Transcutaneous auricular Vagal Nerve Stimulation (taVNS) in mild to moderate Parkinson's Disease
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PROTOCOL TITLE:

Transcutaneous Auricular Vagal Nerve Stimulation (taVNS) in Mild to Moderate Parkinson's Disease

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Study Protocol

Summary

Existing treatment strategies for Parkinson's Disease (PD) are purely symptomatic and incompletely address symptoms, often with intolerable side effects. New strategies are vital to develop more effective treatments for PD. Recent pre-clinical studies conducted by our group indicate potential for symptomatic as well as disease modifying benefit of vagus nerve stimulation (VNS) in PD. Therefore, we propose an investigator initiated pilot study of the acute and subacute effects of transcutaneous auricular vagus nerve stimulation (taVNS) on motor function, cognition, and relevant biomarkers in mild to moderate PD patients.

CONTENTS

1. Background and Significance

VNS and PD: VNS is FDA approved for treatment-resistant epilepsy and depression. The vagus nerve does not have direct connections to cortical regions but rather sends projections through brainstem nuclei, which then relay signals to upper cortical regions. One of these brainstem nuclei used as a relay site is the locus coeruleus (LC), which contains noradrenergic (NA) neurons.[1] VNS increases NA concentrations in the prefrontal cortex of rats, NA neuronal firing rate in LC,[2] as well as brain derived neurotrophic factor (BDNF) expression in the cortex and hippocampus.[3] The loss of LC-NA neurons is significant in PD[4] and occurs earlier in the disease process than the loss of substantia nigra dopamine (SN-DA) neurons.[5,6] Additionally, SN-DA neurons become more sensitive to various insults in the absence of NA nerve fibers.[7,8] NA depletion itself has been implicated in a multitude of pathophysiologic mechanisms in PD, and includes gait dysfunction, dysautonomia, depression, apathy, inattention, and sleep disturbance.[9]

Preliminary data from our neuroscience laboratory (PI-Boger) shows that stimulating the vagus nerve for two weeks improved motor function and resulted in neurorestorative effects on DAergic and NAergic systems, and reduced cellular expression of alpha-synuclein[10] in DA/NA double-lesioned rats. In addition, VNS resulted in a significant increase in BDNF levels in the target regions of the DA and NA neurons. VNS has also been shown to produce an anti-inflammatory effect by decreasing pro-inflammatory cytokines, increasing anti-inflammatory cytokines, and reducing gliosis in models of PD.[10] These results provide pre-clinical evidence for VNS as a potential treatment strategy for PD patients; potentially addressing neuronal damage as well as motor and non-motor symptoms of the damaged noradrenergic and dopaminergic pathways.

Transcutaneous auricular VNS (taVNS): Although cervical VNS is relatively safe, the risks involved in surgical implantation as well as its high cost (about $30-50k) make it less appealing and less available as a study modality. Recently, it has been shown that VNS can be administered non-invasively through stimulating the auricular branch of the vagus nerve located in the ear[11-13] with a transcutaneous electrical nerve stimulation (TENS) device. Our collaborators (Bashar et al.) in the MUSC Brain Stimulation Lab conducted initial safety and feasibility trials of transcutaneous auricular VNS (taVNS) in healthy adults. In these studies, there were no adverse effects of taVNS observed. Their work validates ideal parameters for delivering taVNS, suggesting that one-minute taVNS periods at 200% perceptual threshold delivered at 500μs 25Hz are safe and tolerable and produce regional brain changes consistent with activating the vagus nerve related brain regions involved in neuroplasticity.[13] The same group developed and conducted a concurrent taVNS/fMRI trial and demonstrated that the neurobiological effect of taVNS mimics that of cervically implanted VNS and targets several cortical and subcortical vagus afferent pathway targets. It additionally suggested that taVNS for short time periods activates the vagus nerve (10-25Hz, average 1.8mA). No rapidly accelerated or sustained drops

in HR were seen during the one-minute taVNS stimulation periods. A follow-up study determined that the optimal parameter to modulate the parasympathetic response via taVNS was 500μs pulse width, 10 & 25Hz for 60s duration.[13]

The significance of this study lies in the novelty of stimulating the vagus nerve non-invasively, in order to increase LC-NA neuronal firing and BDNF, and to decrease neuroinflammation. Given the absence of side effects in our human pilot study, we are hopeful to explore a well tolerated treatment option with potential for neuroprotective as well as symptomatic benefits in PD.

2. Objectives

The primary objective of the study is to examine the effect of taVNS on motor symptoms in patients with mild to moderate motor symptoms of PD. The study design is a single site, parallel, randomized, sham controlled, double blinded study, administering taVNS vs sham stimulation 5 days a week for 2 weeks to 35 study subjects. Additional outcomes will be effects on cognition, safety assessments, as well as stimulation effects on relevant biomarkers.

Specific Aim 1: To examine in a 2-week, randomized, sham controlled, double blind trial the effect of taVNS on motor symptoms in patients with mild to moderate motor symptoms of PD.
Hypothesis: taVNS will be superior to sham stimulation on its effects on the motor section of the Movement Disorder Society Unified Parkinson's Disease Rating Scale (MDS UPDRS; primary outcome measure).

Specific Aim 2: To examine in a 2-week, randomized, sham controlled, double blind trial the effect of taVNS on cognitive symptoms in PD.
Hypothesis: taVNS will be superior to sham stimulation on its effects on a composite score of the following cognitive assessment tools: DKEFS, Stroop, Digit symbol, Digit span forward and backwards, Fixation (control), Reflexive, and saccade inhibition tasks, CAARS, PROMIS (secondary outcome measure).

