{(1, 21)}$=11.9, p=0.002) and subjective feelings about freezing ($F_{(1, 21)}$=14.9, p=0.001) were significantly improved during gait.

Interpretation: These findings showed that gait-combined closed-loop tES over the cerebellum improved Parkinsonian gait disturbances, possibly through the modulation of brain networks generating gait rhythms. This new non-pharmacological and non-invasive intervention could be a breakthrough in restoring gait function in patients with PD and related disorders.

Keywords: closed-loop rhythmic electrical stimulation; non-invasive brain stimulation; Parkinson’s disease; walking rehabilitation

Key messages

- What is already known on this topic

Non-invasive brain stimulation could improve physical performance for patients with Parkinson's disease.

- What this study adds

A transcranial electrical stimulation for the cerebellum being synchronized in gait rhythm for successive 10 repetitions of the interventions could have an effect of improving gait function for PD.

- How this study might affect research, practice or policy

Closed-loop brain stimulation combined with an individual rhythm might be one of novel approaches to help to improve physical function for patients with disabilities.

INTRODUCTION

Parkinson's disease (PD) is a neurodegenerative disease that leads to a progressive decline in motor function, including signs of akinesia, rigidity, tremor, postural instability, and gait disorders caused by dysfunction of the nigro-striatal pathway. The increased GABAergic signaling from the output nuclei of the basal ganglia to the subcortical structures decreases the excitatory signaling from the thalamus to various cortical areas, leading to widespread cortical dysfunction, including the motor network [1]. Gait disturbance is particularly important among various movement disorders because it adversely affects quality of life. In addition to hesitancy, shuffling and short steps, freezing and motor blocks, balance deficits, and frequent falls occur during the later stages of PD. Dopamine medications and deep brain stimulation (DBS), which are widely used for PD, are less effective for postural instability, gait disturbance, and freezing, compared to akinesia, rigidity, and tremor [2].

Since these gait disturbances are associated with an impaired cortico-basal ganglia-thalamic network, neurologists often encounter similar Parkinsonian gait in neurological disorders other than PD. Unfortunately, Parkinsonian gait in non-PD disorders is usually resistant to the standard treatment for idiopathic PD. Thus, there is a clinical need for the development of non-pharmacological and non-invasive treatment strategies to improve Parkinsonian gait disturbances, such as transcranial direct current stimulation (tDCS), which can induce neural plasticity at the bedside. Two recent systematic reviews have reported that tDCS improved motor functions in patients with PD [3, 4], which might be

mediated by the modulation of local intracortical circuits and large-scale cortico-basal ganglia-thalamic circuits [5, 6].

Recently, we found that oscillatory transcranial electrical stimulation (tES) to the primary motor cortex (M1) synchronized with individual gait rhythm, or gait-combined closed-loop stimulation, could facilitate gait in healthy subjects [7] and patients with stroke [8], possibly through the entrainment of cortical oscillatory activity coupled to the gait cycle [9]. Moreover, a recent review suggested that tDCS may be a promising complementary approach for neurological disease [3].

Although multiple brain regions contribute to gait, the cerebellum can be an appropriate target for tES intervention in Parkinsonian gait because it primarily affects the spinal locomotor networks through its descending drive and rhythmic bursts, leading to repetitive rhythmic step cycles in animal studies [10]. In human bipedal gait, locomotor regions, including the cerebellum, are activated during both actual and imaginary walking [11]. In patients with PD, gait-induced activation was reduced in the anterior cerebellum, and the improvement of gait function by visual cues is associated with an increase in cerebellar activation [12], suggesting that cerebellar modulation could improve Parkinsonian gait disturbance.

In this pilot study, we investigated whether a personalized gait-combined closed-loop brain stimulation method can improve Parkinsonian gait disturbance using a randomized controlled design. Since the putative effect of our intervention is not necessarily dopamine replacement per se, but the

modulation of the walking-related cortico-basal ganglia-thalamic network, both idiopathic PD and non-PD patients with Parkinsonian gait were enrolled in this study.

SUBJECTS/MATERIALS AND METHODS

1. Participants

Twenty-three patients diagnosed with PD or Parkinson's syndrome were recruited in this sham-controlled study from the Department of Neurology of Nagoya City Hospital. A blinded neurologist assessed each patient's demographic, clinical, and cognitive features using the Unified Parkinson's Disease Rating Scale (UPDRS).

The most common cause of Parkinsonism is idiopathic PD; however, Parkinsonism is not specific to PD but is prevalent in other neurological disorders, including progressive corticobasal syndrome (CBS), multiple system atrophy (MSA), and vascular Parkinsonism (VP). Therefore, participants in this study were selected based on the Yamaguchi criteria [13] for VP due to cardiovascular disease (CVD), Armstrong criteria [14] for CBS, NINDS-SPSP criteria [15] for Progressive supranuclear palsy (PSP) , and the revised Gilman criteria [16] for Spinocerebellar degeneration (SCD).

