TITLE PAGE

Full protocol title: How does the type of waveform affect tolerability with transcutaneous auricular vagus nerve stimulation?

Abbreviated title: taVNS Dosage Study 2

IRB Number (eProst ID): 20240796

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1) Protocol Title

How does the type of waveform affect tolerability with transcutaneous auricular vagus nerve stimulation?

Short Title: taVNS Dosage Study 2

2) IRB Review History*

Not applicable

3) Objectives*

We will compare the tolerability and intended physiological effects for 2 different waveform applications of transcutaneous auricular vagus nerve stimulation (taVNS). Specifically, a crossover design will be used to assess if monophasic pulse waveform (Protocol 1) is equally tolerable and effective to a biphasic pulse waveform (Protocol 2) with a set stimulation intensity (2.5 mA) for 60 minutes. Protocol 1 is based on our prior study which demonstrated that 30 second intervals had slightly better tolerability than 10 second intervals. This study is part of a larger research agenda designed to inform the development of personalized pain relief programs based on using patient preferences and a mechanistic understanding of the interventions to integrate neurostimulation interventions into multimodal care for persistent pain. 30 healthy adults (15 female/15 male) will receive 60-minute sessions of 2 different taVNS protocols. Protocol 1 will have a monophasic pulse waveform and a 50% duty cycle that will be 30 seconds on and 30 seconds off. Protocol 2 will have a biphasic pulse waveform with a continuous duty cycle. Each participant will receive both protocols in random order and on separate nonconsecutive days.

Aim 1: Determine the effects of different taVNS waveforms on tolerability and heart rate variability (HRV): Tolerability will be assessed using semi-structured interviews and questionnaires. HRV will be assessed using a chest-strap heart rate monitor (H10, Polar, Finland).We hypothesize that Protocol-2 will have greater tolerability (assessed with interviews and a questionnaire), and it will be associated with a greater increase in HRV (standard deviation of RR intervals).

Aim 2: Determine if the taVNS protocols differ in their effects on pain sensitivity and cortical excitability response. A subset of the participants (n=12) will be used to assess how the different protocols affect corticospinal excitability and pain sensitivity. Corticospinal excitability will be assessed using transcranial magnetic stimulation (TMS) single and paired pulse paradigms. Pain sensitivity response will be assessed using thermal and mechanical quantitative sensory testing. We hypothesize that protocol 2 will induce greater changes in corticospinal

excitability (i.e., increased inhibition and decreased facilitation) as well as decreased pain sensitivity in comparison to protocol 1.

4) Background*

Transcutaneous auricular vagus nerve stimulation (taVNS) is a non-invasive technique that involves the application of electrical currents through surface electrodes on the outer ear to target the auricular branch of the vagus nerve (ABVN). The vagus nerve is part of the autonomic nervous system, which regulates various bodily functions such as heart rate, blood pressure, digestion, and inflammation.[1] taVNS has shown to have various therapeutic effects on different conditions, such as depression, anxiety, inflammation, pain, migraine, and tinnitus.[2-4] The mechanism of action and influence of stimulation parameters on clinical outcomes are still under investigation.

taVNS is a relatively safe and well-tolerated technique.[5-7] However, there is no clear consensus on the optimal parameters for taVNS. Different studies have used different types of waveforms, frequencies, intensities, durations, and duty cycles for taVNS, making it difficult to compare their results and mechanisms.

One issue that affects comfort with electrical stimulation modalities is the current density. taVNS uses small electrodes that fit onto the ear. Smaller electrode pads concentrate the current in a smaller area. This increases the current density and potentially decreases tolerability for a given intensity. This is important as higher dosages are associated with greater physiological responses.

While many devices use a monophasic waveform, this results in a required duty cycle of 50% or less due to the potential for ion buildup that can cause pain or skin burn. However, with a biphasic waveform, this risk of skin burn is mitigated by the changing polarity of electrodes and consistent transfer of positively and negatively charged ions.[8] Thus, a biphasic pulse waveform allows for using a continuous duty cycle in which twice the amount of stimulation can be safely applied within the same timeframe. Thus, identifying differences in tolerability and physiologic effects between monophasic pulse waveform and biphasic pulse waveform may be the best way to optimize taVNS parameters and volume of dosage. Our aim is to determine the effects of different waveforms on tolerability and response to taVNS in 30 healthy adults. Specifically, a crossover design will be used to assess if monophasic pulse waveform with a duty cycle of 50% (30 seconds on and 30 seconds off)

(protocol 1) is equally tolerable and effective to a continuous biphasic pulse waveform (protocol 2) with a set stimulation intensity (2.5 mA) for 60 minutes.

