RESEARCH ARTICLE

The Effect of taVNS at 25 Hz and 100 Hz on Parkinson's Disease Gait—A Randomized Motion Sensor Study

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ABSTRACT: Background: Transcutaneous electrostimulation of the auricular branch of the vagal nerve (taVNS) has the propensity to reach diffuse neuromodulatory networks, which are dysfunctional in Parkinson's disease (PD). Previous studies support the use of taVNS as an add-on treatment for gait in PD.
Objectives: We assessed the effect of taVNS at 25 Hz (taVNS25), taVNS at 100 Hz (taVNS100), and sham earlobe stimulation (sVNS) on levodopa responsive (arm swing velocity, arm range of motion, stride length, gait speed) and non-responsive gait characteristics (arm range of motion asymmetry, anticipatory postural adjustment [APA] duration, APA first step duration, APA first step range of motion), and turns (first turn duration, double 360° turn duration, steps per turn) in advanced PD.
Methods: In our double blind sham controlled within-subject randomized trial, we included 30 PD patients (modified Hoehn and Yahr stage, 2.5–4) to assess the effect of taVNS25, taVNS100, and sVNS on gait characteristics measured with inertial motion sensors during the instrumented stand and walk test and a double 360° turn. Separate generalized mixed models were built for each gait characteristic.
Results: During taVNS100 compared to sVNS arm swing velocity ($P = 0.030$) and stride length increased ($P = 0.027$), and APA duration decreased ($P = 0.050$). During taVNS25 compared to sVNS stride length ($P = 0.024$) and gait speed ($P = 0.021$) increased and double 360° turn duration decreased ($P = 0.039$).
Conclusions: We have found that taVNS has a frequency specific propensity to improve stride length, arm swing velocity, and gait speed and double 360° turn duration in PD patients. © 2024 The Authors. *Movement Disorders* published by Wiley Periodicals LLC on behalf of International Parkinson and Movement Disorder Society.

Key Words: gait; Parkinson's disease; motion sensors; taVNS; vagus nerve

In Parkinson's disease (PD), the lack of dopamine leads to primary motor symptoms of bradykinesia, rigidity, and the rest tremor. Additionally, gait disturbances are linked to dysfunction in cholinergic and noradrenergic systems, along with impairments in cerebellar-motor pathways.[1-10] With disease progression motor symptoms and gait and balance difficulties continue to deteriorate despite dopamine supplementation. Therefore, treatment of dopamine non-responsive gait disturbances represents an urgent, yet unmet need in the current PD treatment strategies.

There is substantial imaging and neurophysiological evidence in healthy individuals, that noninvasive vagal nerve stimulation (nVNS) can modulate the activity of structures otherwise implicated in gait impairment in PD, including diffuse neuromodulatory nuclei such as noradrenergic locos coeruleus (LC) and serotonergic

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raphe nuclei (SnR),[10-14] cerebellar-motor pathways, and motor cortex.[15-18] There is already some preliminary data on both animal models and PD patients to suggest unilateral vagal nerve stimulation (VNS) may improve gait in PD.[19-25]

nVNS can be applied over the vagal nerve in the neck (cervical nVNS [cnVNS]) or at specific points at the ear (transcutaneous auricular VNS [taVNS]), because certain parts of the ear area, such as cymba conchae, tragus, and the inner ear canal have afferent VN distribution.[26] Compared to cnVNS, taVNS allows patients to use the device hands-free during walking and is more readily available in many countries. Stimulation at 25 Hz (taVNS25) has been the most commonly used frequency for nVNS and was recently shown to reduce subthalamic β power PD patients.[25] However, a high resolution functional magnetic resonance imaging (fMRI) study in healthy participants has found that nVNS at 100 Hz (taVNS100) resulted in the most significant LC activation.[13] Furthermore, we have demonstrated in a transcranial magnetic stimulation study that taVNS100 increased the activity the cerebello-thalamo-cortical pathway, whereas taVNS25 did not, suggesting that taVNS has frequency dependent propensity to activate different circuits.[17]

The primary objective of our study was to assess in advanced PD patients, using wearable inertial motion sensors, the effect of taVNS100 and taVNS25 compared to sham VNS (sVNS), on four most levodopa responsive gait characteristics and four levodopa least responsive gait characteristics, as defined by Curtze et al.[27] As a secondary objective, we added the assessment of the effect of taVNS on turn characteristics, as an approximation of "real life" freezing and gait initiation problems in the laboratory setting.[28]

Methods

Participants

In this randomized placebo controlled within subject trial (ClinicalTrials registration number NCT05683925), we included 30 PD patients who had: (1) modified Hoehn and Yahr (mHY) stage >2; (2) subjective history of gait disturbances while on levodopa treatment, preferably with history of freezings; (3) were able to walk for 50 m; (4) were able to follow simple commands (no significant hearing loss/cognitive impairment); (5) had stable dopaminergic therapy for at least 3 months. Subjective history of gait disturbances was considered positive if the patient, while on levodopa treatment, complained on any of the following: difficulties with gait initiation, unsteady gait, difficulties turning, freezings of gait, or any questions included in the Gait Disorders Questionnaire.[29] The study was performed in accordance with the Declaration of Helsinki and with the approval of the Medical Ethics Committee of the Republic of Slovenia (permit no. 0120–502/2020/3).