Inclusion criteria were as follows: 1) walking ability for more than 6 min without using the device and 2) age ≥ 40 years. Exclusion criteria were: 1) severe dyskinesia or "on-off" fluctuations; 2) need for assistance in activities of daily living; 3) severe motor disability due to other neurological or orthopedic diseases; 3) important cognitive deficit (Mini-Mental State Examination [MMSE] < 23); 4) no history of other neurological or psychiatric diseases; 5) no present pregnancy; and 6) no cardiac pacemaker and no previous surgery involving implants (aneurysm clips or brain or spinal electrodes).

Patients who did not satisfy the inclusion criteria described above and those who could not understand the study procedure were excluded. All participants were classified using the Hoehn and Yahr disease rating scale (HY) and examined according to the motor section of the Unified Parkinson's Disease Rating Scale (UPDRSIII). In addition, freezing of gait (FOG) was evaluated using the FOG questionnaire, and global cognitive impairment was assessed using the MMSE. The FOG-Q is a validated tool for the identification of the freezing of gait, and we administered it on the day of pre- and post-intervention assessments to evaluate patients' current FOG status.

The sample size was estimated to have a power of more than 80% to detect mean group differences using two-way analysis of variance (ANOVA). To determine the sample size requirements for our study, we adopted a conservative effect size of eta squared ($\eta 2$) = 0.8 to 0.1 as a moderate effect, and with a significance level of alpha (α) = 0.05, a total sample size ranging from 20 to 26 patients was determined to provide sufficient power. We used the Consolidated Standards of Reporting Trials (CONSORT) checklist to confirm the guidelines. All participants underwent functional evaluations and gait analysis after providing informed consent to participate in this study, according to the Nagoya City University Hospital Trust Ethics Committee (jRCTs042190007). The experimental procedure conformed to the Ethics Committee of the World Medical Association (Declaration of Helsinki) and was approved by the University Hospital Medical Information Network in Japan.

2. Experimental procedures

All participants were assigned to two interventional groups: (1) patterned tES on the cerebellum of the severe side (tES group) and (2) sham stimulation (sham group) during gait training. A blinded experimenter randomly assigned patients using “the RAND” function in Excel software (Microsoft Office). Both interventions were executed in four sets for 4 min with an intertrain interval of 3 min, twice per week for five weeks (total 10 sessions). Participants were blinded to the condition of the intervention.

On the day before the intervention commencement, stride time was assessed using a sheet-type pressure sensor (2.4-m long; Walk Way, Anima Corporation, Tokyo, Japan) placed in the middle of a 10-m walkway over 30 steps at a preferred speed to determine gait cycle frequency. Gait cycle assessment was set at the time of good physical condition after taking medication. The frequency of patterned tES was applied at the nearest to the pre-measured gait frequency on the severe side with a 0.01 Hz bin in each group. The frequency was different in each subject and remained constant throughout the total 16 min stimulation period (Intervention (4 min*4 sets). During the gait intervention, the participants walked at their own comfortable pace for a total of 16 minutes in either the tES or sham condition on a 135-meter corridor. In the tES group, the stimulus frequency was continuously applied based on the step counts measured during comfortable gait prior to the intervention in each trial.

To check for adverse events or reactions, patients were asked to report if they felt any unusual sensation before, during, and after the experiments. The skin was examined by a physician every day. We asked the participants to continue the same daily physical activity as they had been doing before the intervention started, and we checked it every session.

3. Cerebellar patterned tES intervention

A detailed description of the patterned tES intervention synchronized with the gait cycles applied in this study has already been provided in previous studies by our group with healthy and stroke patients [7, 8, 17, 18] (Fig.1). The electrical currents for tES with a constant positive DC offset were delivered using a DC stimulator Plus (NeuroConn, Ilmenau, Germany). This DC offset was set because using patterned tES with a constant positive DC offset increased cortical excitability and was sustained for more than 20 min [9]. The electrical current waveform was a sinusoidal wave of 2 mA (from 0 to 2 mA) peak-to-peak amplitude with the cycle length fit to the gait cycle of comfortable pace for each individual patient on medication. One cycle of current (rising from 0 to 2 mA and falling from 2 to 0 mA) was started at the moment of foot contact on the severe symptom side, which was digitally detected by pressure sensors (PH-450A, FS amplifier, DKH Co., Ltd., Japan) attached to the bilateral heels during gait. The intervention consisted of 4 min of gait training and 3 min of rest as one block, with a total of four blocks. The closed-loop system that we developed allowed us to dynamically adjust the phase of the

tES to the online detected foot contact timing, despite the fluctuating and unstable gait rhythm of the patients.

For stimulation over the cerebellum on the severe side, the electrode (5 × 5 cm) was centered 3 cm right/left-lateral from the inion, a position that spans the cerebellum. The reference electrode (5 × 5 cm) was placed on the opposite side of the posterior neck. The electrical currents were faded in and out for 60 s, with the electrodes placed in the positions used for the patterned tES. The same procedure was used in the sham group, but the patterned tES current was applied for only the first ten gait cycles, with electrodes positioned on the cerebellum on the severe side. Regarding the role of external cues by the patterned tES, a previous study [7], which confirmed whether subjects felt the rhythmicity of the tES currents, showed that the subjects could not perceive the rhythmicity of this intervention.