Specific Aim 3: To assess safety and tolerability of taVNS in PD.
Hypothesis: Treatment with taVNS will be as well tolerated as sham stimulation on the following parameters: acute monitoring of vital signs, MDS UPDRS motor evaluations and cognition testing to evaluate for worsening PD severity, adverse events or serious adverse events assessments, and the Columbia Suicide Severity Rating Scale (CSSRS). (tertiary outcome measure)

Specific Aim 4:(exploratory aim) To evaluate effects of taVNS versus sham stimulation on biomarkers relevant to VNS neurobiology.
Hypothesis: taVNS and not sham stimulation will lead to a decrease in serum inflammatory biomarkers (TNF-alpha, and IL-6), increase in serum BDNF, and increase in resting pupil diameter.

3. Design

Overview:

This pilot study of taVNS in PD intends to prove basic concepts of acute motor and cognition improvements with taVNS in PD patients. We will accomplish this by a two-week, single site, parallel, randomized, double blinded, sham-controlled pilot study of taVNS in 35 PD patients. There will be 1 screening visit, 10 stimulation visits, and 1 follow up visit.

At the **screening visit** (visit SC) we will obtain informed consent, review inclusion and exclusion criteria, medical history, demographics. We will obtain a baseline MoCA exam, based on proposed exclusionary criteria. For those enrolled, medications used in the treatment of PD will be held constant throughout the study. Baseline blood draw in the amount of 3 teaspoons (<15Ml) for biomarkers will be obtained by appropriately trained personnel. Urine pregnancy test will be administered to women of childbearing potential. We will randomize the patient to sham (n=15) versus active taVNS stimulation (n=15), and obtain baseline cognition battery (in normal *ON PD medication state*) at this time.

At **visit 1**, MDS UPDRS motor evaluation will be videotaped in the *OFF PD medication state* (defined as at least 12 hours without PD medication). Stimulation or sham will occur over 1 hour as defined below, and then repeat MDS UPDRS motor evaluation will be videotaped. A blinded rater will later score the modified (minus rigidity) MDS UPDRS on all videos at completion of the study.

Visits 2-4 are stimulation treatment visits in the *ON med state* with safety assessments, but no efficacy measures.

Visit 5 (study mid point) includes *OFF med* motor evaluation pre/post stimulation as at visit 1, in addition to the stimulation session.

Visit 6 is a taVNS treatment visits in the *ON med* state with additional behavioral saccade testing.

Visits 7-8 are stimulation treatment visits in the *ON med state* with safety assessments, but no efficacy measures.

Visit 9 is an *ON med* stimulation visit, and includes repeat cognition battery following the stimulation session.

Visit 10 (final day of stimulation) includes *OFF med* motor evaluation pre/post stimulation as at visit 1, in addition to the stimulation session. Repeat blood draw.

Visit 11 (Safety follow up visit) six to fourteen days after visit 10, we will check for any adverse events that might have occurred since the last stimulation session.

SCHEDULE OF ACTIVITIES

	Screening Baseline			Maintenance Phase					Follow up
	Visit SC Week 0-2 (-14d)	Visit 1 Stim day 1	Visit 2-4	Visit 5	Visit 6	Visit 7-8	Visit 9	Visit 10	+6-14 days
Written Informed Consent	x								
Eligibility Criteria	x								
MoCA	x								
Med/Neuro History	x								
Physical Exam	x								x
Blood Draw	x								
Cognitive Battery	x						x		
C-SSRS	x				x		x		
Pre-stim MDS UPDRS		x		x				x	
stimulation thresholds		x							
taVNS vs sham stimulation		x	x	x	x	x	x	x	
pupillometry		x							
saccade					x				
Vital Signs		x	x	x	x	x	x	x	
Post-stim MDS UPDRS		x		x				x	
Monitoring AE		x	x	x	x	x	x	x	x

MDS-UPDRS- Movement Disorder Society- Unified Parkinson's Disease Rating Scale (motor portion used)