Because this was the first study assessing the effect of different frequencies of taVNS in PD patients in a within-subject trial design, sample size calculation of 30 participants was made on the assumption that standard deviations of the difference between means will not be larger than 10% of the mean,[27] and the minimal significant standard difference between means we want to detect is 5%, with 0.8 power and a type I error of 0.05. We have predefined the number of participants in our ClinicalTrial registration (NCT05683925).

Study Overview

Each participant completed one visit, in which the instrumented stand and walk test (iSAW) and a double 360° turn (d360°t) were performed during each stimulation condition. The experiment was performed in the ON state, at the time of day determined by participant at their "best." The baseline measurements were performed first, followed by three different stimulation conditions: sVNS, taVNS25, and taVNS100 administered in randomized order (Fig. 1A). During each stimulation, the participants preformed iSAW and d360°t twice in silence and twice while simultaneously performing calculations aloud (ie, double tasking condition to provoke gait disturbances in the laboratory setting), consisting of counting backward by three from numbers above 100. To avoid practice effect, we changed the initial number with each repetition.

For iSAW, participants first stood still for 30 seconds, after which they started walking in a straight line for 7 m, turned and walked back to the start. For d360°ts, the participants were standing in a narrow space (~1 square meter, with the back 30 cm from the wall, a chair and a table were used on the left and right to limit the space). After an audio signal they first performed a 360° turn into one direction and an immediate turn back to the starting position.

If the participant was unable to perform all repetitions of iSAW or d360°t because of poor motor condition, we omitted the counting condition (first for d360°t, then for iSAW). In two cases, the whole d360°t task was omitted to shorten the experiment (Supplementary Appendix; Fig. S1). The participants were equipped with six inertion Opal motion sensors on both feet, both wrists, on the lumbar back, and on their sternum. Only two participants required walking aids to perform the experiment. The data was collected in the gait laboratory at the Department of Neurology in Ljubljana, Slovenia.

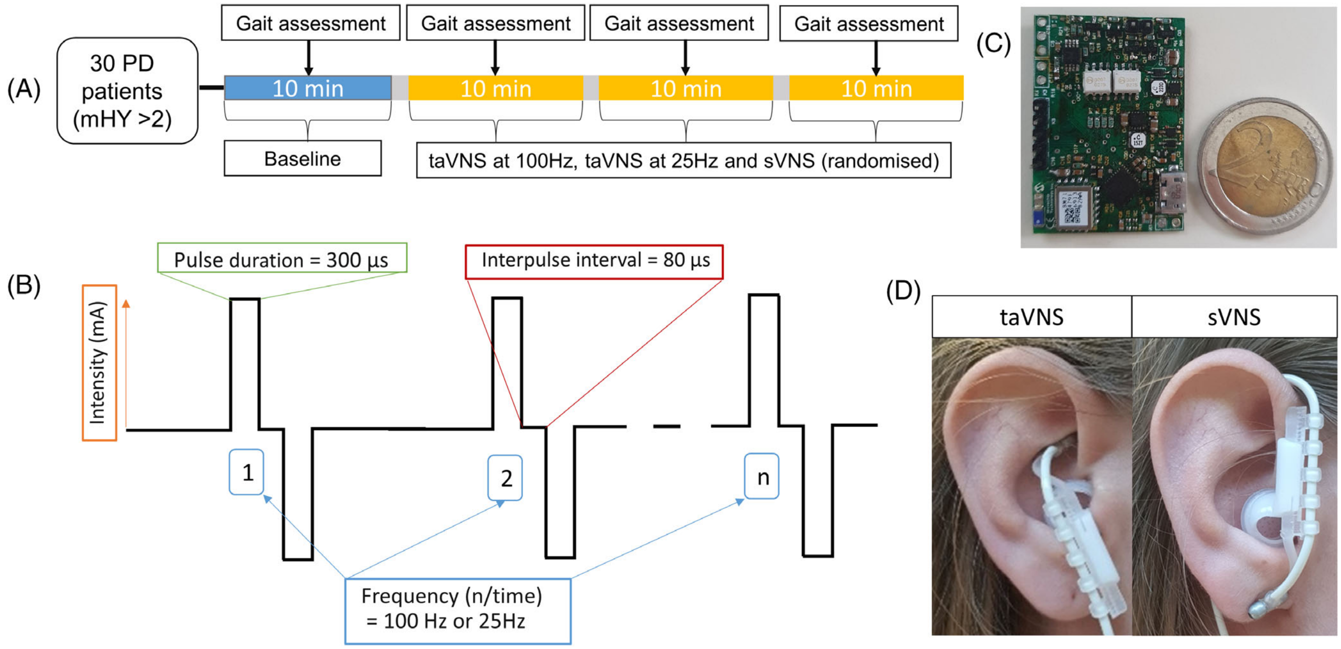

FIG. 1. (**A**) Study overview. (**B**) Stimulation parameters. (**C**) Stimulator next to a 2 euro coin. (**D**) Position for transcutaneous auricular stimulation (taVNS) and sham earlobe stimulation (sVNS). [Color figure can be viewed at wileyonlinelibrary.com]