4. Data analysis

Motor function assessments, including the UPDRS part III and H-Y scale, and gait function assessments were performed by experimenters blinded to the stimulation type. To evaluate the change in gait function following the intervention, several gait parameters on the more impaired side were assessed using a sheet-type pressure sensor, operating at a sampling frequency of 100 Hz, including gait speed, swing phase time, stance phase time, and stride length. In addition, a symmetry index (SI)

based on gait cycle time on both legs was calculated. A stance or swing time symmetry were calculated according to the following equation [19].

$$Symmetry\ Index\ (SI) = \frac{Longer\ stance\ /\ swing\ time}{(Longer + Shorter\ stance\ /\ swing\ time)}$$

For each measure, a value of 0.5 reflect perfect symmetry. The pressure-sensitive carpet system recorded the temporal and spatial gait cycle parameters as the subject walked on the carpet. The participants were instructed to walk along the walkway at a comfortable pace. They repeated the the10-m walk two times, and the average parameters were calculated. Pre- and post-assessments were conducted at the beginning and end days of the intervention. The FOG-Q total score ranges from 0 to 24, with higher scores corresponding to more severe FOG. This questionnaire was also asked by the same neurologist at the beginning and end of the intervention to assess the state of freezing gait. In addition, unblinded investigators performed a short clinical assessment to monitor the safety of tES.

5. Statistical Analysis

For demographic and clinical characteristics, the Student's t-test and Mann-Whitney U test were used to examine baseline clinical characteristic data between the tES and control intervention groups. Descriptive statistics are reported as means and standard deviations (SDs).

To evaluate the effects of the tES intervention on gait function, we performed a linear mixed model ANOVAs to test the factors of interventional conditions (tES vs. sham) and time (pre vs. post)

on gait, including gait speed, stance phase time, stance phase time, and stride length on the severe side. In addition, we also investigated gait symmetries in stance time and swing phase using the SI. We chose a linear mixed model because we wanted to investigate the effects of tES on gait function for each group by removing errors, which were a different length of gait training for every participant. Random intercepts and fixed slopes were used for each participant in the mixed-effects model. In addition, a linear mixed model analysis was used to test the effect on freezing of gait assessed by FOG-Q with factors of interventional conditions (tES vs. sham) and time (pre vs. post). In the secondary measurement, intergroup comparisons were assessed using Cohen's d for the change ratio to calculate the effect size, which is equivalent to the z-score of a standard normal distribution. The effect size estimation was corrected using the Hedges' correlation. Greenhouse-Geisser corrected the degrees of freedom that were used to correct for violations of the assumption of sphericity. Bonferroni procedures were used to correct for multiple comparisons in the post hoc analysis of gait function. A paired t-test was used to examine differences in motor function between the two intervention groups. Group comparisons of clinical and gait characteristics and significant changes in gait were considered significant at $p < 0.05$. All other comparisons obtained from model-based contrasts were secondary. Statistical significance was set at $p < 0.05$. Statistical analyses were performed using the R studio (ver. 3.6.1).

In addition, we performed a similar subanalysis using only patients with idiopathic PD.

Results

There were no dropouts, and the compliance of both groups was good and comparable. There were no reports of phosphenes, vertigo, or skin irritation from stimulation. Participant characteristics, including age (p = .753), sex (p = .867), duration from onset (p = .740), and MMSE (p = .790), are presented in Table 1, and there was no significant difference between the intervention groups. The UPDRS motor scores (p = .419), H-Y score (p = .559), and FOG-Q at baseline (p = .189) were not significantly different. Behavioral and neurophysiological assessments were performed by a neurologist in the same manner before and after the intervention.

There was a significant main effect of time ($F_{1,21}$ = 12.63, p = .002) and an interaction ($F_{1,21}$ = 12.99, p = .002) for the speed of the self-paced walk (Fig. 1), but no significant main effect for condition ($F_{1,21}$ = 0.07, p = .799). Post hoc analysis revealed that gait speed was significantly faster after the real intervention than after the sham intervention (p < .001, Hedges's g = 1.450). (Fig.2).