C-SSRS- Columbia-Suicide Severity Rating Scale

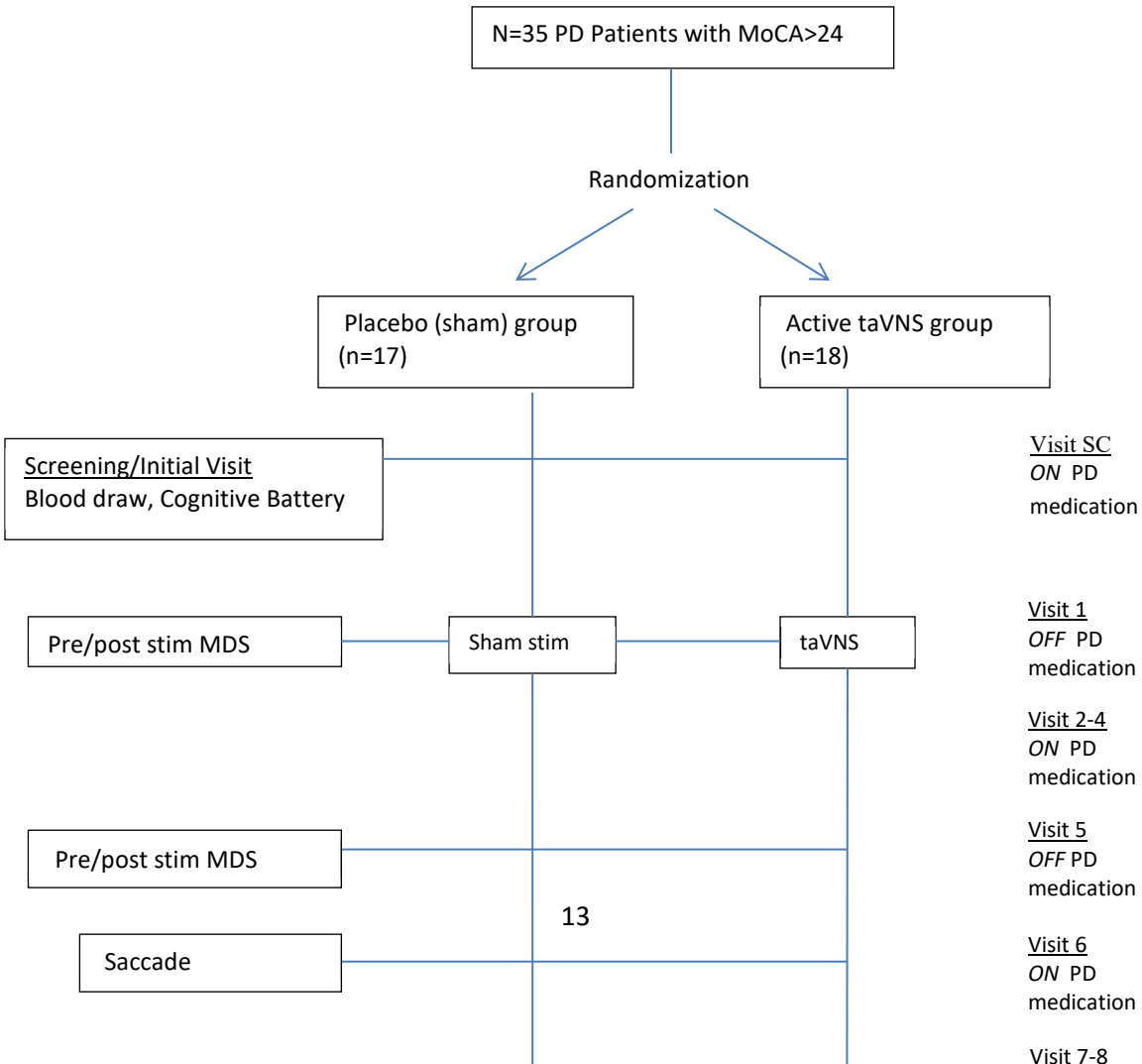
taVNS- transauricular vagal nerve stimulation

saccade- Fixation (control), Reflexive, and saccade inhibition tasks. This is INCLUDED in cognitive battery, but will be done as a stand-alone test on stim day 6

Cognitive Battery- DKEFS, Stroop test, Digit-Symbol test, Digit Span, saccade,

Patient reported measures- CAARS, PROMIS (fatigue, sleep related, Applied Cognitive Abilities)

Figure 1: Overview of Study Flow / Assessments


N=35 PD Patients with MoCA>24
Randomization
Placebo (sham) group (n=17)
Active taVNS group (n=18)
Screening/Initial Visit
Blood draw, Cognitive Battery
Visit SC
ON PD medication
Pre/post stim MDS
Sham stim
taVNS
Visit 1
OFF PD medication
Visit 2-4
ON PD medication
Pre/post stim MDS
Visit 5
OFF PD medication
Saccade
Visit 6
ON PD medication
Visit 7-8

4. Eligibility Criteria

Inclusion Criteria:
Age: 40-82 y
Idiopathic Parkinson's Disease Diagnosis
Disease Stage: Hoehn and Yahr stage 2-3
Patient requires a minimum of 3 doses of levodopa daily
Willingness to be videotaped

Exclusion criteria:
Dementia or MoCa <24
PD psychosis
Ear trauma
Facial pain
TBI or clinical history of stroke
Metal implants above the shoulders
History of myocardial infarction or arrhythmia, bradycardia
Active respiratory disorder
Alcohol or substance use disorders
History of DBS or other brain surgery
Epilepsy
Pregnancy
B-Blockers, dopamine blocking agent, antiarrhythmic medication, acetylcholine esterase inhibitor, midodrine, florinef, droxidopa, or anticholinergic drugs

5. Randomization and masking

Randomization of the 35 subjects will be designed to yield an expected assignment ratio of 1:1 for taVNS and sham stimulation. Treatment assignments will be masked to the patients and to all study personnel with exception of PI and others vital to providing the actual treatment. Patients will be masked to transcutaneous stimulation of the auricular branch of the vagus nerve at the tragus, versus sham stimulation. Sham stimulation involves identical perceptual threshold finding and stimulation parameters as active stimulation, with the exception of stimulation target. Sham stimulation will be delivered to the left earlobe, a target

believed to have little to no vagal nerve innervation. A blinded MUSC Movement Disorder Neurologist will rate six videotaped MDS UPDRS examinations. Rater will be blinded to sham versus active stimulation, pre-stimulation versus post-stimulation state, and to the stimulation day of study.

6. Treatment plan

6.1 Treatment groups

Patients will be assigned to one of the following groups:
- Active taVNS
- Sham stimulation

6.2 Treatment dosing

taVNS will be delivered to the left ear only in one configuration: active (tragus) using a Digitimer Type DS7AH (Digitimer Ltd, Hertfordshire, England) TENS unit cleared by the FDA for electrical nerve stimulation. A clip electrode will be used to stimulate at one of the two positions.

