Photobiomodulation for Management of Temporomandibular Disorder Pain

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STATEMENT OF COMPLIANCE

The study will be conducted in accordance with the International Council for Harmonization guidelines for Good Clinical Practice (GCP) (ICH E6) and the Code of Federal Regulations on the Protection of Human Subjects (45 CFR Part 46). National Institutes of Health (NIH)-funded investigators and clinical trial site staff who are responsible for the conduct, management, or oversight of NIH-funded clinical trials have completed Human Subjects Protection and ICH GCP Training.

SIGNATURE PAGE

The signature below constitutes the approval of this protocol and the attachments, and provides the necessary assurances that this study will be conducted according to all stipulations of the protocol, including all statements regarding confidentiality, and according to local legal and regulatory requirements and applicable US federal regulations and ICH guidelines.

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LIST OF ABBREVIATIONS

|  |  |
| --- | --- |
| AE | Adverse Event/Adverse Experience |
| CATI | Computer Assisted Telephone Interview |
| CFR | Code of Federal Regulations |
| CMP | Clinical Monitoring Plan |
| CONSORT | Consolidated Standards of Reporting Trials |
| CRF | Case Report Form |
| CTSI | Clinical Translational Science Institute |
| DC/TMD | Diagnostic Criteria for Temporomandibular Disorders |
| DHHS | Department of Health and Human Services |
| DSD | Daily Symptom Diary |
| eCRF | Electronic Case Report Form |
| FDA | Food and Drug Administration |
| FFR | Federal Financial Report |
| FWA | Federalwide Assurance |
| GCP | Good Clinical Practice |
| GEE | General Estimating Equation |
| HIPAA | Health Insurance Portability and Accountability Act |
| ICF | Informed Consent Form |
| ICH | International Council for Harmonisation |
| ICMJE | International Committeeof Medical Journal Editors |
| IDE | Investigational Device Exemption |
| IRB | Institutional Review Board |
| LED | Light Emitting Diode |
| LMM | Linear Mixed Models |
| MOP | Manual of Procedures |
| N | Number (typically refers to participants) |
| NDA | New Drug Application |
| NIDCR | National Institute of Dental and Craniofacial Research, NIH, DHHS |
| NIH | National Institutes of Health |
| NRS | Numerical Rating Scale |
| OCTOM | Office of Clinical Trials Operations and Management, NIDCR, NIH |
| OHRP | Office for Human Research Protections |
| PBM | Photobiomodulation |
| PEG | Pain, Enjoyment, General Activity (PEG) Scale |
| PI | Principal Investigator |
| PO | Program Official, NIDCR, NIH |
| PPT | Pressure Pain Threshold |
| QA | Quality Assurance |
| QC | Quality Control |
| SAE | Serious Adverse Event/Serious Adverse Experience |
| SMT | Standardized Movement Task |
| SOP | Standard Operating Procedure |
| TMD | Temporomandibular Disorder |
| UP | Unanticipated Problem |
| US | United States |