taVNS

Non-invasive electrostimulation was applied to the left ear. taVNS was applied to the cymba conchae and sVNS to the earlobe (Fig. 1D). The main experimenter explained to the participants that different stimulation positions and different stimulation parameters are compared, to ensure adequate blinding. Both taVNS and sVNS were applied through the Nemos electrode with a small wearable Bluetooth-adjustable stimulator developed by Jan Slapšak s.p. (Jesenice, Slovenia) (Fig. 1C).[16] The stimulation parameters are given in Figure 1B. taVNS25 and sVNS were applied at 25 Hz and taVNS100 was applied at 100 Hz. The intensity for each stimulation was adjusted at the beginning of the experiment, so that the participants were able to feel all three stimulations types, ensuring that there was no subjective intensity difference between real and sham stimulation. A second researcher determined the order of taVNS100, taVNS25, and sVNS by drawing paper cards from a box and placed the electrode in the ear, which was then covered by cotton pads and an electroencephalography cap to hide the electrode position from the main experimenter. The order of stimulation was not revealed until the automated data and statistical analysis.

Data Preparation

To assess gait, the raw accelerometer, magnetometer, and gyroscope data were analyzed with the Mobility Lab software. To limit our research question, we only analyzed the four most (arm swing velocity, arm range of motion, stride length, gait speed), four least levodopa responsive gait characteristics (arm range of motion asymmetry, anticipatory postural adjustment [APA] duration, APA first step duration, APA first step range of motion).[27] Because iSAW and d360°t were repeated twice with and twice without counting during each stimulation condition (baseline, taVNS25, taVNS100, and sVNS), for each of the above eight gait parameters, the mean value of two test repetition was calculated (separately for conditions with and without counting and separately for each side for bilateral gait characteristic).

We additionally investigated the effect of taVNS on turn characteristics, however, the measurements produced by the Mobility Lab software were severely affected by the presence of freezings and irregular steps, which were not detected as part of the turn, thereby fragmenting the detected turn and producing unreliable data. Therefore, to analyze four turn characteristics (first turn duration, d360°t duration, number of steps in first turn, and sum of number of steps in both turns) we developed a custom built algorithm that took into account all steps and analyzed the turn regardless of interruptions by freezings. We first validated the algorithm on 10 randomly selected recordings, by correlating manually counted/measured values with algorithm produced values. Detailed analysis description and validation results can be found in Supplementary Appendix S1.

Statistical Analysis

Main Models

For each of the selected gait/turn characteristics, we divided the value during each condition by the baseline value for each participant, thereby obtaining the relative values during taVNS100, taVNS25, and sVNS, with and without counting.

Statistical analysis was performed in IBM SPSS Package (22; IBM Corp, Armonk, NY). Separate generalized linear mixed model (GLMM) with a gamma or linear probability distribution were built to assess the effect of the three stimulation types on each gait/turn characteristic. Plotted residuals were used to assess whether the homoscedasticity assumptions were met for each measure. Sattherwaite approximation was used to account for potentially unbalanced variance types and robust covariance estimation in cases where potential violations of model assumptions were suspected. Stimulation type (taVNS100, taVNS25, and sVNS) was used as a fixed effect, and individual intercepts were added as random effects. We additionally checked whether the side of measurement (more affected, less affected), gait test repetition (first after baseline, second after baseline, third after baseline) and the presence of counting (yes/no) significantly affected the gait measure and significant predictors were added as fixed factors to the final model. Interactions between the stimulation type and the significant predictors were added only if this increased the fit of the model using the Akaike and Bayesian information criterion. Accordingly, Counting*VNS was added for anticipatory postural adjustment duration in the main model. We further computed the effect of VNS on gait measures per condition (ie, the estimated marginal means of linear trends) and compared the contrasts between the estimated marginal means during each stimulation in a pairwise fashion with Benjamini-Hochberg (BH) correction for multiple comparisons.

Follow-up Analyses

In the follow-up analysis, we further investigated the effects of double tasking (yes/no), preceding stimulation (baseline, taVNS100, taVNS25, and sVNS) and Unified Parkinson's Disease Rating Scale part III (UPDRS-III) score (total UPDRS scores part III were divided into tertiles) on the response to VNS, by looking into main effects and (if applicable) interactions with VNS. Detailed description can be found in Supplementary Appendix S1.

Data Sharing

All anonymized gait characteristics data, R scripts, SPSS scripts, and full trial protocol are available on reasonable request to the researchers, as well as details regarding the stimulator design and code.

Results

From September 2022 to February 2023 we recruited 30 participants (of those seven females) with the average age 72.15 (±10.5) years and average disease duration 9.4 years (±4.45). Average mHY stage was 2.95 (±0.48), average UPDRS-III was 43 (±11.3; first tertile, 36.57; second tertile, 46.42), and average levodopa equivalent daily dose was 1096 (±551). Detailed demographic and clinical characteristics are given in Supplementary Appendix Table S1. No side-effects were reported by any of the participants.