For swing phase time on the severe side, the linear mixed model measure ANOVAs showed a significant main effect of time ($F_{1,21}$ = 6.48, p = .019) and interaction ($F_{1,21}$ = 11.90, p = .002), but no significant main effects for condition ($F_{1,21}$ = 0.55, p = .467). Post hoc analysis revealed that the swing phase time on the severe side was significantly longer after the real intervention than after the sham intervention (p = .002, Hedges's g = 1.388). There was also a significant interaction ($F_{1,21}$ = 13.10, p = .002) for stance phase time on the severe side, but no significant main effects for time (F

$_{1,21}$ = 2.74, p = .112) and condition ($F_{1,21}$ = 1.42, p = .247). Post hoc analysis revealed that the stance phase time was significantly shorter after the real intervention than after the sham intervention (p = .01, Hedges's g = -1.053) (Fig. 2). In addition, there were also significant main effects for time ($F_{1,21}$ = 7.76, p = .011) and interaction ($F_{1,21}$ = 5.82, p = .003) for a SI of swing time, but not significant main effects for condition ($F_{1,21}$ = 2.07, p = .165). Post hoc analysis revealed that the SI of swing time was symmetry after the real intervention (p = .001, Hedges's g = 1.007).

Furthermore, for stride length on the severe side, the linear mixed model ANOVAs also showed a significant main effect of time ($F_{1,21}$ = 12.91, p = .002) and interaction ($F_{1,21}$ = 8.89, p = .01), but no significant main effects for condition ($F_{1,21}$ = 0.19, p = .665). Post hoc analysis revealed that stride length on the severe side was significantly longer after the real intervention than after the sham intervention (p = .001, Hedges's g = 1.200) (Fig. 2).

For the FOG-Q score, the linear mixed model ANOVAs showed a significant main effect of time ($F_{1,21}$ = 14.35, p = .001) and interaction ($F_{1,21}$ = 14.93, p < .001), but no significant main effects for condition ($F_{1,21}$ = 0.64, p =.433). Post hoc analysis revealed that FOG-Q was significantly decreased after the real intervention compared to the sham intervention (p < .001, Hedges's g = -1.555) (Fig.3). According to the change in gait parameters following interventions, effect sizes were statistically significant, and differences were high; Hedges' g values range from 0.96 to 1.30.

Regardless of the improvement in gait parameters by the real intervention, UPDRS III scores

(tremor, rigidity, and bradykinesia) were not significantly changed after the interventions.

Additionally, we performed subgroup analysis only for idiopathic PD patients (n = 15, 7 participants were in the real group and 8 participants were in the sham group). There was a significant main effect of time ($F_{1,14} = 6.96$, $p = .02$) and an interaction effect ($F_{1,14} = 12.62$, $p = .003$) on the speed of the self-paced walk. Post hoc analysis showed that gait speed in the real intervention group was significantly faster after the intervention ($p = .003$, Hedges's g = 1.602). For the swing phase time on the severe side, there was a significant main effect of time ($F_{1,14} = 5.51$, $p = .034$) and interaction ($F_{1,14} = 18.99$, $p < .001$). Post hoc analysis showed that the swing phase time on the severe side for the real intervention group was significantly longer after the intervention ($p = .002$, Hedges's g = 1.985). There was also a significant interaction effect ($F_{1,14} = 5.31$, $p = .037$) for stance phase time on the severe side, but no significant main effects for time ($F_{1,14} = 3.45$, $p = .08$) and condition ($F_{1,14} = 0.36$, $p = .563$). Post hoc analysis showed no significant difference (p = .062, Hedges's g = -1.038). There were no significant differences in the stride length on the severe side. For the FOG-Q score in idiopathic PD patients, it showed a significant main effect of time ($F_{1,14} = 5.82$, $p = .030$) and interaction ($F_{1,14} = 14.87$, $p = .002$). Post hoc analysis revealed that FOG-Q was significantly decreased after the real intervention compared to the sham intervention ($p = .002$, Hedges's g = 0.865).

DISCUSSION

This study aimed to evaluate the effect of individualized gait-combined closed-loop tES over the cerebellum on Parkinsonian gait disturbance. The patients were randomly assigned to real and sham intervention programs. The real intervention group showed a significant improvement in gait parameters, including speed, gait symmetry, and stride length, on the severe side of symptoms after 10 repetitions of the intervention. Regarding the change of temporal symmetry for gait by intervention, the effect on swing phase time has led to improve of gait asymmetry. In addition, FOG questionnaire scores significantly improved after the intervention. These findings suggest that the present brain stimulation, whose pattern matches the individual gait cycle in terms of frequency and phase, could achieve functional recovery of Parkinsonian gait and might be used as an add-on therapy for gait rehabilitation in the future.

Several studies suggested that applying sinusoidal currents simultaneously to many neurons could modulate oscillatory network dynamics in a frequency-specific manner [20, 21] even if the externally applied current is small. Furthermore, neural entrainment may be a generic way in which electric fields can affect neuronal networks. With regard to oscillatory tES in human subjects, it has reported superior efficacy for memory function compared to traditional tDCS [22]. Thus, our gait-combined closed-loop tES system might be a suitable way to interact with endogenous gait-related oscillations in the brain by driving stimulation at the individualized frequency imposed to induce

synchronization between the tES and brain network, which might be associated with alternating de- and hyperpolarization of membrane potentials at a given frequency [21]. A recent study reported modulation of human M1 excitability via cerebello-cortical connectivity by stimulating the cerebellum [23], also supporting our hypothesis that cerebellar tES can modulate the cortical-basal ganglia-thalamic network.