PROTOCOL SUMMARY

|  |  |
| --- | --- |
| **Title:** | *Photobiomodulation for Management of Temporomandibular Disorder Pain* |
| **Précis:** | Evidence-based treatments for Temporomandibular Disorders (TMD) are lacking, and the most common treatments for TMD, intraoral appliances and pain medication, provide suboptimal pain control, and the latter often leads to treatment-limiting side effects. Photobiomodulation therapy (PBM) shows substantial potential for the management of pain in people with TMD; however, its efficacy for TMD pain reduction has not been rigorously tested. We will conduct a double-blind randomized, placebo-controlled clinical trial of multimodal PBM (vs. placebo) for TMD pain. This single-site trial will randomize 130 participants with chronic TMD, age 18 years and older, who will be enrolled at the University of Florida, College of Dentistry. Interested participants will complete the pre-screening computer assisted telephone interview (CATI). Individuals meeting pre-screening inclusion criteria will be invited to a pre-randomization visit (V0) that will include informed consent and completion of a detailed medical history to confirm eligibility criteria, followed by a clinical exam to confirm TMD case status according to the Diagnostic Criteria for TMD (DC/TMD). Those who remain eligible after V0 will be randomized at V1 to an actual or placebo photobiomodulation (PBM) condition. Then, participants will complete eight treatment visits (V1 to V8) scheduled within 3 to 5 days apart, a post-intervention visit (V9) that will include questionnaires, TMD exam, Pressure Pain Threshold (PPT), and blood draw, and a 6-month follow up visit (V10) that will also include a TMD exam, questionnaires, and pressure pain threshold as in visit 9. Additionally, the Pain Enjoyment, General Activity (PEG) questionnaire will be collected at 1 and 3 months post-intervention. Analyses will determine intervention effects on the primary outcome (pain intensity) and multiple secondary outcomes and will examine whether changes in inflammation and pain sensitivity mediate intervention response. |
| **Objectives and Outcomes:** | Primary: To determine the efficacy of a multimodal PBM intervention (vs placebo) for reducing pain in participants with chronic TMD pain. The primary outcome measure will be the average of daily pain ratings on the numerical rating scale (0 = no pain, 100 = worst pain imaginable) from the Daily Symptom Diary (DSD) at V9, which will be compared for active versus sham PBM while controlling for baseline pain intensity obtained before V1.  Secondary: To investigate the efficacy of this multimodal PBM on changes in jaw function, pain interference, mechanical pain sensitivity, and inflammatory responses. Also, we will examine the efficacy of PBM in reducing use of pain medication in participants with TMD, and the long-term analgesic effectiveness of PBM. The secondary outcome measures include:  **Pain with jaw function Pain:** Pain intensity and range of motion during a standardized movement task (SMT) at V9 will be compared between intervention groups, controlling for V1 values of this measure.  **Pain Intensity & Interference:** The combined pain intensity and interference from the PEG at V9 will be compared between intervention groups, controlling for V0 PEG scores.  **Pain sensitivity:** PPT during V9 will be compared between intervention groups, controlling for V0 PPT.  **Long term analgesic effectiveness of PBM:** DSD pain intensity scores obtained before V10 will be compared between intervention groups, controlling for baseline (V1) DSD pain intensity scores. Also, PEG Scores obtained at 1,3 and 6 months will be compared between intervention groups, controlling for V0 PEG Scores. Similar analyses will be conducted for PPT and pain during SMT.  **Inflammation:** Circulating cytokines measured at V9 will be compared between intervention groups controlling for baseline (V0) cytokine levels.  **Pain medication use:** The amount of pain medication recorded in the DSD from the week prior to V9 will be compared between intervention groups, controlling for amount of pain medication recorded in the DSD from the week prior to V1. |
| **Population:** | 130 participants age 18 or older meeting diagnostic criteria for chronic TMD will be randomized. Eligibility criteria include facial pain of at least 3 months duration with average pain intensity over the past week of ≥ 30 on a 0-100 numerical rating scale at Visit 0. |
| **Phase or Stage:** | Phase II |
| **Number of Sites:** | One Clinical Site - University of Florida |
| **Description of Intervention:** | **Laser A** (Single Diode Laser) this laser has a trigger point locator and it will be applied on trigger points found in the following areas bilaterally: Temporalis (2 trigger points in the anterior temporalis); masseter (4 trigger points in the body of the masseter) sternocleidomastoid (4 trigger points close to the muscle’s origin and close to muscle’s insertion ), and trapezius ( 2 trigger points on the length of the muscle), and occipital (2 trigger points).  **Laser B** (Laser Cluster): This laser probe has five laser diodes and four LEDs and it will be applied in 14 areas along the nerve pathway at the spinal cord and trigeminal nucleus by treating the C2-C5 area and peripherally targeting the trigeminal area around the TMJ on both sides. Also targeting the sternocleidomastoid muscle and trapezius muscle.  **Laser C** (LED Cluster): This LED probe will be applied in four areas as follows: Pre auricular area, and cervical chain (bilaterally).  Placebo: The PBM equipment (THOR® LX2.3) has a randomizing switch box that allows the assignment of numeric codes for both sham and active intervention. The placebo condition mimics features of active PBM (e.g., light from the protective eyewear, warmth from the probe), but does not provide actual photobiomodulation. Protective eyewear will be worn by staff as well as by participants during both active and placebo therapy. The participant’s goggles also emit LED light inside (behind the lenses), which prevents them from discerning sham versus active intervention. |
| **Study Duration:** | Approximately 5 years |
| **Subject Participation Duration:** | Total participant duration will be approximately 220 days and will include: Telephone Pre-Screening, One Pre-Intervention visit (Screening and Baseline), Eight Intervention visits, One Post- Intervention visit, 1- and 3-month follow-up assessments via internet or telephone, and a 6-month follow-up visit. |
| **Estimated Time to Complete Enrollment:** | Four years |