For APA duration, APA first step duration, and APA first step range of motion, because of dyskinesias or prominent tremor in the first 30 seconds of iSAW when participants were supposed to stand still, we only obtained 58% of measurements during quiet standing and 48% of measurements during counting. For arm range of motion asymmetry, we obtained 91% and 88% of measurements, while standing still and counting, respectively. All other measurements were obtained in 100%.

Effect of taVNS100 and taVNS25 in the Main Models

The effect of VNS was significant for arm swing velocity ($P = 0.03$, $F[2,179] = 3.581$), stride length ($P = 0.016$, $F[2,128] = 4.298$), gait speed ($P = 0.027$, $F[2,172] = 3.702$), d360°t duration ($P = 0.028$, $F[2,138] = 3.660$), and APA durations ($P = 0.050$, $F[2,66] = 3.141$). A trend toward significance was observed in APA first step duration ($P = 0.058$, $F[2,28] = 3.17$) (Table 1). BH corrected pairwise comparisons showed that during taVNS100 compared to sVNS, arm swing velocity ($P = 0.030$) and stride length increased ($P = 0.027$), and APA duration decreased ($P = 0.050$) (Table 2 and Fig. 2). Furthermore, BH corrected pairwise comparisons showed that during taVNS25 compared to sVNS stride length ($P = 0.024$) and gait speed ($P = 0.021$) increased and d360°t duration decreased ($P = 0.039$) (Table 2 and Figs. 2 and 3; plots showing individual data points per participant are available in Supplementary Appendix S1).

No significant differences between taVNS25 and taVNS100 were observed.

Gait test repetition was significant for all levodopa responsive gait features (Table 1), with improvement observed with each repetition. With gait test repetition, also the sum of steps in turns, first turn duration and d360°t duration decreased (Table 1), indicating a presence of a practice effect. Counting was significant for stride length, gait speed, APA duration, APA first step duration, and APA first step range of motion (Table 1). In the follow-up analyses, no significant interaction between counting and VNS were observed, suggesting that counting introduced variability into the measurements independently of the stimulation type.

TABLE 1 *Fixed effects for the main models of all analyzed gait and turn characteristics. Significant p-values are bolded.*

Gait measure	Model	Fixed parameters	F	df1	df2	*p*-value
Arm swing velocity	gamma	VNS	3.679	2	163	**0.027**
		Test repetition	4.734	2	107	**0.011**
Arm range of motion	gamma	VNS	0.949	2	42	0.395
		Test repetition	3.172	2	19	0.065
Stride length	gamma	VNS	4.298	2	128	**0.016**
		Test repetition	8.316	2	99	**0.000**
		Counting	15.371	1	133	**0.000**
Gait speed	gamma	VNS	3.702	2	172	**0.027**
		Test repetition	14.126	2	136	**0.000**
		Counting	28.041	1	157	**0.000**
Arm range of motion asymmetry	Linear	VNS	0.711	2	51	0.498
APA duration	gamma	VNS	3.14	2	66	**0.050**
		Counting	7.696	1	25	**0.010**
		VNS*Counting	0.363	2	59	0.697
APA first step duration	gamma	VNS	3.17	2	28	0.058
		Counting	2.739	1	1	0.294
APA first step range of motion	gamma	VNS	2.972	2	7	0.116
		Counting	4.493	1	9	0.062
Steps in first turn	Linear	VNS	2.633	2	131	0.076
Sum of steps	Linear	VNS	5.608	2	86	0.145
		Test repetition	1.961	2	119	**0.005**
First turn duration	Linear	VNS	2.45	2	109	0.091
		Test repetition	4.588	2	69	**0.014**
		Counting	3.359	1	61	0.072
Double 360° turn duration	Linear	VNS	3.660	2	138	0.028
		Test repetition	11.223	2	68	**0.000**

Abbreviations: VNS, stimulation type; APA, anticipatory postural adjustments; CI, confidence interval; df, degrees of freedom; F, F statistic.

"Carry-Over" Effect Exploration

Previous stimulation was a significant predictor for stride length ($P = 0.005$, $F[2,74] = 5.714$), gait speed ($P = 0.000$, $F[3,108] = 8.261$), and arm swing velocity ($P = 0.020$, $F[2,119] = 3.417$), and d360°t duration ($P = 0.000$, $F[3,91] = 6.934$) (Supplementary Appendix Table S2). However, only for stride length, the preceding taVNS100 significantly improved the measurement during the subsequent condition compared to preceding sham, preceding taVNS25, or preceding baseline ($P = 0.003$, $P = 0.006$, and $P = 0.001$, respectively). In other measures, the effect of preceding taVNS was not significantly different to preceding sVNS (Supplementary Appendix Table S3).