The functional role of the cerebellum in gait control is thought to provide a rhythmic pattern and contribute to speeding modifications for supraspinal control of locomotion. The cerebellar locomotor region (CLR), which lies at the midline of the cerebellar white matter, is an important region in the hierarchical network of the supraspinal locomotion center. Therefore, activation of this region by electric stimulation could induce rhythmic output in experimental animals, and the cerebellum integrates information from higher and lower brain centers to produce precise coordination of ongoing locomotion [10]. Moreover, it has also been reported that electrical stimulation of the output fibers of the fastigial nucleus [24], which is strongly influenced by the cerebellar vermis, results in augmentation of the postural muscle tone of cats.

The cerebral-cerebellar interaction is based on multiple closed-loop circuits, while anatomical studies have suggested that the dentate nucleus projects to the striatum and that the subthalamic nucleus of the basal ganglia projects to the cerebellar cortex [25]. Neuroimaging study has demonstrated increased activation in the cerebellum of patients with PD during motor execution [26],

during the motor learning process [27], and in the resting state. Thus, it has been suggested that the functional role of increased activity or connectivity in the cerebello-thalamo-cortical loop in PD could compensate for hypofunction in the striato-thalamo-cortical circuit [26]. Given this complimentary balance between the cerebellum and basal ganglia, it is likely that tES intervention over the cerebellum, as in our intervention strategy, might be especially useful for improving Parkinsonian gait. Our technique using the present study supports the effect of closed-loop feedback systems similar to those already implemented in animal studies, in which the stimulation waveform is dynamically adjusted to suppress ongoing pathological activities [28, 29]. It is possible that the symmetry index could have been improved in the current study, given the crucial role of the cerebellum in balance control, although it has been reported that temporal asymmetries in gait patterns are more difficult to change than spatial asymmetries [30]. These findings have shown that closed-loop intervention might have a potential role in treating neurological disorders and can be used to rebalance activity in abnormally functioning neural circuits.

In the present study, FOG, which is a very disabling paroxysmal symptom affecting over half of patients with PD, was also significantly improved by tES intervention. FOG clinically presents substantial variability within and between patients; therefore, the patterned tES synchronized to personalized gait rhythm could be an appropriate strategy for ameliorating FOG. A recent review also suggested that PD patients with FOG may benefit from a future on-demand treatment system [31], and

this novel intervention has shown similar effects to DBS or medication for FOG.

Imaging studies in PD with FOG showed neural disruption of the pedunculopontine nucleus (PPN) and altered white matter connectivity in the corticopontine and pontine-cerebellar tracts [32]. The pedunculopontine nucleus (PPN) is part of the mesencephalic locomotor region (MLR) in the upper brainstem. The MLR anatomically has connections to the basal ganglia and cerebellum and therefore is an essential point of interaction in the locomotor network for motor information out of the basal ganglia and cerebellar loops. Thus, it has been reported that DBS of the PPN could improve gait disturbances, and DBS over the unilateral PPN intensifies cerebral blood flow bilaterally into the central thalamus and cerebellum [33]. Moreover, an advanced non-invasive method using magnetic resonance-guided focused ultrasound ablation has recently been reported to be well tolerated and to improve motor function [34]. However, the direct effects of this novel intervention on gait function remain unknown. Improvement of gait function, including FOG, induced by our gait-combined closed-loop tES might be caused by modulation of the functional connectivity between the PPN and cerebellum.

Recent systematic reviews have found that tDCS improves motor function in PD [3, 4]. One small randomized controlled trial (RCT) with a sample of 10 patients demonstrated a positive effect on gait, FOG, and motor performance after five sessions of anodal tDCS over M1 [35]. Costa-Ribeiro et al. also reported that tDCS over the motor-related areas combined with cueing gait training can lead

to prolonged improvements in PD patients [5]. Moreover, physical training combined with tDCS over M1 or the dorsolateral prefrontal cortex has been shown to produce more significant improvements in gait, balance [36], and cognitive function [37] in PD patients than physical training alone. In contrast, repetitive administration of anodal tDCS over bilateral M1 has been found to improve levodopa-induced dyskinesias, but not other motor symptoms [38]. The cerebellum is also being increasingly considered as a potential target for tDCS due to its involvement in a range of conditions, including cerebellar ataxia, PD, and dystonia [6]. Workman et al. has reported that cerebellar tDCS at a high stimulus intensity (bilateral 4mA) did not improve gait despite improvement in balance function in patients with PD [39]. However, these previous studies employed independent tDCS and motor rehabilitation interventions, making it difficult to compare directly with our closed-loop tES system synchronized with gait rhythm. Overall, there is a lack of clear evidence on the effectiveness of tDCS for gait disturbance, and there is a need to develop personalized tDCS approaches and optimize its clinical use for gait rehabilitation.

It is possible that the interaction between dysfunction of the dopamine system in patients and dopamine replacement might modify the effects of intervention in a complex way because neuroplasticity is significantly affected by dopamine [40].