Stimulation will be delivered at a constant current (200% perceptual threshold). Perceptual threshold is defined by the minimum amount of current perceived by the participant. This ranges between 0.5mA to 1.5mA depending on individual anatomical differences. Each taVNS treatment visit will be identical: 1 hour of taVNS (duty cycle: 1min on, 30 sec off)

6.3 Management and reporting adverse events

Management: All adverse events occurring after randomization and during the treatment period, regardless of adherence to study treatment, will be recorded at all visit contacts. Procedures will be promptly discontinued if the participant reports significant distress that does not improve with time or intractable side effect of taVNS.

Importantly, all of these methods have been done before, and published by our and other groups in adults, and we believe these extra precautions will assure the safety and well-being of any participants enrolled.

Patients and caregivers will be asked about visits to doctors, healthcare providers, and emergency departments for other than routine care. The Principal Investigator (PI) will be responsible for monitoring the safety of patients, as well as for appropriate medical care of patients during the study in connection with study procedures.

Safety assessments will include physical exam, vital signs (heart rate primary measure, with additional blood pressure, respiratory rate and pulse oximetry monitoring), MDS UPDRS examinations and Cognitive examinations, Columbia- Suicide Severity Rating Scale

(C-SSRS), monitoring of adverse events, and monitoring and maintenance of concurrent medication records.

Reporting:

The supervising Institutional Review Board (IRB) will be notified of adverse events, risks, or unanticipated problems according to their reporting requirements. If any adverse events are noticed which are not described in the consent or are serious, we will inform the IRB before continuing with the next enrolled subject. Data collected regarding serious adverse events will include the treatment provided, outcome, and presumed relationship to study stimulation and will be updated as new information becomes available; a narrative description also will be provided.

Data Safety Monitoring Board will be utilized. See full description below; An independent review committee charged to monitor the conduct of the protocol to ensure the safety of participants and the validity and integrity of the data.

7. Screening and Baseline visit

7.1 Overview

Obtain Informed Consent, as outlined below. Review inclusion and exclusion criteria, medical history, demographics. We will obtain a baseline MoCA exam, based on proposed exclusionary criteria. For those enrolled, medications used in the treatment of PD will be held constant throughout the study. Baseline blood draw for biomarkers will be obtained. We will randomize the patient to sham (n=15) versus active taVNS stimulation (n=15), and obtain baseline cognition battery (in normal *ON PD medication state*) at this time. Cognitive battery outlined below.

7.2 Overview of consent issues

Informed consent will be obtained from the participant prior to the initiation of any study related procedures. Either the study coordinator or the physician investigator will obtain the consent. The informed consent session will be conducted and documented according to local IRB requirements, as outlined in consent procedure section below

7.3 Review Medical History

The principal investigator or designated study physician must see the patient before randomization. The physician is responsible for fully assessing whether the patient has any of the conditions listed as contraindications, chiefly a history of myocardial infarction or

arrhythmia, bradycardia, epilepsy, metal implants above the shoulders, active respiratory disorder. The presence of any contraindicated condition or medication, which are itemized in the baseline eligibility, constitutes an exclusion criterion.

8. Stimulation Visits

At **visit 1**, MDS UPDRS motor evaluation will be videotaped in the *OFF PD medication state* (defined as at least 12 hours without PD medication).
Stimulation dosing will be assessed as above. Then taVNS or sham stimulation will occur over 1 hour. Acute pupillometry testing will be done. Then repeat MDS UPDRS motor evaluation will be videotaped (assessment of acute taVNS effect). A blinded rater will later score the modified (minus rigidity) MDS UPDRS on all videos at completion of the study.

Visits 2-4 are routine stimulation treatment visits in the *ON med state* with safety assessments, but no efficacy measures.

Visit 5 (study mid point) includes *OFF med* motor evaluation pre/post stimulation as at visit 1, in addition to the stimulation session. (used as mid point motor evaluation).

Visit 6 is a stimulation treatment visits in the *ON med* state with additional behavioral saccade testing. (used as mid point cognition marker free of practice effects, not administered at visit 5, as visit 5 is an *OFF med* visit)

Visits 7-8 are taVNS treatment visits in the *ON med state* with safety assessments, but no efficacy measures.

Visit 9 is an *ON med* stimulation visit, and includes repeat cognition battery following the stimulation session. (The cognition battery is not administered at visit 10, as visit 10 is an *OFF med* day.)

Visit 10 (final day of stimulation) includes *OFF med* motor evaluation pre/post stimulation as at visit 1, in addition to the stimulation session. Repeat blood draw.

9. Specimen Collection

Consented patient participants will be assigned a random *4-digit study ID number and blood serum samples will be drawn by appropriately trained staff. There will be two blood draws per patient; one baseline and one at the conclusion of the study. Blood samples will be delivered to Dr. Boger's lab as soon as possible, following all standard procedures for safe transportation and storage. Dr. Boger's personnel will be blinded to sample name, diagnosis, and all other personal identifiers.

A paper log containing names and corresponding study IDs will be kept in a locked room and locked filing cabinet in the Movement Disorder Program Suite. This room will not be accessed by patients, and is separate from the room where patient evaluations are performed.

10. Follow up visit

Visit 11 (Safety follow up visit) 6-14 days after visit 10, we will check for any adverse events that might have occurred since the last stimulation session, just as we have with every visit.

11. Outcomes Assessments

11.1 Primary outcome: Clinically meaningful treatment difference of the MDS - UPDRS motor section

Videotaped, modified UPDRS evaluations have been established as a reliable research method with very high correlation to full UPDRS assessments.[14] The UDPRS is modified due to the fact rigidity is not assessed by a rater watching a videotaped examination.