Effect of Motor Symptoms on Response to taVNS

The interaction between stimulation type and UPDRS-III tertiles was significant for arm swing velocity ($P = 0.003$), stride length ($P = 0.003$), gait speed ($P = 0.024$), APA duration ($P = 0.008$), and APA first step range of motion ($P = 0.024$) (Supplementary Appendix Tables S4 and S5). BH corrected pairwise comparisons showed (Supplementary Appendix Tables S6 and S7) that in the third UPDRS-III tertile, taVNS100 compared to sVNS significantly increased arm swing velocity ($P = 0.000$) and stride length ($P = 0.022$). taVNS100, compared to both taVNS25 and sVNS, decreased APA duration ($P = 0.001$ and $P = 0.001$). taVNS25 compared to sVNS, increased stride length ($P = 0.018$), whereas taVNS25 compared

TABLE 2 *Contrasts estimates of pairwise comparisons between estimated marginal means for each stimulation type with Benjamini Hochberg adjusted significance levels.*

Gait measures	Pairwise contrasts	Contrast	*p*-value	95% CI
Arm swing velocity	taVNS100-sVNS	0.085	**0.03**	0.021, 0.150
	taVNS25-sVNS	0.065	0.065	0.000, 0.130
	taVNS100-taVNS25	0.02	0.566	0.083, 0,044
Arm range of motion	taVNS100-sVNS	0.058	0.419	−0.049, 0.165
	taVNS25-sVNS	0.062	0.419	−0.032, 0.155
	taVNS100-taVNS25	−0.004	0.914	−0.072, 0.065
Stride length	taVNS100-sVNS	0.021	**0.027**	0.004, 0,038
	taVNS25-sVNS	0.024	**0.024**	0.006, 0,041
	taVNS100-taVNS25	−0.003	0.728	−0.020, 0,014
Gait speed	taVNS100-sVNS	0.016	0.159	−0.003, 0,035
	taVNS25-sVNS	0.027	**0.021**	0.007, 0.046
	taVNS100-taVNS25	−0.011	0.249	−0.030, 0,008
ROM asymmetry	taVNS100-sVNS	−0.331	0.437	−0.902, 0.241
	taVNS25-sVNS	−0.162	0.437	−0.580, 0.255
	taVNS100-taVNS25	−0.168	0.437	−0.560, 0.223
APA duration	taVNS100-sVNS	−0.359	**0.05**	−0.652, −0.066
	taVNS25-sVNS	−0.145	0.245	−0.391, 0.102
	taVNS100-taVNS25	−0.214	0.217	−0.504, 0.076
APA first step duration	taVNS100-sVNS	0.01	0.755	−0.051, 0.071
	taVNS25-sVNS	−0.075	0.102	−0.156, 0.006
	taVNS100-taVNS25	0.085	0.063	0.014, 0.155
APA first step range of motion	taVNS100-sVNS	−0.11	0.132	−0.225, 0.006
	taVNS25-sVNS	−0.047	0.425	−0.193, 0.100
	taVNS100-taVNS25	−0.063	0.132	−0.136, 0.010
Steps in first turn	taVNS100-sVNS	0.049	0.097	0.003, 0.096
	taVNS25-sVNS	0.005	0.843	−0.043, 0.052
	taVNS100-taVNS25	0.044	0.097	−0.003, 0.092
Sum of steps	taVNS100-sVNS	0.02	0.337	−0.019, 0.058
	taVNS25-sVNS	−0.019	0.337	−0.059, 0.020
	taVNS100-taVNS25	0.039	0.15	−0.078, 0.000
First turn duration	taVNS100-sVNS	0.021	0.311	−0.020, 0.061
	taVNS25-sVNS	−0.022	0.311	−0.062, 0.018
	taVNS100-taVNS25	0.042	0.087	0.0004, 0.081
Full turning duration	taVNS100-sVNS	−0.01	0.607	−0.48, 0.027
	taVNS25-sVNS	0.047	**0.039**	0.010, 0.084
	taVNS100-taVNS25	0.038	0.06	0.002, 0.073

Abbreviations: taNVS, transcutaneous auricular vagal nerve stimulation; taVNS100, taVNS at 100 Hz; taVNS25, taVNS at 25 Hz; sVNS, sham earlobe stimulation; APA, anticipatory postural adjustments; ROM, range of motion; CI, confidence interval.