Several potential limitations should be noted. First, the study included patients with various neurological diseases, leading to a high degree of clinical heterogeneity and limiting the

generalizability of the results. Second, while the severity of freezing of gait (FOG) was assessed using the FOG-Q questionnaire, which is based on patients' subjective judgment, an objective assessment such as the observation of FOG events may provide a more comprehensive understanding of FOG status. Third, the entrainment of tES with the gait cycle may enhance cortico-spinal excitability and modulate cortical control of muscle activity during gait, but the optimal oscillatory brain stimulation combined with gait has yet been evaluated (e.g., the effects of different types and methods of brain stimulation such as tDCS). Fourth, this study did not consider other aspects of gait, such as standing balance and body coordination measures that may also be sensitive indicators of neurological disorders. Additionally, spatial information during gaits, including kinematics and kinetics, could have provided further insight into the effect of the intervention. Finally, while we did not compare ON/OFF medication states, it is possible that some effects attributed to closed-loop stimulation may be related to medication.

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TABLES

Table 1. Baseline patients' characteristic

Group	Age	Diagnosis	Sex	Stim Freq (Hz)	Duration(m)	Yahr	UPDRS III	Domi	MMSE	FOGQ	LEDD (mg)
Real	64	PD	F	1.02	144	3	38	R	30	17	519
	76	PD	F	1.08	66	2	14	R	23	6	500
	58	CBS	F	1.23	29	2	30	L	30	7	300
	78	CVD	M	0.67	58	1	7	L	29	18	0
	80	CBS	F	1.05	12	3	22	R	29	2	400
	69	PD	F	1.04	18	3	17	L	29	14	250
	53	PD	F	0.96	27	2	11	R	30	10	200
	63	PD	M	0.92	53	3	18	L	29	5	300
	68	PD	M	1.03	64	3	22	L	30	13	200
	76	PD	M	1.05	116	3	20	L	28	18	325
	79	SCD	F	0.59	79	3	14	R	30	16	0
	73	CVD	F	0.98	48	3	18	R	28	11	0
Sham	73	PD	M		53	3	12	R	25	8	350
	61	PD	M		48	3	22	L	29	1	300
	45	CBS	F		45	3	22	L	30	15	250
	70	PD	M		45	3	24	L	24	1	540
	66	PD	M		132	2	31	R	29	14	739
	80	PSP	M		7	3	21	L	34	13	200
	80	PD	F		6	2	18	L	23	5	200
	71	PD	F		29	3	17	L	29	11	200
	54	PD	F		71	2	8	R	30	8	0
	66	PD	F		157	3	38	R	30	11	589
	86	SCD	F		132	3	31	L	30	6	550

Data are presented as mean (SD) or n. MDS-UPDRS= Movement Disorders Society-Unified Parkinson's Disease Rating Scale. According to both the dyskinesia rating scale (items 1-11 for on-dyskinesia and 12-15 for off-dystonia) and the MDS-UPDRS IV. Off-medication dystonia in all patients was restricted to the most affected side of the body. The MDS-UPDRS III scores range from 0 to 108, with higher scores indicating more severe clinical features. The off-medication state was defined as a minimum 12 h overnight withdrawal of standard-release anti-parkinsonian drugs and a 24 h withdrawal of prolonged-release anti-parkinsonian drugs. The on-medication state was defined by both the patient and clinician, indicating that the medication had been effective for at least 30 min after intake.