5) Inclusion and Exclusion Criteria*

We will recruit 30 healthy participants (15 males, 15 females).
Inclusion Criteria: 1) between the ages of 18-80 years, 2) English speaking (must be able to consent and complete the interviews in English)

Exclusion Criteria: 1) chronic pain (average intensity >2/10 on 0-10 scale, for longer than 3 months), 2) acute pain of intensity greater than 3/10, 3) chronic inflammatory conditions that are poorly controlled (e.g., diabetes, autoimmune disease), 4) any unstable medical condition or medical contraindication to moderate physical exertion (e.g., unstable angina or cardiac arrythmia), 5) pregnancy, 6) currently taking Buprenorphine or recently stopped taking (within 1 month), 7) presence of cognitive impairment or language barrier that impairs full autonomy in the consent process or in the ability to participate in detailed interviews, 8) implants in the head or neck, cochlear implants, or pacemaker, 9) head or neck metastasis or recent ear trauma, 10) history of epilepsy, 11) history of autonomic nervous system dysfunction (e.g., postural orthostatic tachycardia syndrome).

6) Number of Subjects*

30

7) Study-Wide Recruitment Methods*

Only healthy individuals (no patients) will be recruited for this study. All recruitment will be in accordance with the UM Human Subjects and Research Office guidelines. We will only recruit participants via flyers.

Participants will be recruited from the University of Miami Department of Physical Therapy, The University of Miami Student Complex, and The University of Miami Health and Wellness Centers, and the community.

15 female and 15 male healthy individuals will be recruited. Flyers advertising the study will be posted in University of Miami campuses (i.e., Plumer Building, Wellness Center) and in community centers (e.g., churches and gyms). The flyer will also be distributed to UM Physical Therapy faculty, students, and alumni via email listserv.

Participants will receive $10 ($5 per visit), if participating in the Plumer site data collection, or $30 ($15 per visit) if participating in the Christine Lynn Center data collection, for their time. We have designed the protocol this way to enhance feasibility and to minimize participant burden. As tolerability and HRV are the primary endpoints, all participants will receive these measures. Many of our healthy participants find it more convenient to go to the Coral Gables campus rather than the medical campus, and we are mindful of this preference and the added burden of commute, parking, and

navigating the medical campus for these people. However, it is also important for us to explore potential mechanisms underlying the outcomes and this requires use of the equipment in our laboratory (i.e., TMS and QST). Thus, we are asking a subset of participants to undergo additional measures at the Lynn Center, and we are compensating them at a higher rate given the added time and burden.

8) Study Design

Primary Purpose: The primary purpose is to determine the effects of different waveforms on tolerability and response to taVNS in healthy adults.
Study Phase: N/A
Interventional Study Model: Crossover design
Number of Arms: 2
Masking: Participants will be blinded to which protocol they are receiving during data collection. The investigator responsible for processing HRV data will be blinded to group assignment, and investigators will be blinded to group assignment when conducting qualitative data analysis.
Allocation: Block randomization with stratification by sex
Enrollment: 30

9) Study Timelines*

Each participant will complete 2 visits on separate nonconsecutive days. 18 of the participants will be at the Plumer location and visit 1 and 2 will take ~2 hours each to complete. 12 of the participants will be at the APReCIAT Laboratory in the Lynn Rehabilitation Center and visit 1 and 2 will take ~3 hours to complete. Thus, participants will be enrolled during the time of consent to the completion of visit 2 (total of ~6 hours).

10) Study Endpoints*

A. Primary Outcome Measure Title: Tolerability measured by tolerability questionnaire

Primary Outcome Measure Description: Tolerability will be assessed with a questionnaire with scores ranging from 0-10 for comfort.

Primary Outcome Measure Timeframe: at visit 1 and 2 [Time Frame: Up to 3 hours.]

B. Primary Outcome Measure Title: Tolerability measured by self-report

Primary Outcome Measure Description: Tolerability will be assessed with semi-structured interviews.

Primary Outcome Measure Timeframe: at visit 1 and 2 [Time Frame: Up to 3 hours.]

C. **Secondary Outcome Measure Title:** Change in Heart Rate Variability (HRV)
Secondary Outcome Measure Description: HRV will be measured with an H10 chest strap device (Polar, Finland). Post values will be subtracted from pre values; thus, a positive number will indicate an increase in HRV post taVNS.

Secondary Outcome Measure Timeframe: at visit 1 and 2, 15 minutes at rest pre taVNS application and 15 minutes post taVNS [Time Frame: Up to 3 hours.]

D. **Exploratory Outcome Measure Title:** Effects on pain sensitivity (thermal and mechanical)

Exploratory Outcome Measure Description: : For 12 of the 30 participants, quantitative sensory testing utilizing pinprick, pressure pain threshold, and hot/cold thermodes will be assessed pre and post taVNS application

Exploratory Outcome Measure Timeframe: at visit 1 and 2, pre and post taVNS application

E. **Exploratory Outcome Measure Title:** Change in corticospinal excitability

Exploratory Outcome Measure Description: For 12 of the 30 participants, corticospinal excitability will be assessed using TMS single and paired pulse paradigms.