**Schematic of Study Design:**

Intervention Visits

Visits 6-8

V5+4 days, V6+4 days, V7+4 days (±3 Days)

**Final Assessments**

*PEG, DC/TMD, Blood draw, JFLS, PPT, and review DSD*

Evaluate approximately 200 participants for study inclusion. Obtain informed consent, medical history, DC/TMD clinical exam, Pressure Pain Threshold (PPT), Pain, Enjoyment, General Activity (PEG) Score, Jaw Functional Limitation Scale (JFLS), Psychological Questionnaires, Daily Symptom Diary (DSD) will be provided.

DSD

Randomization

Review Daily Symptom Diary (DSD) to confirm eligibility.

Administer Pre-treatment Questionnaire, collect blood draw.

Review Daily Symptom Diary (DSD).

Administer study intervention (photobiomodulation (PBM) or placebo).

DSD

Randomization Visit 1 Day 0

Screening and Baseline Visit 0 Day -21 to -7

PEG, AEs and concomitant therapies obtained via telephone/internet

DSD

1- and 3 Months Post- Intervention Follow up assessments

Review Daily Symptom Diary (DSD), DC/TMD, PEG, JFLS, Pressure Pain Threshold, and blood draw.

DSD

Review Daily Symptom Diary (DSD)

Administer study intervention (PBM or placebo).

DSD

Review Daily Symptom Diary (DSD), DC/TMD, PEG, JFLS, Pressure Pain Threshold, intervention (PBM or placebo), and blood draw.

DSD

Post-Intervention Assessment Visit 9

Visit 8+ 5 days ± 9 Days

Obtain verbal consent and prescreen via phone approximately 600 participants to identify approximately 200 who meet initial inclusion/exclusion criteria. Collect contact information.

Pre-screening

Day-49 to Day 7

Intervention Visits

Visits 1-4

V0+4 days, V1+4 days, V2+4 days, V3+ 4 days (±3 Days)

Mid-Intervention Visit 5

Visit 4 + 4 days ± 3 Days

Final Study Visit 10 Visit 9 + 180 days ± 30 Days

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# KEY ROLES AND CONTACT INFORMATION

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# INTRODUCTION: BACKGROUND INFORMATION AND SCIENTIFIC RATIONALE

## Background Information

### Background on Temporomandibular Disorder and treatment needed

At least 100 million U.S. adults suffer with chronic pain, and temporomandibular disorders (TMDs) are the 4th most common pain condition in the U.S. population. The prevalence ranges from 5% to 12%, with annual cost estimate around $4 billion (1). TMDs are a set of complex musculoskeletal disorders characterized by pain and limited jaw function and are associated with considerable healthcare costs and morbidity. Although painful TMDs are associated with considerable morbidity, societal costs, and reduced quality of life, evidence-based treatments are lacking, and the most common treatments for TMDs, intraoral appliances and pain medication, provide suboptimal pain control, and medication often leads to treatment-limiting side effects (e.g., GI effects, dizziness, sedation) (2-4). Therefore, the development of new safe and effective therapies is crucial to improve the quality of life for patients suffering from this painful condition.

### Photobiomodulation and treatment of TMD

Recent years have witnessed a burgeoning interest in the therapeutic potential of photobiomodulation (PBM) for treating pain. PBM therapy delivers energy to target tissues to impact biological processes and comprises both the use of low-level laser therapy (LLLT) and Light Emitting Diodes (LED). Briefly, both LLLT (laser) and LED light stimulate photoreceptors in the target tissue(s), activating secondary mediators (e.g., ATP, cAMP, and nitric oxide) and thereby influencing multiple biological processes, including gene expression, cell signaling, cellular metabolism, and cytokine release (5). Therapeutic PBM devices in the form of laser and/or LEDs are often manufactured to emit similar wavelengths in the red or near-infrared spectrum; however, these two modalities differ in important ways. First, laser-based systems are usually higher power than LEDs and the laser beams are smaller, which means the intensity (irradiance) is higher in lasers than in LEDs. Biologically, it has been suggested that the higher irradiance laser will produce stronger direct effects on nociceptive and analgesic pathways than LED (6-8). In contrast, LEDs emit lower power light over a larger area, and this lower irradiance and lower dose produce less penetration, which may have greater effects on inflammation, blood flow, and lymphatic flow. Notably, multiple LEDs can be arranged into arrays, which increases the area of tissue that can be stimulated (5). Thus, while these two forms of PBM share similarities, there are important differences that render them potentially highly complementary in the treatment of musculoskeletal pain. Hence, our protocol will use both light sources, laser, and light emitting diodes.