The six motor examinations will be compared for both acute as well as cumulative effects of stimulation:

1) Visit 1 pre-stimulation comparison to visit 1 post stimulation: acute effect
2) Visit 5 pre-stimulation comparison to visit 5 post stimulation: acute effect
3) Final Visit 10 pre-stimulation comparison to Visit 10 post stimulation: acute effect
4) Visit 5 pre stimulation comparison to Visit 10 pre-stimulation: subacute effect
5) Visit 1 pre-stimulation comparison to final Visit 10 pre-stimulation: subacute effect
6) Visit 1 pre-stimulation comparison to final Visit 10 post-stimulation: subacute effect

11.2 Secondary outcomes: Cognition and Biomarkers

To examine taVNS effect on cognition, a battery of behavioral and cognitive tasks relevant to PD will be assessed. We will utilize both objective and subjective measures. We will assess baseline, midpoint, and final cognitive function.

Cognitive battery:

1) DKEFS (Delis-Kaplan Executive Function System) verbal fluency and switching: 10 minutes
2) Stroop written test: 4 minutes (including start up and instructions)
3) Digit symbol computer based: 4 minutes (including start up and instructions)
4) Digit span forward and backwards: 3 minutes
5) Fixation (control), Reflexive, and saccade inhibition tasks: 5 minutes (including start up and instructions)

Additional subjective patient reported measures:

1) CAARS (Conners' Adult ADHD Rating Scales) (3 minutes)
2) PROMIS Sleep-Related Impairment (1 minute), PROMIS Applied Cognitive Abilities (1 minute), PROMIS Fatigue (1 minute)

To examine taVNS effect on serum markers associated with PD and neurobiology of VNS, measures of BDNF, TNF-alpha, and IL-6 will be obtained at baseline and after completion of final stimulation session. Laboratory samples will be assayed using ELISA.

To examine whether taVNS leads to the predicted pupillary resting state dilation, Eye tracking with computerized software will be used.

Pupillometry will involve a computer-based system in which an individual will receive stimulation while software records the movements and dilation of the pupil. The system used involves a setup in which infrared cameras are used to monitor the pupil in a luminance-controlled room. Participants will look at a computer screen and be instructed to maintained gaze within a specific area of the screen. IR cameras will track and monitor pupil diameter during stimulation.

11.3 Tertiary outcome: Safety and Tolerability

Safety and tolerability will be monitored for mean change, at each visit, from baseline to follow up. All stimulation visits include monitoring adverse events as well as vital signs. 3 visits include C=SSRS admin, 2 visits include cognition battery, 2 visits include additional physician physical examination. The follow up visit will be one additional visit 6-14 daysfollowing stimulation to specifically monitor for delayed adverse events, and will include a physical examination. Adverse events will be documented.

12. Statistical Analysis

12.1 Primary analysis: MDS UPDRS

A two-tailed independent samples t-test will be used to compare change in MDS UPDRS motor score (post-stimulation at final "on" visit" - baseline) in the treatment versus placebo group. A priori alpha level=.05. Cohen's d will be used to examine effect of stimulation on motor scores.

12.2 Secondary analysis: Cognition and Serum markers

Measures of central tendency and distribution characteristics will be examined for all measures. Appropriate transformations and screening of outliers will be applied as indicated.

Generalized eta-squared[15] will be calculated from repeated measures ANOVAs to examine cognitive change between groups over the 2 time points. For each time point, a composite score for cognition will be estimated by the average of demographically-corrected raw scores. The subjective measures of cognitive functioning will be analyzed in a similar manner. Cohen's d will be used to examine effect of stimulation on serum biomarkers.

12.3 Power Analysis

This study has been designed to provide adequate power to address the primary hypothesis and corresponding analysis. Significant placebo effects have been found in many clinical trials of pharmacologic interventions for motor symptoms in PD. However, a recent meta-analysis found no significant placebo effect associated with sham rTMS on short or long-term changes in UPDRS in PD.[16] Given that there are no prior studies from which to estimate treatment or placebo effects associated with taVNS in PD, we powered our study to detect a clinically meaningful change of -3.25 on the UPDRS,[17] with treatment taVNS. assumed a small and statistically non-significant change of -1 on the UPDRS with sham taVNS, and a standard deviation (pooled) of 2.0. At 80% power and alpha=.05, using a two-tailed t-test for difference in means, the total sample size required is 26 (13 per group). Prior studies in this clinic have found attrition to be about 10% for short-duration clinical trials. Accordingly, a sample of 35 participants will be enrolled.

13. Data, safety, and quality assurance monitoring

13.1 Data Safety Monitoring Board (DSMB)

An independent review committee charged to monitor the conduct of the protocol to ensure the safety of participants and the validity and integrity of the data. The DSMB will include a movement disorder neurologist, an Epilepsy Neurologist with specialized knowledge of Vagal Nerve Stimulation use.

Only the DSMB will review summary data that contains partially unblinded comparisons of AEs and outcomes. The actual study team members will be partially blinded to group assignment for portions of the study. The study reports for review by the DSMB will be prepared by the study coordinator.

Meetings will be held every prior to commencing recruitment, mid enrollment and at the end of enrollment and data collection. The study reports for review by the DSMB will be prepared by the study coordinator. Each report provides cumulative summary statistics including: 1) the number of subjects in each phase of the protocol, and the number who have completed the protocol; 2) the amount of data lost (including subject withdrawal); 3) the presence of any AE, and 4) the presence of any new information, regarding the expected efficacy and safety of taVNS.