Exploratory Outcome Measure Timeframe: at visit 1 and 2, pre and post taVNS application

F. **Exploratory Outcome Measure Title:** current affect

Exploratory Outcome Measure Description: Affect and emotional state will be measured via Positive and Negative Affect Schedule (PANAS). The PANAS will be administered prior to each visit and used to determine if current affect is a confounder.

Exploratory Outcome Measure Timeframe: at visit 1 and 2 prior to taVNS and HRV assessment

11) Arms and Interventions

All participants will receive both taVNS protocols in random order. Thus, half of the participants will receive Protocol-1 first followed by Protocol-2, and the other half will receive Protocol-2 first followed by Protocol-1.
Protocol-1 will consist of a monophasic pulse waveform with 30 seconds on and 30 seconds off (50%) duty cycle, and Protocol-2 will consist of a continuous biphasic

pulse waveform. All other parameters will remain the same between protocols (Table 1).

Table 1. Common Parameters between Protocols 1 and 2	
Stimulation intensity	2.5 mA
Session Duration	60 minutes
Frequency	25 Hz
Pulse width	500µS

12) Procedures Involved*

Data collection will take place at the APReCIAT Laboratory located in the Lynn Rehabilitation Center and at the Plumer Building on the UM Coral Gables Campus (Table 2). All participants will complete 2 visits, receiving both taVNS protocols in random order. All participants will also receive HRV assessment for 15 minutes before and after each taVNS trial, and they will complete interviews and questionnaires after each trial. Only the 12 participants who complete data collection at the APReCIAT Laboratory at the Lynn Center and will receive additional assessments of quantitative sensory testing (QST) and transcranial magnetic stimulation (TMS).

Table 2. Study Overview

Location	Pre-trial assessment	taVNS Trial	Post-trial assessment
Plumer Building (n=18)	PANAS, HRV	Protocol 1 and Protocol 2*	Semi-structured interview, tolerability questionnaire, HRV
APReCIAT Laboratory (n=12)	PANAS, HRV, QST, TMS	Protocol 1 and Protocol 2*	Semi-structured interview, tolerability questionnaire, HRV, QST, TMS

taVNS=transcutaneous auricular vagus nerve stimulation; PTQ= Perseverative Thinking Questionnaire; HRV= Heart rate variability; QST= quantitative sensory testing; TMS=transcranial magnetic stimulation; PANAS=Positive and Negative Affect Scales; *= random order

A detailed description of each procedure is provided below:

Positive and Negative Affect Schedule (PANAS)[9]: The PANAS is a 20 item self-report, Likert-type scale designed to measure the recent or general state of

positive and negative emotion. It is expected to take less than 10 minutes to complete, and it will be completed at visits 1 and 2.

Semi-structured interviews: Post-trial semi-structured interviews will be used to examine the perceived experience of each taVNS protocol. An interview template will be created for each visit. Please see templates. Each interview is expected to take ~5 minutes and will be conducted at the end of visits 1 and 2.

Tolerability questionnaire: A taVNS tolerability questionnaire will be used to assess pain ratings (with scores ranging from 0 for no pain to 10 for the worst pain), potential side effects, and belief in taVNS after each session. This questionnaire is expected to take ~5 minutes to complete and it will be administered at visits 1 and 2.

Heart rate variability (HRV): HRV will be measured with an H10 chest strap device (Polar, Finland) and will serve as the primary endpoint for response to taVNS in Aim 2. Participants will wear the chest strap continuously throughout the application of taVNS, and for 15 minutes prior (baseline) and 15 minutes post taVNS trial at each session.

Quantitative Sensory Testing (QST): Each of the 12 participants who complete data collection at the APReCIAT Laboratory will undergo pressure pain threshold (PPT) using the Algomed Digital Algometer (Medoc, Israel), thermal pain testing using the TSA2 Air (Medoc, Israel), and temporal summation pain sensitivity (wind-up ratio) testing using the PinPrick stimulator (MRC Systems GmbH, Germany). The entire QST protocol will take less than 15 minutes to complete. The purpose of these tests are to determine the threshold for pain; thus, the stimulus will be discontinued immediately as soon as the participant indicates that the stimulus has become painful.

PPT: PPT is the force at which pressure begins to be perceived as pain. PPT assists in quantifying static mechanical hyperalgesia to blunt pressure and has been recommended for capturing pain sensitivity. PPT will be measured at the mid-belly of the upper trapezius with a probe area of 1 cm2 (probe diameter of 1.1 cm). The tip of the algometer will be placed perpendicular to the skin and gradual pressure will be applied at a rate of 30 kPa/s. Participants will be asked to press a button to indicate as soon as a painful sensation is perceived, and then rate the intensity of the painful sensation. The PPT will be determined as the average of three trials.