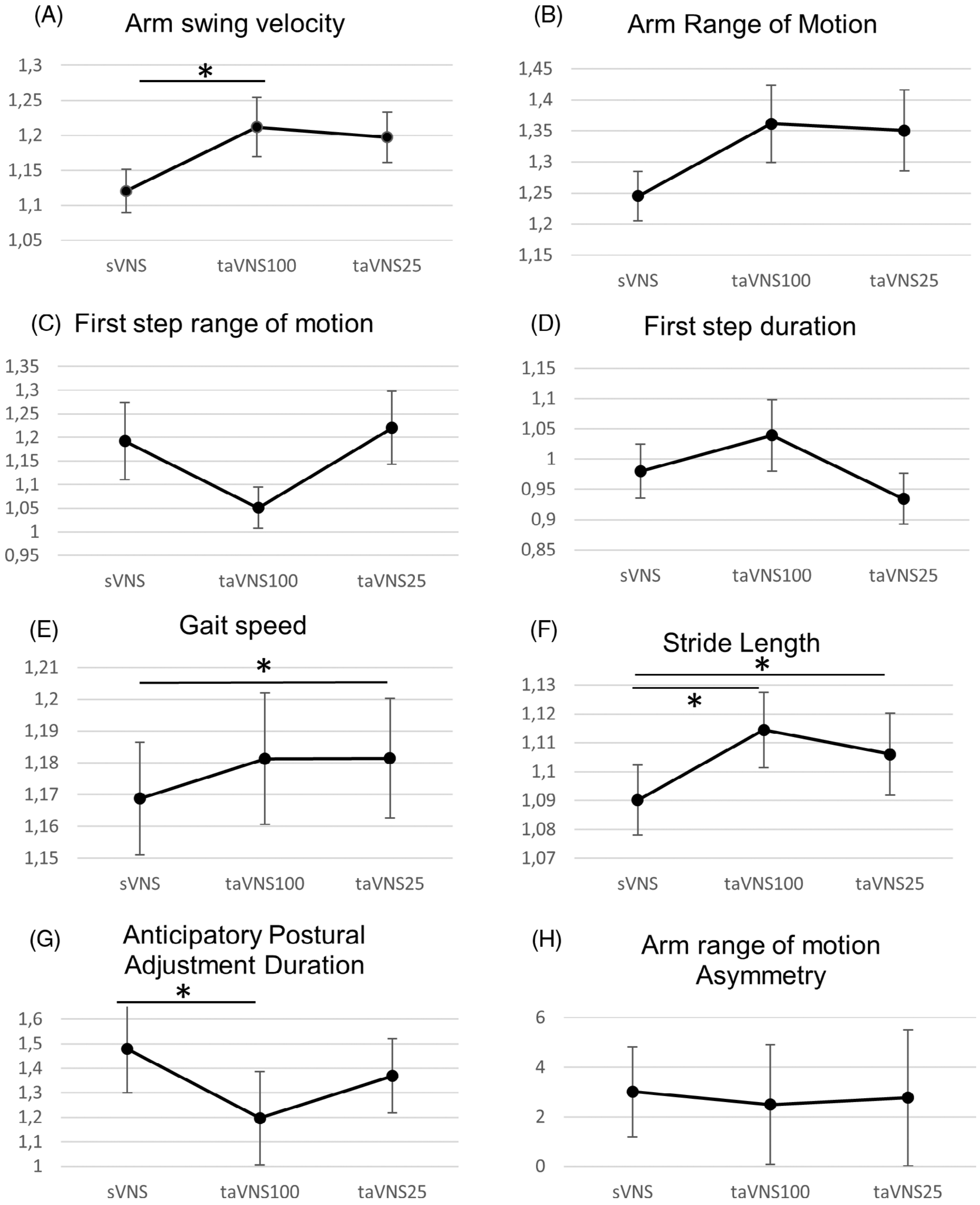

FIG. 2. Visualization of the effect of taVNS25, taVNS100, and sVNS on gait characteristics. Averages of relative values with standard errors of the mean (SEM) during each stimulation condition are shown. taNVS, transcutaneous auricular vagal nerve stimulation; taVNS100, taVNS at 100 Hz; taVNS25, taVNS at 25 Hz; sVNS, sham earlobe stimulation.

to taVNS100 decreased first turn duration ($P = 0.009$). In the second UPDRS-III tertile, taVNS25 increased gait speed compared to sVNS ($P = 0.027$), and decreased stride length compared to taVNS100 ($P = 0.039$). No significant interactions were observed in the first tertile.

Discussion

To the best of our knowledge, this is the first study where the "online" effects on gait of taVNS100, taVNS25, and sVNS were compared (ClinicalTrials.gov ID: NCT05683925). This was made possible by