From these data, the DSMB will make a determination of whether research progress is satisfactory, whether subjects' risk/benefit ratio has changed, and whether any changes need to be made to any protocol. The DSMB will consult with the database manager, for assurance that no breach of confidentiality has occurred and that quality control procedures are working well. The DSMB will write an annual letter stating that they have reviewed the data and approved the study to continue, that will be included in the annual IRB renewals of the protocol.

13.2 Quality assurance monitoring

The best assurance device is quality-conscious personnel. PI will meet regularly with the research staff, review data collected, and review consent documents for accuracy and completeness. In addition, staff will be reminded of their responsibilities in adhering to high ethical standards, in protecting the privacy of trial participants and confidentiality of records, and in collecting accurate data. They will be reminded to make known to proper authorities any suspicious or wrongful actions in relation to the study. Each member of the research group will sign a statement indicating knowledge and understanding of the above and to disclose potential conflicts of interest. The next best set of assurances lies in the use of design strategies that protect the results from treatment-related biases. This includes random assignment of patients to active treatment or placebo, and masked data collection and outcome assessment. Assurances also depend on the documents, and procedures used for data collection and monitoring.

13.3 Safety Plan

In the event that a participant discloses suicidal ideation or intent to harm him or herself during the formal interview (C-SSRS) or at any other time during the study visit, research staff will contact the PI as soon as possible. The PI will further assess suicidality and develop an appropriate safety plan. If at all possible, the study participant will not be left alone during this time; however, if it is necessary to leave the participant alone in order to contact the PI or enlist other resources, the time left alone will be minimized. Although not anticipated, other situations requiring immediate response from a mandated reporter (i.e., intent to harm someone else, abuse or neglect of a child or dependent adult), will be handled similarly in accordance with applicable state and federal laws.

14. Protection of Human Subjects

14.1 Human subjects

Participants will be men and women with idiopathic PD, with inclusion and exclusion criteria detailed above. There will be no restrictions to participation based on gender, race, or ethnicity. We expect to enroll participants that are representative of the ethnic and racial diversity of the geographic and patient populations of our institution (Medical University of South Carolina).

14.2 IRB approval

Notice of IRB approval and the stamped consent form are required before any study activities may begin. The IRB will be informed promptly of any changes to the study protocol. The appropriate protocol amendment and changes will be submitted to the IRB in writing. Reporting of adverse events will be done according to MUSC IRB policy. A summary adverse event data (not by treatment assignment) will be submitted on an annual basis to the IRB.

14.3 Recruitment and Consent procedures

Patients will be recruited from the Movements Disorders Program at MUSC, and local patient support groups. The MUSC Movement Disorders clinic neurologists are part of the PI's group practice, but are not members of the study team (except for the sub-I). When a patient is seen in clinic, or contacts the site expressing interest, he/she will be informed of the nature of the study by their neurologist or the study coordinator. If the patient chooses to learn more, there may be a short delay between discussing the nature of the study, and the coordinator contacting the patient for a screening visit. If the patient chooses to learn more, he/she will read the consent or have the consent read to him/her. Discussion(s) with prospective subjects will take place in private setting. The patient will then be given the opportunity to discuss the protocol and ask questions. The consent will be signed by the

participant at that time or they will agree to return for a research screening visit and sign the consent form at that time in the presence of the study coordinator. In addition to this recruitment effort, potential candidates will be contacted via telephone by the study coordinator after (or in advance of) their routine clinic visits if the patient's neurologist identifies that person as a potential participant. In this case, the neurologist will give the names of potential participants to the coordinator who will contact the patient, briefly explain the study, and assess the patient's interest. If interested, the coordinator will ask the patient to come in for a screening visit at which time the consent form will be reviewed and signed. The signed consent form will be maintained in a specific study folder in a locked file cabinet. The participant will be given a copy of the signed consent.

Those able to obtain consent include the Principal Investigator, Co-investigator(s), and site research coordinators.

This study will not include patients with dementia. However, capacity to give consent will be carefully assessed in clinical interviews of patients by study staff experienced in clinical PD research. In the course of these interviews, study staff will assess the ability of patients to:

- Comprehend the study and its consent form, by asking them to repeat the key elements of the research
- Understand the study and its consent form, by answering questions about the key elements of the research
- Appreciate the consequences of what will or could happen to them should they agree to participate
- If in this process, a potential patient is found not capable of fully providing consent for participation, then this person will be excluded from study participation

14.4 Potential risks and benefits

Potential Risks of taVNS

taVNS is nothing more than transcutaneous electrical nerve stimulation (TENS) of the auricular branch of the vagus nerve that innervates the ear. Although this novel therapeutic modality is still in the development and optimization process, risks are a combination of those to be expected by both the peripheral TENS and implantable cervical VNS.

TENS devices are FDA approved for pain relief and are available over the counter. The main risks associated with TENS are electrical hazards that may result in user discomfort or injury. The unit used in these studies (Digitimer DS7AH) is a 510(k) cleared electrical stimulator that meets the rigorous electrical standards of the FDA. Skin irritation, redness, or inflammation may occur under the stimulating electrodes if TENS current is delivered for a prolonged period of time.

Implantable cervical VNS is FDA approved for the treatment of intractable epilepsy and treatment resistant depression. Cervical VNS has risks associated with the procedure of

implanting the nerve, and the surgery. None of those apply here. Cervical VNS does have some minimal risks that are due to the actual stimulation of the nerve within the neck such as skin irritation. taVNS also has associated risks that may arise from the direct brain effects stimulating the vagus nerve. These theoretical risks associated with neuromodulation of the parasympathetic nervous system would also be applicable in the administration of noninvasive taVNS. They are the following: reduction of heart rate, blood pressure, and vasovagal syncope.