Thermal pain testing: All measurements will be completed using a 30x30mm thermode. Cold Pain Threshold (CPT) is the temperate at which cold begins to be perceived as pain. CPT assists in quantifying static thermal hyperalgesia to contact thermodes. CPT will be measured on the forearm. The thermode will be placed perpendicular to the skin and gradual decreasing temperature will be applied at a rate of $5℃/s$ with a maximum cut-off at 0℃. Participants will be

asked to press a button to indicate as soon as a painful sensation is perceived, and then rate the intensity of the painful sensation on a NRS. The CPT will be determined with three series of descending stimulus intensities, as described above, at each point over 3 rounds.

Hot Pain Threshold (HPT) is the temperature at which heat begins to be perceived as pain. HPT will be measured on the forearm. The thermode will be placed perpendicular to the skin and gradual increasing temperature will be applied at a rate of $5℃/s$ with a maximum cut-off at 50℃. Participants will be asked to press a button to indicate as soon as a painful sensation is perceived, and then rate the intensity of the painful sensation on a NRS. The HPT will be determined with three series of ascending stimulus intensities, as described above, at each point over 3 rounds.

Wind-up ratio: A 256 mN pinprick will be used to assess wind-up ratio over the participants' hand. The perceived intensity of a single pinprick will be compared with that of a series of 10 repetitive pinprick stimuli of the same physical intensity (1/s applied within an area of 1 cm2). The participant will be asked to give a pain rating representing the single stimulus, and the estimated mean over the whole series of 10 stimuli using a '0–10' numerical rating scale. The whole procedure will be repeated 3 times. Wind-up ratio (WUR) will be calculated as the ratio: mean rating of the 3 series divided by the mean rating of the three single stimuli. Quantitative sensory testing is expected to take <15 minutes to complete and will be completed pre and post taVNS at visits 1 and 2 for a subset of the participants (n=12).

TMS single pulse, short-interval intracortical inhibition (SICI), and intracortical facilitation (ICF): Paired-pulse transcranial magnetic stimulation (i.e., SICI and ICF) has become an increasingly popular method of using TMS techniques to probe intra and inter cortical connections. For paired-pulse transcranial magnetic stimulation, a subthreshold conditioning stimulus is followed by the suprathreshold individual test stimulus (TS) with varying interstimulus intervals. ICF can be elicited with ISI of 6–25 ms while SICI is elicited at 1–6 ms. Both, SICI and ICF prove intracortical excitability but through different mechanisms.

SICI and ICF will be assessed using TMS paired-pulse paradigms with the Magpro X100 (MagVenture, Alpharetta, GA). These TMS paired-pulse measures will serve as secondary endpoints for response to taVNS in Aim 2.

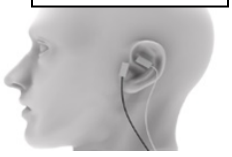
Participants will be comfortably seated in an armchair with the head on a foam headrest. TMS will be delivered through a focal figure-of-eight shaped magnetic coil. The coil will be placed flat on the head over the left motor cortex (Figure 1), at an approximate angle of 45° to the sagittal plane, inducing a current in the brain roughly perpendicular to the central sulcus, flowing from posterior to anterior, as this has been reported to be the most effective way to activate the corticospinal system transsynaptically. Motor evoked potentials (MEP) will be recorded using surface EMG Ag/AgCl electrodes placed over the right first dorsal interosseus muscle in a belly-tendon montage. The optimal coil placement will be determined by recording MEPs while varying the coil position. The coil position leading to the highest peak-to-peak amplitude of the MEP ('hot spot') will be marked using the TMS navigation system (Localite, Germany) to ensure accurate coil positioning throughout the testing. A fixed sequence of TMS measurements will be followed: First, TS and resting motor threshold (RMT) will be defined as the lowest stimulator output intensity that induced MEP peak-to-peak amplitude greater than 50μV in five out of 10 consecutive trials.

Figure 1. TMS coil placement

Then the single pulse and paired-pulse parameters, SICI and ICF, will be obtained in a pseudo-random order. EMG response to single pulse stimuli of 130% of RMT will be recorded. For paired pulse, the conditioning stimulus will be set to an intensity of 70% of the RMT as this does not produce MEPs, and the intensity of the following suprathreshold test stimulus will be set to 130% of RMT. We will use multiple interstimulus intervals (ISIs), including 1, 2, 3, and 5 milliseconds for SICI and 7, 10, or 15 milliseconds for ICF, with 10 pulses at each interval, to produce a response curve. The average of the 10 trials will be used to define the amplitude of the peak-to-peak MEP for each condition.

taVNS: A portable stimulator DS8R Biphasic Constant Current Stimulator (Digitimer, United Kingdom) will be used with flexible hydrogel electrodes to administer taVNS at the cymba concha (Figure 2, with one electrode on the cymba concha and the other on the tragus), using the parameters described in Table 1. This device is approved for research purposes, and these parameters have been shown to safe in healthy people and across a variety of patient populations. Participants will be quiet sitting in a comfortable chair during the application of taVNS.
Please see the Risks to Subjects section for procedures performed to lessen the probability or magnitude of risks.