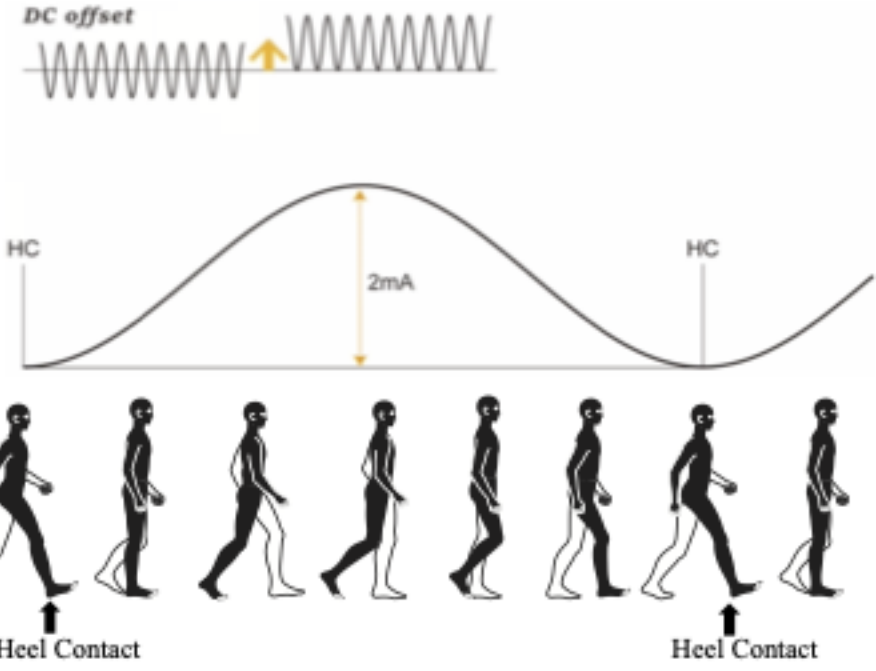
FIGURE LEGENDS

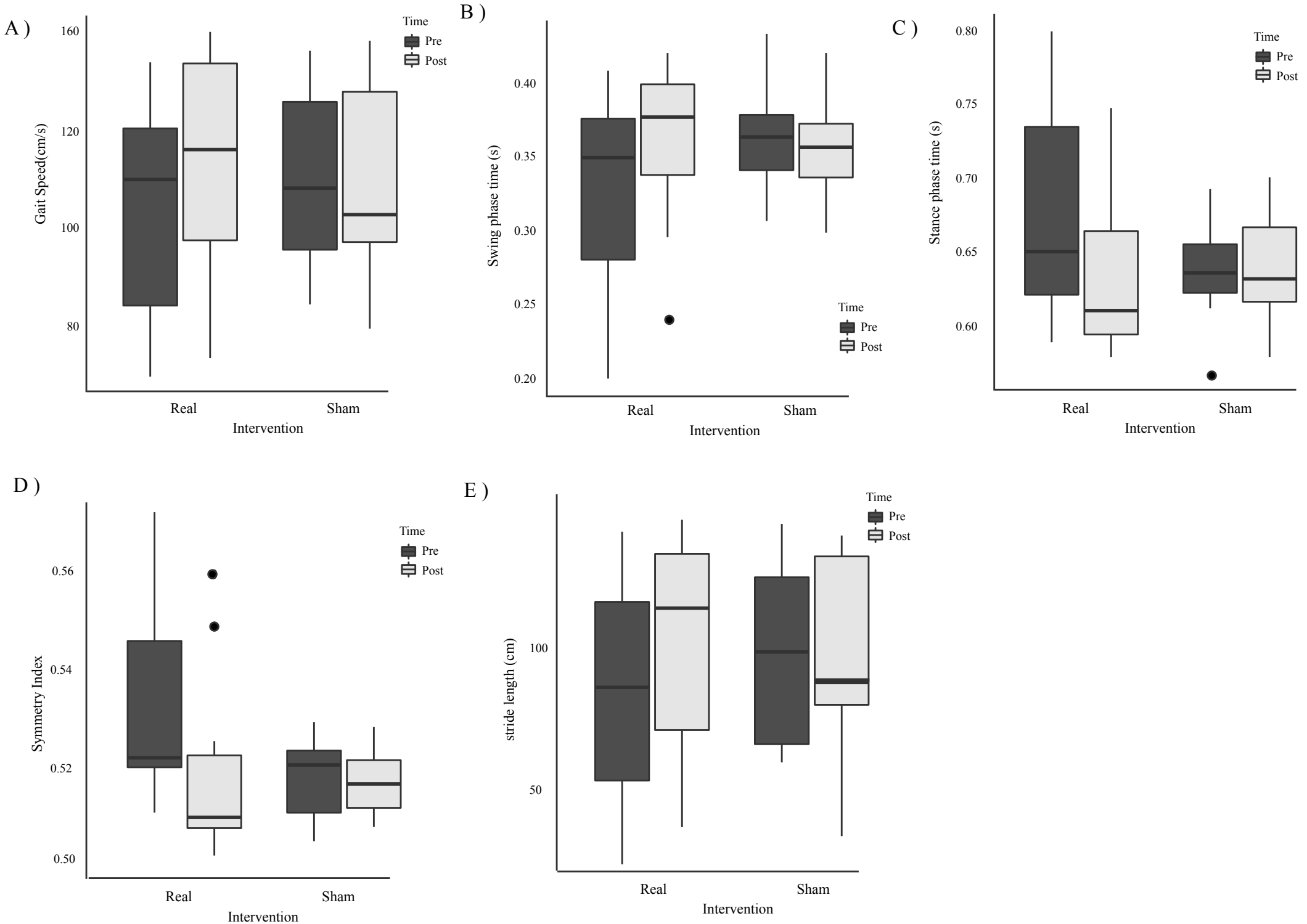
Figure 1. Experimental protocol: In the tES gait condition, electrical current was delivered with a sinusoidal waveform with 2 mA peak. Each current started at the time of heel contact on the severe side during a self-paced 4 min gait. The active electrode (5×5 cm) was applied 3 cm left or right from the inion for cerebellum stimulation. The counter electrode was placed over the opposite position to stimulate the cerebellum.