There have been dozens of studies in which taVNS has been used on humans, none of them reporting adverse events. A simple PubMed search for "transcutaneous vagus nerve stimulation" shows 24 peer-reviewed articles, 15 of which have been published within the past 3 years. See below table for 10 recent publications on PubMed involving taVNS on the auricular branch of the vagus in humans:

Author, Year	**Aim (subject number)**	**Side effects/Risks/AEs**
Aihua et al. 2014	Epilepsy (n= 60)	Dizziness, drowsiness
Capone et al, 2014	Cortical excitability (n=10)	No modification of instantaneous HR, systolic BP, diastolic BP, and mean BP
Kreuzer et al, 2014	Tinnitus (n=50)	Twitching and pressure at electrode site
Rong et al, 2014	Epilepsy (n=144)	None reported
Kraus et al, 2013	taVNS/fMRI (n=16)	A bright, prickling sensation, twinge or stabbing pain
Rong et al, 2012	Depression (n=120)	None Reported – trial still under progress
Kreuzer et al, 2012	Safety Study (n=24)	In those subjects with no known pre-existing cardiac pathology, preliminary data do not indicate arrhythmic effects of tVNS
Busch et al, 2013	Pain (n=48)	No relevant alterations of cardiac or breathing activity or clinical relevant side effects were observed during t-VNS
Stefan et al, 2012	Epilepsy (n=10)	Hoarseness, headache, or constipation
Polak et al, 2009	Far field potentials (n=20)	Slight pain at electrode site

An extremely through review of all taVNS literature has been performed and no harm or adverse events have been observed and any side effects were resolved by decreasing current intensity. There also is currently a commercial taVNS device available for purchase on the European market (Cerbomed - Nemos device; www.cerbomed.com) that is marketed as a take-home treatment for epilepsy.

Not only does the current literature show a lack of harm done by taVNS, but implantable cervical VNS also has an incredible safety record. According to Cyberonics, the company that supplies the cervical VNS implantable devices, there has been over 100,000 implanted patients being monitored by over 3,000 providing physicians. Most side effects range from

alteration of voice, coughing, pharyngitis, hoarseness, headache, and nausea. Cardiac evaluations have been made on hundreds of VNS patients with no changes in cardiac function (Handforth et al, 1998; Sackeim et al, 2001; Morris and Mueller, 1999) with long-term safety confirmed in recent large sample retrospective studies (Menascu et al, 2013; Ryvlin et al, 2014; Choi et al, 2013).

Given the minimal risk both of these already FDA approved methods introduce, we suspect taVNS will be a very safe procedure. taVNS is not intended to be a therapy for currently existing conditions and all subjects will be healthy controls with no previous history of neurological disorders or trauma.

Additionally, there has been study of acute taVNS in 50 PD subjects and 50 control subjects to assess peripheral structural integrity via short pulse stimulation sessions at relatively high amperage. Although not a specific outcome of study, there were no reported significant adverse events. However, an especially noteworthy outcome measured in this study was heart rate variability by several indices that did not differ significantly between the PD and control groups.[18]

The blood draw may result in slight pain, bleeding, bruising, or infection. To decrease the likelihood of such complications, all blood draws will be performed by appropriately trained staff. To protect confidentiality, each subject will be identified with a unique numerical identifier, which will be used for all testing. The data will be kept in a locked file cabinet in a locked office when not being used.

The Cognitive Assessments may cause fatigue or frustration. Subjects will be free to stop a test at any time if they are frustrated, fatigued, embarrassed, or simply chose not to continue. Questionnaires may contain questions that are sensitive in nature. Subjects are able to refuse to answer a question that makes them feel uncomfortable. If the subject has any concerns, they are urged to contact their doctor.

Loss of confidentiality: There is a risk of a loss of confidentiality of personal information as a result of participation in this study. The information we collect will be maintained in a secure manner and access to this information will be limited to study team members only

Participants will hold one dose of medication for the “off medication” condition with is a routine procedure for Parkinson’s studies. Risks to the subjects during the “off medication” state include temporary increase in symptoms of slowness, stiffness, and tremors.

Potential Benefits of taVNS:

The severity of the participants PD symptoms might improve with study intervention. However, this is currently unknown and based on the study hypothesis only. The benefits to society of this study will include important new data on the treatment of PD.

14.5 Safety monitoring

Safety assessments will include physical exam, vital signs (heart rate primary measure, with additional blood pressure, respiratory rate and pulse oximetry monitoring), MDS UPDRS examinations and Cognitive examinations, Columbia- Suicide Severity Rating Scale (C-SSRS), monitoring of adverse events, and monitoring and maintenance of concurrent medication records. Prolonged stimulation or high amperage will not be used to minimize skin irritation.

taVNS is easily discontinued if needed. Study personnel will have frequent contact with participants in person at study visits.

14.6 Confidentiality of patient data

We will keep all patient and caregiver data in a secure locked office, in a locked cabinet and away from patient access areas. Names, addresses, and other such personal data will not be part of the REDcap database.

Study data will be collected and managed in a REDCap database (Research Electronic Data Capture). The database will be managed by a data manager from the MUSC SCTR (SCTR= South Carolina Clinical and Translational Research Institute, that provides infrastructure for enhancing clinical research studies). The PI will perform error checking, and ensure that human subjects data is de-identified, that it meets all regulatory requirements, and that it is stored and backed-up in a secure fashion. REDCap is a secure, web application designed to support data capture for research studies, providing user-friendly web-based case report forms, real-time data entry validation, audit trails and a de-identified data export mechanism to common statistical packages. The database is hosted at the MUSC Datacenter. The system is protected behind a login and Secure Sockets Layer encryption.