Figure 2.



13) Data and Specimen Banking*

NA

14) Data Management*

The principal investigator (Marlon Wong) will be responsible for all data management and analyses. A research assistant will assist with data entry and qualitative data coding. All quantitative data will be entered and stored in the University of Miami REDCap system. NVivo software (QSR International, United States) will be used for qualitative data management and analyses, and SPSS version 28 (IBM, United States) will be used for quantitative analyses.

Qualitative Analysis: The Interviews will be recorded and transcribed verbatim. Transcripts will be checked for accuracy against recordings. We will apply rapid qualitative analysis to the interview transcripts to create structured templates and matrix displays. Analysis will be performed independently by multiple coders to minimize bias and ensure credibility and dependability of findings. First, qualitative team members will use the interview guide, while reviewing the transcripts, to create summaries (structured templates) of the interviews. The structured templates will be aggregated to create matrices that enable systematic comparison between participants. After rapid qualitative analyses are completed, the transcripts will be uploaded into NVivo for coding.

Quantitative Analysis: Descriptive statistics will be assessed, including sample mean, standard deviation, median, interquartile range for each continuous variable, and frequencies and percentages for categorical variables. We will test distributional assumptions of all variables by using normality tests and visual inspection. Paired T tests will be used to assess differences in post-trial values between protocols.

Convergence of Qualitative and Quantitative Data: An exploratory sequential mixed methods approach will be used to integrate qualitative and quantitative data, when appropriate, for determining tolerability.

15) Provisions to Monitor the Data to Ensure the Safety of Subjects*

16) Withdrawal of Subjects*

To address the challenge of early identification of an increased risk of a known adverse event, all adverse event data will be tracked and evaluated by the study team on a regular basis. A participant may be taken out of the study if any of the following criteria are met: (i) the participant chooses to withdraw from the study; (ii) the participant's physician determines that the participant should not continue for physical or mental health reasons; (iii) a physical injury that would prohibit continuation of the study is sustained by the participant; (iv) continuing involvement in the study would result in a degradation of participant health; (v) personal, family, or other obligations prohibit further involvement in the study.

No special precautions are needed for the participants before, during, or after the study, including medication, dietary restrictions, or any lifestyle changes. No special care is required for or by the participants.

Participants may be withdrawn from the research without their consent if it is deemed that they are unsafe to participate (e.g., abnormal vital signs, signs of distress such as unexplained diaphoresis, change in cognitive status, and/or signs of intoxication) or if there has been a change in their medical status (e.g., recent surgery or ongoing illness).

If a participant is withdrawn without their consent (fully or partially), the reason will be explained to them and will be notated in REDCap.

17) Risks to Subjects*

The risks associated with participation in the study are deemed to be extremely low. Nevertheless, this study involves the administration of assessment instruments that may cause minor discomfort, the application of an experimental intervention, and the risk of loss of confidentiality. All participants will be informed that all aspects of this study are voluntary. It is also important to note that we have used very similar procedures in two previous studies one of which involving healthy subjects (IRB#20240134) and the other involving cancer survivors with chemotherapy induced peripheral neuropathy (IRB#20230154) both with no adverse effects reported from any participants.

Transcutaneous Vagus Nerve Stimulation is reported in numerous systematic reviews to be effective in treatment of insomnia and relief of acute and chronic pain. Given the non-invasive nature of taVNS, its use is growing for clinical purposes as well as in healthy populations for basic research in cognitive neuroscience and related fields.

Risks/discomforts associated with taVNS: A systematic literature review on the safety and tolerability of tVNS has evaluated 51 studies, independent of the area of application. The authors report that the most prevalent side effect was local skin irritation from electrode placement, occurring in about 18% of included subjects following long-term stimulation. Headache (3.6%) and nasopharyngitis (1.7%) were more rare side effects, and only 2.6% of participants dropped out of studies due to side effects. taVNS will be administered in a laboratory by an investigator and participants will be monitored for any adverse responses. Adverse responses will be tracked and reported to the SCCC Data & Safety Monitoring Committee.