PBM therapy has been demonstrated to confer several benefits on muscle tissues, including: prevention of post-exercise muscle damage, reducing delayed onset muscle soreness (DOMS), increasing workload capacity of muscle, improving fatigue resistance, and hastening muscle recovery (9, 10). These benefits to muscle function could potentially improve TMD pain, which commonly originates in muscle. Another mechanism implicated in TMD pain is inflammation. Indeed, increased circulating levels of pro-inflammatory cytokines, such as interleukin1β (IL-1β), IL-6, tumor necrosis factor-alpha (TNF-α), and monocyte chemoattractant protein-1 (MCP-1) have been observed in participants with TMD, including both myalgia and arthralgia. For example, in a cohort of participants with mixed arthralgia and myalgia TMD, those reporting high disability showed elevated levels of IL-6, and TNF-α compared to controls25. Also, individuals with mixed myalgic and arthralgic TMD (with or without fibromyalgia) had elevated salivary levels of IL-1β compared to controls (11), and women with TMD myalgia showed elevated intramuscular cytokines compared to controls (12). PBM therapy has been shown to decrease the release of several cytokines (e.g.; IL6, TNF-α, MCP-1, etc.) (13-17), which could, in turn, reduce TMD pain and associated hyperalgesia. Finally, TMD has been associated with peripheral and central sensitization, partially mediated by glutamate, N-methyl-D-aspartate (NMDA)(18, 19), as well as with increased oxidative stress (20, 21). Interestingly, PBM has been shown to reduce peripheral nociceptive drive (22-24) and to decrease expression of pro-nociceptive proteins such as TRPV1, CGRP and substance P following trigeminal nerve injury (25).

## Rationale

The development of new non-pharmacological approaches to therapy is crucial to improve the quality of life of patients suffering from TMDs. As detailed in a recent meta-analysis (26), current evidence supporting the efficacy of PBM for TMD pain is of low quality for several reasons. First, the majority of previous studies have used PBM treatment parameters comprised of a single wavelength, which limits the ability to target multiple pathophysiological mechanisms. Second, most studies failed to apply validated approaches to TMD case definition, resulting in considerable heterogeneity of participant populations. Finally, previous trials have failed to include carefully masked placebo conditions (26, 27). Therefore, we will conduct a double-blind, randomized, placebo-controlled trial that will address these shortcomings. We propose to rigorously test a mixed modality (i.e., comprised of both laser and LED light sources and including different wavelengths, power densities and energy densities) PBM protocol to treat chronic TMD, targeting several potential pathophysiological pathways implicated in TMD pain. We hypothesize that, **compared to sham PBM, active PBM will significantly reduce TMD pain and the analgesic effects of PBM will be associated with reductions in mechanical pain sensitivity and systemic inflammation.**

## Potential Risks and Benefits

### Potential Risks

**TMD Examination and Psychophysical Tests.** Three types of assessments will be performed that may produce transient discomfort: the TMD examination, tests for Pressure Pain Threshold (PPT), and a manual tender point examination. Assessments of muscle and TMJ sensitivity to digital palpation during the TMD examination, responses to masticatory muscle pressure threshold tests, and responses to pressure pain tests are designed to evoke brief pain or discomfort during the application of the stimulus; however, none of these assessments is expected to result in lasting discomfort or damage to affected tissues. Transient redness or tenderness of skin is possible after these tests. All testing modalities will be conducted within a range of stimulus intensities that produce a level of pain or discomfort that is acceptable to the participant. No test will exceed this range, and all tests will end upon the completion of the modality or upon the participant’s request, whichever comes first.

**Blood Collection.** Risks of drawing blood include discomfort at the site of puncture; possible bruising and swelling around the puncture site; rarely an infection; and, uncommonly, faintness from the procedure. Trained personnel will perform the blood collection using standard procedures.