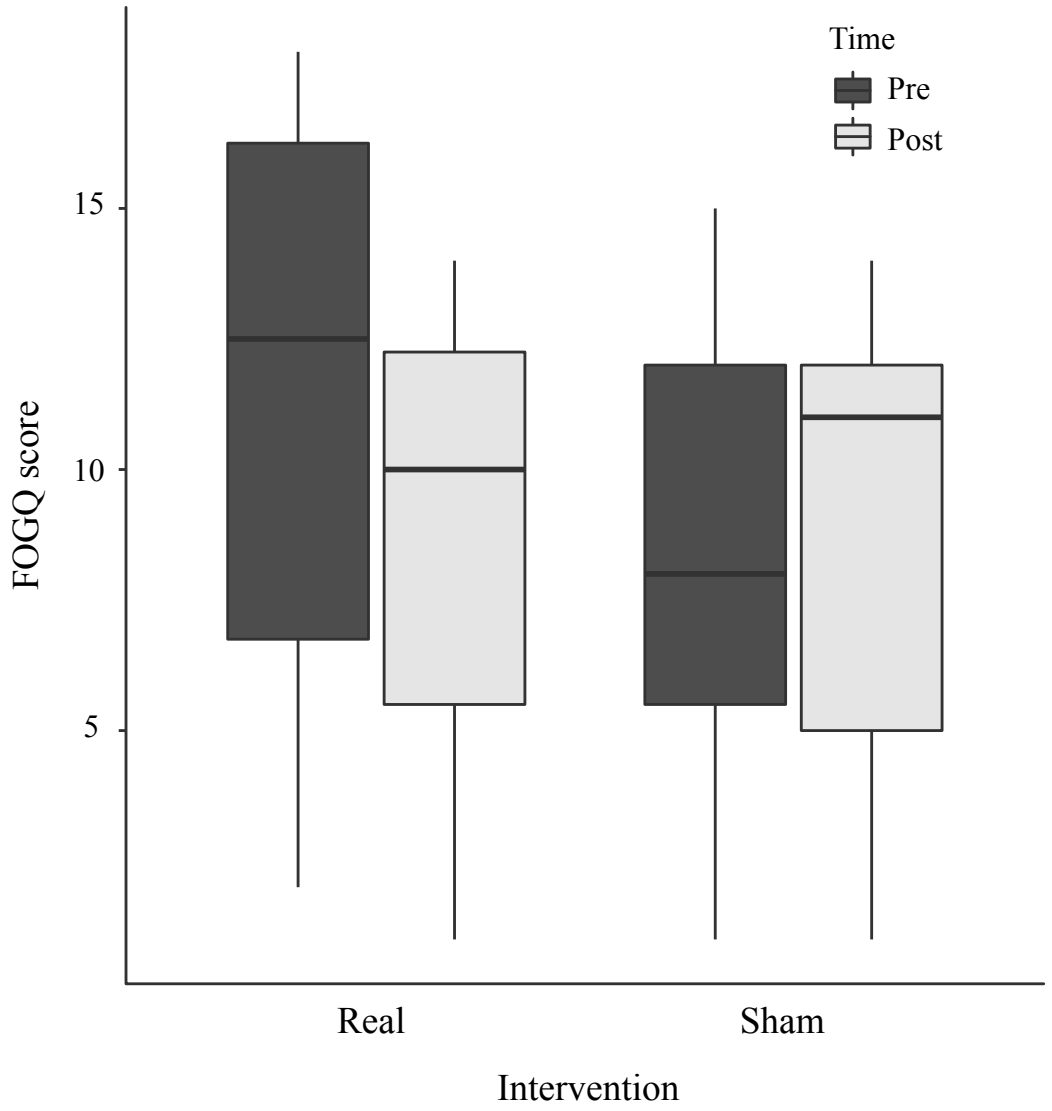
Figure 2. Gait parameters: Effects of 10 times administration of intervention on gait parameters. A) The speed of the comfortable pace, B) length of stride in the comfortable pace, C) ratio of the swing phase on the severe side, D) symmetry index in swing phase time, E) ratio of stance phase on the severe side were improved after the tES gait intervention, compared with those after sham intervention.

Figure 3. Freezing of Gait Questionnaire: The effects of tES or sham stimulation on self-reported severity of freezing of gait (FOG). Participants were asked to rate their change in FOG severity using a Likert scale ranging from 0 to 24 points, showing that higher scores correspond to more severe FOG. The tES synchronized with gait intervention showed significant improvement in FOG after the intervention.

Generate of stimulus




A)
Gait Speed(cm/s)
160
120
100
80
Time
Pre
Post
Real
Sham
Intervention
B)
Swing phase time (s)
0.40
0.35
0.30
0.25
0.20
Time
Pre
Post
Real
Sham
Intervention
C)
Stance phase time (s)
0.80
0.75
0.70
0.65
0.60
Time
Pre
Post
Real
Sham
Intervention
D)
Symmetry Index
0.56
0.54
0.52
0.50
Time
Pre
Post
Real
Sham
Intervention
E)
stride length (cm)
100
50
Time
Pre
Post
Real
Sham
Intervention


Time
Pre
Post
15
10
5
FOGQ score
Real
Sham
Intervention