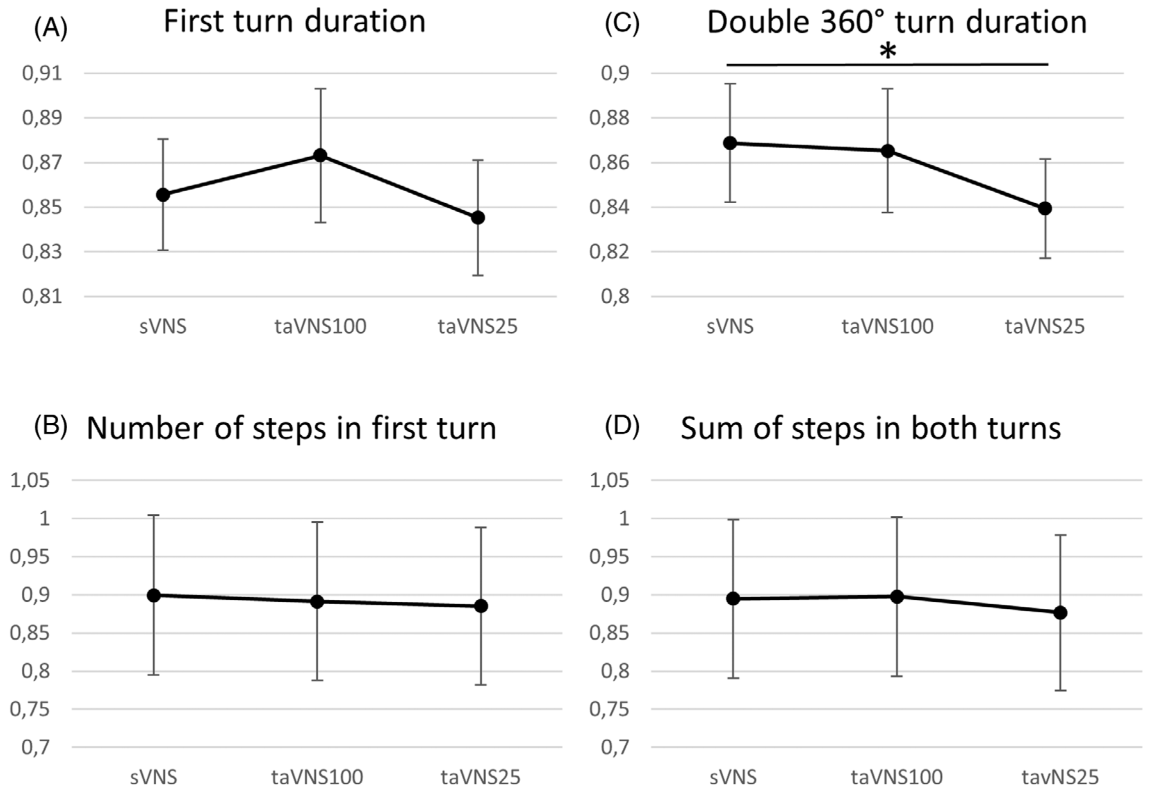

FIG. 3. Visualization of the effect of taVNS25, taVNS100, and sVNS on turn characteristics. Averages of relative values with standard errors of the mean (SEM) during each stimulation condition are shown. taNVS, transcutaneous auricular vagal nerve stimulation; taVNS100, taVNS at 100 Hz; taVNS25, taVNS at 25 Hz; sVNS, sham earlobe stimulation.

delivering electrostimulation using a compact wearable Bluetooth-adjustable stimulator.

We found overall better effect of taVNS on levodopa responsive gait characteristics, with frequency dependent effects. Although both taVNS100 and taVNS25 improved stride length, only taVNS100 improved arm swing velocity, whereas only taVNS25 improved gait speed, all compared to sVNS. The only levodopa-responsive characteristic that was not improved with either form of taVNS was arm range of motion. This may be attributed to a ceiling effect, because our patients were tested in ON state and this characteristic exhibited the highest responsiveness to levodopa.[27] The effect on levodopa non-responsive gait characteristics was less robust. taVNS100 improved APA duration, whereas there was no effect of taVNS25 on any of the measurements. However, it should be noted that APA measurements were not obtained if the patient was not able to maintain sufficient stillness for 30 seconds and patients with dyskinesias or pronounced tremor contributed less measurements, notably reducing sample size and, therefore, diminishing the power of our analysis.

Although no significant effect of either taVNS was observed on number of steps in turns, we found an increased full turning speed during taVNS25. The observation that taVNS25 did not decrease the duration of the first turn implies that the taVNS25 did not alleviate issues related to gait initiation. Instead, it suggests that taVNS25 may either improved akinetic freezing episodes, characterized by a temporary absence of movement or complete akinesia,[30] or enhanced gait speed, as also noted during the iSAW.

Our results are in accordance with recent study in eight PD patients, where increased walking speed and decreased test duration was observed after taVNS25. Our results are also in line with a study on 12 PD patients, showing that taVNS at 20 Hz improved reaction time, total gait time, rotation time, gait speed, stride length, and swing amplitude.[24,25] However, in neither of the two studies, real and sham stimulation were directly compared.

When potential lasting effect of the preceding stimulation is concerned, we observed that only preceding taVNS100 significantly increased stride length in the subsequent stimulation, suggesting that taVNS at 100 Hz may have a prolonged effect that outlasts the duration of stimulation itself.

Furthermore, our follow-up analysis on the interaction between VNS and UPDRS-III tertiles suggests that taVNS may have more pronounced effect in participants with more advanced PD. However, because our study was not powered for this subanalysis, a cautious interpretation is warranted and a larger sample is needed to further investigate this effect.