Risks/discomforts associated with TMS:
Seizure or syncope: Induction of seizures is the most severe acute adverse effect of TMS; however, the risk is very low. A published search of the literature only identified 41 reports of seizures. The estimated standardized risk for paired-pulse TMS stimulation is 2/100,000 sessions. Such a seizure has never been reported to result in any permanent damage, and the risk only occurs during the stimulation: there is no heightened risk of seizure after the stimulation. A licensed physical

therapist, and certified basic life support (CPR and AED) provider, will be present at all times. In the case of a seizure, TMS will be terminated immediately, and the provider will follow guidelines for seizure managment4 and activate emergency response if needed. The subject will be assisted in controlled reclining without impact. Airway breathing and circulation will be assessed. Unless tonic-clonic seizure activity occurs, the subject will be turned on one side to help clear the airway and avoid aspiration and maintained in that position until recovery of awareness. Delayed recovery of normal consciousness beyond 30 s following a seizure will mandate further medical evaluation.

Hearing: There is a very small risk of hearing damage. After exposure to the TMS stimulus, a few adult subjects have experienced transient increases in auditory thresholds, and there was a single report of permanent small threshold shift in a person whose ear plug slipped out during stimulation. To minimize the risk of hearing damage, participants will be fitted with ear plugs.

Local pain, headache, or discomfort: TMS is generally well tolerated and experienced as painless by most participants; however, there is a small risk of pain or discomfort with paired-pulse TMS. Less than 2% of patients in TMS trials discontinued treatment due to pain.
Risks/discomforts associated with completion of questionnaires: Some questions may be emotionally distressful to some participants or make them uncomfortable. Participants will be instructed that they can skip any questions which they do not wish to answer.

Risks/discomforts associated with measurement of sensory function (QST): Pain thresholds will be measured for pressure as well as for pinprick. This will result in slight discomfort for the participant. As soon as the participant indicates that the stimulus has become painful, the stimulus will be discontinued immediately. The equipment used for sensory testing has been used in our laboratory for the last few years, without any adverse events. This equipment is also used in many other laboratories and has an excellent track record.

Risks/discomforts associated with heart rate variability monitoring: Heart rate variability data will be gathered through two standard ECG pads on the chest that will be worn continuously. There are no significant risks associated with heart rate variability monitoring. Some people have minor discomfort or skin irritation where the sensors (electrodes) are placed, and we will inspect the skin after assessment.

Risks/discomforts associated with completion of questionnaires: Some questions may be emotionally distressful to some participants or make them uncomfortable. Participants will be instructed that they can skip any questions which they do not wish to answer.

18) Potential Benefits to Subjects*

There are no direct benefits to participants.

19) Vulnerable Populations*

- NA

20) Multi-Site Research*

- NA

21) Sharing of Results with Subjects*

NA

22) Setting

The research procedures will be conducted in the APReCIAT Laboratory located at the Lynn Rehabilitation Center and at the Plumer Building on the UM Coral Gables Campus. The Christine E. Lynn Rehabilitation Center was designed from scratch to be one of the country's elite facilities for patients recovering from traumatic brain injury, spinal cord injury, cancer treatment, and other complex conditions. The stunning 250,000-square-foot, nine-story structure opened in 2020, and it houses 80 inpatient beds and features world-class amenities and next-generation rehabilitative technology. Every aspect of the facility underscores a focus on comprehensive care. The building's layout ensures that clinical care and research share space, so that patients and families see the scientists who are developing treatments, and researchers interact with real people who need the results of their work—a constant reminder of the way Lynn Rehabilitation Center integrates academic study with the day-to-day treatment of patients.

Dr. Wong's APReCIAT Pain Research Lab is located in the Lynn Rehabilitation Center. His dedicated lab space is equipped with a PC, laptop, secured internet access, filing cabinets, office and medical supplies, and research equipment and supplies.

The Plumer Building is the hub of the UM Physical Therapy Department educational activities. It includes multiple floors of educational and research laboratory facilities and Dr. Wong's primary office is located in the Plumer Building. His office is equipped with a PC, laptop, secured internet access, filing cabinets, office and medical supplies, and research equipment and supplies. Research activities at the Plumer building will take place in a laboratory space that is equipped with a PC, laptop, secured internet access, filing cabinets, office and medical supplies, and research equipment and supplies.

23) Resources Available

Research Team:

Marlon Wong, PT, DPT, PhD

Principal Investigator
Dr. Marlon Wong is an Associate Professor of Clinical at the University of Miami Miller School of Medicine within the Department of Physical Therapy. Dr. Wong has extensive research and clinical experience in the assessment and management of pain in a variety of patient populations. As the PI, he will provide expertise and oversee the execution of the aims outlined in this research project. He will develop and conduct the methodology, data collection, data analysis, and manuscript preparation.

Gabriel Gonzalez, PT, DPT
Research Coordinator, PhD Student
Dr. Gabriel Gonzalez, PT, DPT has three years of clinical experience working with chronic pain patients. In addition, he is currently a PhD student in the University of Miami, Physical Therapy Department, under Dr. Wong. Dr. Gonzalez will coordinate the study activities including participant recruitment, scheduling, data collection, and data management.