Patient video will be collected within the secure "BOX" password protected network storage application that is MUSC IT security approved for patient PHI. There will be no end user device storage of PHI during collection or access. Single sign-in is required for access on any end user device. Folder access will be managed by the physician obtaining the video, the PI, and the blinded Movement Disorder Neurologist. Only after individual login will these key physicians be able to discover the study folder. Data collected from study evaluations and interviews will be identified only by study ID codes, which will be the patient ID and 4-letter code assigned at eligibility evaluation.

Consented patient participants will be assigned a random *4-digit study ID number and blood serum samples will be drawn by appropriately trained staff. There will be two blood draws per patient; one baseline and one at the conclusion of the study. Blood samples will be delivered to Dr. Boger's lab as soon as possible, following all standard procedures for safe transportation and storage. Dr. Boger's personnel will be blinded to sample name, diagnosis, and all other personal identifiers.

A paper log containing names and corresponding study IDs will be kept in a locked room and locked filing cabinet in the Movement Disorder Program Suite. This room will not be accessed by patients, and is separate from the room where patient evaluations are performed.

15. Literature cited

1. Fornai F, Ruffoli R, Giorgi FS, Paparelli A. The role of locus coeruleus in the antiepileptic activity induced by vagus nerve stimulation. Eur J Neurosci. 2011 Jun;33(12):2169-78.
2. Dorr AE, and Debonnel G. Effect of Vagus Nerve Stimulation on Serotonergic and Noradrenergic Transmission. J of Pharm and Exp Ther. 318:890–898, 2006
3. Biggio F, Gorini G, Utzeri C, Olla P, Marrosu F, Mocchetti I, Follesa P. Chronic vagus nerve stimulation induces neuronal plasticity in the rat hippocampus. Int J Neuropsychopharmacol. 2009 Oct;12(9):1209-21
4. Chan-Palay V, Asan E. Alterations in catecholamine neurons of the locus coeruleus in senile dementia of the Alzheimer type and in Parkinson's disease with and without dementia and depression. J Comp Neurol. 1989 Sep 15;287(3):373-92
5. Chan-Palay V. Locus coeruleus and norepinephrine in Parkinson's disease. Jpn J Psychiatry Neurol. 1991 Jun;45(2):519-21
6. H. Braak, U. Rüb, W. P. Gai, and K. Del Tredici. Idiopathic Parkinson’s disease: possible routes by which vulnerable neuronal types may be subject to neuroinvasion by an unknown pathogen. J Neural Transm (2003) 110: 517–536
7. Dymecki J, Freed WJ. Effects of transplantation of cross-species substantia nigra into the lateral ventricle of rats with experimentally induced hemiparkinsonism. Adv Neurol. 1990;53:551-7.
8. Marin C, Aguilar E, Bonastre M. Effect of locus coeruleus denervation on levodopa-induced motor fluctuations in hemiparkinsonian rats. J Neural Transm (Vienna). 2008 Aug;115(8):1133-9
9. Espay AJ, LeWitt PA, Kaufmann H. Norepinephrine Deficiency in Parkinson’s Disease: The Case for Noradrenergic Enhancement. Movement Disorders, Vol. 29, No. 14, 2014
10. Farrand AQ, Helke KL, Gregory RA, Gooz M, Hinson VK, Boger HA. Vagus nerve stimulation improves locomotion and neuronal populations in a model of Parkinson's disease. Brain Stimul. 2017 Nov - Dec;10(6):1045-1054
11. De Ridder D, Vanneste S, Engineer ND, Kilgard MP. Safety and efficacy of vagus nerve stimulation paired with tones for the treatment of tinnitus: a case series. Neuromodulation: Technology at the Neural Interface. 2014;17(2):170-9.
12. Ventureyra EC. Transcutaneous vagus nerve stimulation for partial onset seizure therapy. Child's Nervous System. 2000;16(2):101-2.
13. Badran B, Glusman C, Badran A, Austelle C, DeVries W, Borckhardt J, et al. The physiological and neurobiological effects of transcutaneous auricular vagus nerve stimulation (taVNS). Brain Stimulation: Basic, Translational, and Clinical Research in Neuromodulation. 2017;10(2):378.

14. Abdolahi A, Scoglio N, Killoran A,Dorsey R, Biglan KM. Potential reliability and validity of a modified version of the Unified Parkinson's Disease Rating Scale that could be administered remotely. Parkinsonism Relat Disord. 2013 February; 19(2): 218–221.
15. Bakeman R. Recommended effect size statistics for repeated measures designs. Behavior Research Methods 2005, 37 (3), 379-384
16. Zanjani A, Zakzanis KK, Daskalakis ZJ, Chen R. Repetitive transcranial magnetic stimulation of the primary motor cortex in the treatment of motor signs in Parkinson's disease: A quantitative review of the literature. Movement Disorders, Vol. 30, No. 6, 2015
17. Horvath K, Aschermann Z, Acs P, Deli G, Janszky J, Komoly S, Balazs E, Takacs K, Karadi K, Kovacs N. Minimal clinically important difference on the Motor Examination part of MDS-UPDRS. Parkinsonism and Related Disorders 21 (2015) 1421e1426.
18. Weise D, Adamidis M, Pizzolato F, Rumpf J, Fricke C, Classen J. Assessment of Brainstem Function with Auricular Branch of Vagus Nerve Stimulation in Parkinson's Disease