The mechanism of taVNS effect in patients with dopaminergic neurodegeneration is not well understood. The main hypothesis is that stimulation of vagal afferents activate diffuse neuromodulatory networks, including the noradrenergic and serotonergic networks.[31] Those could in turn modulate both the activity of the motor cortex and the cerebellar activity, thereby surpassing the dopaminergic system. Indeed, previous studies have shown that taVNS has a frequency dependent potential to modulate both the cerebello-thalamo-cortical pathway, and to activate GABA-ergic intractortical circuits.[12,15,17,32] However, because we found a better improvement of levodopa responsive gait features, other explanations should also be considered. taVNS has been shown to activate SnR in healthy individuals because serotonergic neurons have the ability to metabolize exogenous levodopa into dopamine,[33-35] it is possible that the modulation of serotonergic projections with taVNS increases the availability of levodopa in the striatum, thereby improving dopamine responsive gait features. Finally, the existence of kinesia paradoxa in PD underscores the idea that significant non-pharmacological amelioration of motor symptoms is possible even in advanced patients with extensive dopaminergic degeneration, provided they are exposed to suitable stimuli.[36]

Strengths and Limitations of the Study

We compared two different stimulation frequencies of nVNS, namely 25 Hz, which is most commonly used in VNS, and 100 Hz, which has recently emerged as a superior frequency for activation of LC and nucleus tractus solitarii (NTS) and modulation of cerebello-thalamo-cortical pathway.[13,17] We further postulated that because the auricular branch is a sensory branch, simulation parameters similar to trigeminal stimulation should be considered.[37,38]

Our results indicate that nVNS exhibits a frequency-dependent tendency to enhance gait parameters in PD, therefore, underscoring the importance of exploring and comparing various frequencies when evaluating the impact of taVNS in PD.

We have assessed online effect of taVNS by using a portable electrostimulator. This paves the way for the potential implementation of "on-demand" stimulation, where the stimulation is administered as necessary in real-life scenarios, either when a gait issue is identified by sensors or in situations where difficulties may be anticipated by patients.

Our patients underwent testing while on dopaminergic medications, to explore the potential advantages of incorporating taVNS alongside their optimal pharmacological treatment. Curtze et al[27] used the same sensors to compare gait features in ON versus OFF state in advanced PD patients, and found a 37% improvement of arm swing velocity, 5.4% improvement in stride length, and 6.9% improvement in gait speed. Similar relative responses were achieved during taVNS (compared to baseline) in the ON state in our study, implying that detected improvement of gait in our study was an add-on improvement, surpassing the benefits achieved by the best available dopaminergic treatment.

We intentionally recruited participants with advanced PD. Early-stage PD patients taking medication typically exhibit less severe gait impairment, which could potentially result in a ceiling effect, obscuring the outcomes, similar to what would be anticipated in healthy individuals. Moreover, as early PD is managed effectively with pharmacotherapy, improvement of gait among advanced patients carries greater clinical significance for PD community.

We did not collect UPDRS gait scores after each stimulation or gather patients' impressions regarding the effects of taVNS. Consequently, we were unable to determine whether the observed improvement in gait detected with sensitive tools in our study reflects a clinically significant difference. However, we believe that UPDRS gait scoring would overlook clinically relevant changes, because it only captures gross alterations. Furthermore, in our pilot study, we observed that when participants were asked about their subjective impressions, they not only exhibited suggestibility but also showed subsequent changes in performance. Consequently, we made the decision to refrain from collecting subjective reports in this study.

In our study, the stimulation was applied to the left ear, in line with established practice for both invasive and noninvasive VNS.[31] NTS harbor bilateral functional connectivity[39] and unilateral taVNS has been shown to activate bilateral brainstem nuclei in healthy individuals.[13,14] However, a recent study in 10 PD patients showed that taVNS25 significantly decreased β-power in the contralateral subthalamic nucleus, while a trend was observed ipsilateraly.[25] This warrants further studies to investigate benefits of an individualized choice of the side of stimulation, based on the laterality of motor symptoms.

For bilaterally obtained gait parameters (gait speed, stride length, etc.), we explored whether these differed between the more and less affected side by adding this as a fixed effect. However, side did not emerge as a significant predictor in the models, possibly because of more advanced disease stage characterized by a reduction in motor asymmetry. Furthermore, gait in advanced PD has significant axial correlates and therefore may not be the most optimal for exploring the laterality of the effects.

Last, we used six Opal inertial motion sensors and Mobility Lab software to compute gait characteristics, eliminating potential bias from subjective motor assessments by unblinded researchers. However, Mobility Lab software detects representative gait cycles only, excluding irregular ones, which hinders assessment of cycles affected by freezing. To address this, we analyzed turns using custom-built software. Despite this limitation, because both the sensors and Mobility Lab software are validated for use in PD with numerous published studies, we believe that our gait results hold clinical significance.[27,40-43]

Conclusions

taVNS has a frequency specific propensity to improve stride length, arm swing velocity, gait speed, APA duration, and d360°t duration in advanced PD patients. Given that the observed effect in our study occurred during optimal levodopa treatment, these results endorse the utilization taVNS as a supplementary treatment for advanced PD patients experiencing gait impairment. ■

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Trial Registration

The trial was registered on ClinicalTrials under the name "Effect of Transcutaneous Auricular Vagus Nerve Stimulation on Gait Characteristics in Parkinson's Disease" (NCT05683925).

Data Availability Statement

All anonymized gait characteristics data, R scripts, SPSS scripts and full trial protocol can be made available upon reasonable request to the researchers. Furthermore, details regarding the stimulator design and code are available upon reasonable request to researchers.

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Supporting Data

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