**Questionnaires**. A participant may experience discomfort associated with being asked personal questions about their health history, symptoms, or feelings. Participants will be told that they may choose not to answer any questions that cause discomfort. If a participant experiences or expresses significant distress when completing questionnaires, a study team member will discuss this with the participant and provide referral to appropriate mental health services if needed.

**PBM therapy.** PBM may produce minimum discomfort such as light warmth or heat on the area where the light is being applied, redness in the area of application, itching, dryness, and dry mouth. If the participant feels uncomfortable, they can stop at any time. All interventionists and participants will be provided with goggles to protect their eyes from the light. Treatment directed to the head and neck with high irradiance laser may cause pain as the melanin in the fine superficial hair follicle absorbs a lot of the laser energy. Participant skin tone will be recorded using the Monk Skin Tone Scale (as described in section 5.6.8.2 of the MOP) to monitor whether laser sensitivity varies by skin tone. If treatment becomes painful, the treatment probe will be removed from contact and treatment will continue ~15mm from the skin surface.

**Violation of Confidentiality.** The risk of violation of confidentiality exists because human participants are providing personal information, including biological data. Strategies to minimize this potential risk include collecting only minimal identifying information, using unique study codes for participants, and storing data collection documents on secure, password-protected servers or in a locked private office within locked filing cabinets. Password-protected computers will be used, and only individuals involved in the study will have access. Compliance with all IRB regulations concerning data collection, data analysis, data storage, and data destruction will be strictly observed.

### Potential Benefits

Participants may or may not benefit from receiving the PBM and may experience decreased pain and improved functionality. In an attempt to reduce pain related to TMD as well as decrease the overuse of pain medication, this search for an efficacious, safe, and affordable treatment may benefit individuals suffering from TMD in the future.

# OBJECTIVES AND OUTCOME MEASURES

## Primary

| **Objective** | **Brief Description/Justification of Outcome Measure** | **Outcome Measured By** | **Time Frame** |
| --- | --- | --- | --- |
| To determine the efficacy of multimodal PBM (vs. placebo) for reducing pain in participants experiencing chronic TMD pain. | The primary outcome measure will be the average of daily pain ratings on the numerical rating scale (0 = no pain, 100 = worst pain imaginable) from the Daily Symptom Diary. Daily pain ratings averaged over one week prior to randomization will be treated as the baseline variable. The average daily pain for one week prior to Visit 9 will be treated as the primary endpoint. This outcome measure averaged across multiple days provides a stable measure of pain that is less subject to recall bias. | The primary outcome measure will be assessed using the electronic Daily Symptom Diary, whereby participants will report their average pain each day using a 0-100 Numerical Rating Scale. The average participant scores at Visit 9, controlling for pain intensity obtained before V1, will be compared between the intervention group and placebo group to assess intervention efficacy. | The Daily Symptom Diary will be completed for at least 4 days during the week (7 days) prior to Visit 1 (randomization) and again for at least 4 days during the week following completion of the intervention (or placebo) prior to Visit 9. These two visits are expected to occur approximately 4-6 weeks apart. |

## Secondary

| **Objective** | **Brief Description/ Justification of Outcome Measure** | **Outcome Measured By** | **Time Frame** |
| --- | --- | --- | --- |
| To investigate the effectiveness of PBM in reducing pain related to TMD up to 6 months after completion of the intervention. | The primary outcome measure will be the average of daily pain ratings on the numerical rating scale (0 = no pain, 100 = worst pain imaginable) from the Daily Symptom Diary. Daily pain ratings averaged over one week prior to randomization will be treated as the baseline variable. The average daily pain over the one week prior to Visit 10 (6-month visit) will be treated as the primary endpoint. This outcome measure averaged across multiple days provides a stable measure of pain that is less subject to recall bias. | The primary outcome measure will be assessed using the electronic Daily Symptom Diary, whereby participants will report their average pain each day using a 0-100 Numerical Rating Scale. The average participant scores at Visit 10, controlling for pain intensity obtained before Visit 1, will be compared between the intervention group and placebo group to assess intervention efficacy. | The Daily Symptom Diary will be completed for at least 4 days during the week prior to Visit 1 (randomization) and again for at least 4 days during the week prior to the final study visit, Visit 10. These two visits are expected to occur approximately 28-32 weeks apart. |
| To investigate the effectiveness of PBM in reducing pain intensity and interference related to TMD up to 6 months after completion of the intervention. | The outcome measure will be the Pain, Enjoyment, General Activity (PEG) score V10, controlling for the V0 PEG score. PEG scores will be assessed at baseline (V0) and at 1,3, and 6 months (V10) after completion of the intervention. The PEG is a valid and reliable measure of pain intensity and interference and is sensitive to intervention-induced changes. | The secondary outcome will be measured by using the PEG questionnaire to evaluate participant pain intensity and pain interference at 1, 3, and 6 (V10) months after completion of the intervention. The PEG scores at V10, controlling for baseline (V0) PEG, will be compared between the intervention and placebo groups. Additionally, PEG scores will be assessed between intervention groups at 1 and 3 months after completion of the intervention, which is the timeframe between V0 and V10. | The PEG questionnaire will be obtained at V0, the 1- and 3-month follow-up assessments, and at V10. Therefore, the total timeframe is expected to be approximately s 28-32 weeks apart. |