Juan Gonzalez, DPT
Research Assistant, PhD Student
Dr. Juan P. Gonzalez PT, DPT, has four years of clinical experience specializing in the assessment and management of patients with chronic pain. In addition, he is currently a PhD student in the University of Miami, Physical Therapy Department, under Dr. Wong. Dr. Gonzalez has developed expertise with quantitative sensory testing and data management and will assist with those aspects of the project.

Facilities and Equipment:

Dr. Wong's Clinical Pain Research Lab is located in the Lynn Rehabilitation Center, room 3.412. His dedicated lab space is equipped with a PC, laptop, secured internet access, filing cabinets, office and medical supplies, and research equipment and supplies (detailed below). Only the Magventure device used in this study is considered to be a medical device and it has received 510(k) clearance. The DS8R Biphasic Constant Current Stimulator is not considered a medical device, nor does it have any medical device certification. However, it has been designed for safe use in human research applications, and it will be used according to the approved indications.

Item	Description
1. DS8R Biphasic Constant Current Stimulator	A constant current stimulator for human research studies involving nerve and muscle stimulation via surface electrodes. it is not an FDA regulated device and it has been designed for safe use in human research applications and it meets all of the following standards: • 60601-1 General requirements for basic safety and essential performance. • 60601-1-2 General requirements for basic safety and essential performance -Collateral

	Standard: Electromagnetic disturbances – Requirements and Tests. • 60601-1-6 General requirements for basic safety and essential performance -Collateral standard: Usability. • 60601-2-10 Particular requirements for the basic safety and essential performance of nerve and muscle stimulators. Additionally, it is externally audited to ISO 13485 for medical devices, and each device is subjected to both high voltage isolation tests and electrical safety tests, during production. The Digitimer DS8R has been manufactured and sold since 2016. While it doesn't have any official IDE/IRB exemptions, there have been many inquiries made and IRB/IDE approvals granted based on the information above.
2. MagVenture Magpro X100	high performance magnetic stimulator designed primarily for transcranial magnetic stimulation (TMS) research. It can provide both biphasic and monophasic pulse waveforms, electronically reverse current direction, complex pulse firing patterns, and high frequency stimulation up to 100 Hz. (510[k] # K091940)

24) Prior Approvals

NA

25) Local Number of Subjects

30

26) Confidentiality

Confidentiality will be maintained by assigning participant identification numbers at the beginning of the study. All data will be identified using these ID numbers. The personal information included as data will consist of the gender, age, race, ethnicity, and cancer type and treatment. This information will be kept in a locked file cabinet and stored in a password-protected digital file on the encrypted database at the University of Miami Physical Therapy Department located at 5915 Ponce De Leon Blvd, Plumer Building 1st Floor, Miami, FL 33146. Signed consent forms will be kept in a master file in a locked file cabinet. Only the

principal investigator and research assistant will have access to participant's personal information and data.

Choose the statements below that are applicable to this research:

26(a). Will the research collect protected health information or personally identifiable information from the EMR or from subjects at UHealth and/or JHS?

☐ Yes (If checked go to 26(b))

☒ No (If checked, go to Section 27)

26(b). Check the box next to the correct statement below

☐ Research Subjects will sign a HIPAA Authorization before the research will collect this data.

☐ Research Subjects will not sign a HIPAA Authorization for this data collection and the research is requesting a waiver of HIPAA authorization from the IRB for recruitment purposes only.

26(c). How will the research store the data?

☐ On a University of Miami electronic device (e.g. encrypted, password-protected computer)

☒ On a cloud-based storage system that is approved by the University of Miami (REDCap)

☐ On the secured JHS SharePoint environment

☐ Other, specify: Click here to enter text.

26(d) Select one of the following:

☐ The Principal Investigator (and/or Study Team members) will record (e.g. write down, abstract) data acquired in a manner that does not include any indirect or direct identifiers (listed in the instructions for Section 26 of this protocol), and the recorded data will not be linked to the individual's' identity.

OR

☒ The Principal investigator (and/or Study Team members) will record (e.g. write down, abstract) the data collected in a manner that does not include any direct identifiers (see list in the instructions for Section 26 of this protocol) of any subject. Instead, the Principal Investigator and/or Study Team members shall will assign a code (that is not derived in whole or in part from any direct or indirect identifiers of the individual) to each study subject and link the code to the study subject's identity. The link to each subject's identity and/ or other identifiable information will be maintained on a document separate from the research data.

26(e) Additional requirement for Jackson Health System Data:

NA

☒ Not-applicable, no data will be acquired from JHS under a waiver of authorization.

☐ JHS data, including Protected Health Information (PHI) and/or Personally Identifiable Information (PII), acquired from JHS for this research under a waiver of authorization shall only be stored on the secured JHS SharePoint environment made available by JHS. I and the Study Team members shall not copy or store the JHS sourced personally identifiable information (PII), including protected health information (PHI) data to any other system, including any systems maintained or provided by the University of Miami. I and the Study Team shall only copy or transfer JHS-sourced data that has been properly de-identified in accordance with all requirements contained in the HIPAA Rules by removing all of the identifiers listed in the instructions for Section 26 of this protocol.

27. Biospecimens

☒ Not applicable. No biospecimens will be collected

☐ *Bio*-Specimens obtained for this research will be stored without any direct or indirect identifiers.

☐ *Bio*-Specimens obtained for this research will be stored in a de-identified coded manner.

☐ When required to transport data or bio-specimens for this research, the research team will transport the data and bio-specimens in a de-identified (or anonymous) manner with a link to the individual subject's identity maintain separately from the data and/or bio-specimen.

27) Provisions to Protect the Privacy Interests of Subjects

Only IRB approved research team members will have access to study records. Overall study results will not be shared with study participants, individually. Confidentiality will be maintained by assigning subject identification numbers at the beginning of the study. All data will be identified using these ID numbers. This information will be kept in a locked file cabinet and stored in a password-protected digital file on the encrypted database in the office of the PI in the University of Miami Department of Physical Therapy located at 5915 Ponce de Leon Boulevard, 2nd Floor, Coral Gables, FL 33146. Signed consent forms will be kept in a master file in a locked file cabinet. Only the principal investigator and research assistant will have access to subject's personal information and data.

28) Compensation for Research-Related Injury

Compensation will not be provided for research-related injury. Funds to compensate participants for pain, expenses, lost wages, and other damages caused by injury are not available. However, treatment will be available in most cases and their insurance may be billed or participants will have to pay.

29) Economic Burden to Subjects

There are no direct costs associated with participating in this study. However, transportation and parking may be an economic burden on some participants. The 18 participants who complete data collection at the Plumer building will be paid $10 to compensate them for the economic and time burden, and those who complete data collection at the Lynn Rehabilitation Center will be paid $30. All participants will be paid upon completion of visit 2.

30) Consent Process

We will follow SOP: Informed Consent Process for Research (HRP-090). The consent process will begin with the phone screening when interested people call to inquire about participation. If they provide verbal consent to the phone screening and are deemed eligible for the study, then they will be invited to the Plumer Building or Lynn Center. Potential participants will have the opportunity to consult with family members or others and will have all questions addressed by the PI or other members of the study team. Potential participants will be briefed on their rights as a research subject, including their right to withdraw at any time with no negative consequences.

The study procedures will be reviewed with each participant at each phase of the study and verbal consent will be re-obtained to ensure ongoing consent. Each time that the verbal consent is obtained, it will be reiterated to the patient that they may choose to withdraw from the study at any time.

Waiver or Alteration of Consent Process (consent will not be obtained, required information will not be disclosed, or the research involves deception)

- NA

Cognitively Impaired Adults

- NA

Adults Unable to Consent

- NA

Adults Unable to Consent

- NA

31) Process to Document Consent in Writing

Consent will be documented in REDCap via e-Consent prior to initiating onsite data collection. Please see the attached consent form. A printed

copy will be provided to the participants. During the meeting we will review and explain the information in the consent document, answer any questions the participant has about the study, ask the participant questions about the study to confirm their comprehension, and ask the participant if they consent to participate. If the participant agrees, then they will be asked to sign and date the consent document electronically. Then we will explain to the participant that they will receive a copy of the signed documents via email.

32) Authorization for Use and Disclosure of Protected Health Information (HIPAA)

Since we are collecting data on healthy participants, and not patients, this is not applicable.

Healthy participants only. No data is being collected from or put into participants medical records.

If you are collecting health information from JHS under a waiver of authorization, you must read the paragraph below and sign the signature block to indicate your agreement:
☒Not applicable. This research will not collect data from JHS record under a waiver of authorization

Notwithstanding the preceding "I confirm" statements above, I agree that neither I nor any member of the study team listed on the IRB submission for this Protocol shall ever re-use or re-disclose any of the information acquired from Jackson Health System in any format, whether **identifiable or de-identified,** to any individual or entity without first obtaining written permission from Jackson Health System, even if such re-use or re-disclosure is permissible by law (e.g., HIPAA).

_____

PI Signature Date

33) Drugs or Devices

The DS8R Biphasic Constant Current Stimulator device is approved for research purposes and is being used in this study as intended. The device will be stored and used in our labs at the Lynn Rehabilitation Center and Plumer Building, and it will only be used on study participants and administered only by authorized investigators. Our lab space is not a shared space and always remains locked when not in use. No patient care takes place in the lab space.

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