## Tertiary/Exploratory

| **Objective** | **Brief Description/Justification of Outcome Measure** | **Outcome Measured By** | **Time Frame** |
| --- | --- | --- | --- |
| To investigate the extent to which PBM will increase jaw function | The Standard Movement Task (SMT) that is part of the (DC/TMD) will be used to determine participants’ range of motion and pain upon movement. This SMT assesses the range of jaw movement during opening, functions that are often impacted in participants with TMD pain. Pain is assessed during these maneuvers using a numeric rating scale (0-100), which provides a clinically valid measure of movement-evoked pain. | This exploratory outcome will be measured by comparing the pain intensity and range of motion in millimeters for unassisted and assisted mouth opening. Range of motion and pain intensity at V9, controlling for these measurements at V0, will be compared between the intervention and placebo groups. | These measurements will be made at V0, V5, V9, and V10 during the DC/TMD exam, therefore, these sessions are expected to occur over a 220 day period. |
| To investigate the extent to which PBM will reduce the inflammatory response | Levels of circulating cytokines at each of three timepoints (V1, V5, V9) will be measured. The goal is to evaluate if PBM will reduce pro-inflammatory cytokines at the end of the intervention (V9) compared with baseline (V1). We will also measure cytokines at the intervention midpoint (V5) to evaluate if the inflammatory response begins to change by midway through the intervention. | To measure circulating cytokines in whole blood, we will use the Luminex xMAP multiplexing (Bio-Rad Laboratories, CA, USA) 8-plex (MCP1, IL-1ra, IL-8, IL6, IL-1β, IL10, and TNF-α) following manufacturer’s guidelines. Samples will be analyzed in duplicate to evaluate intersample differences. Cortisol will be measured by immunoassay and used as a control variable.  Cytokine levels at V5 to V9, controlling for levels at V1, will be compared between the intervention and placebo groups. | Blood draws will be collected at V1, V5, and V9, therefore the total time frame is expected to be 4-6 weeks apart. |
| To investigate the extent to which PBM will reduce mechanical pain sensitivity. | Pressure Pain Threshold (PPT) will be measured using an algometer applied to three cranial and two extracranial sites. PPT assessed during V0 will be compared to PPT during V5 (intervention midpoint) and V9. PPTs reflect mechanical pain sensitivity, which is associated with risk for TMD and may be reduced by PBM, which may partially explain its therapeutic effects in TMD. | PPT will be assessed with a handheld algometer bilaterally at temporalis, masseter, TMJ, trapezius, and lateral epicondyle.  PPT at V5 and V9, controlling for PPT at V0, will be compared between the intervention and placebo groups. | The PPT will be performed at V0, V5, V9, and V10, therefore the total time frame is expected to be 220 days. |
| To investigate the extent to which PBM-induced changes in inflammation and pain sensitivity will be associated with the magnitude of reductions in clinical pain, following the PBM intervention | The goal is to evaluate if changes in the level of cytokines and changes in PPT after PBM intervention will be associated with a reduction in clinical pain evaluated by the daily diary, DC/TMD exam, and the PEG. | Change scores for each variable will be computed by subtracting the pre-treatment (V0 or V1) value of the variable from the post- intervention (V9) value and will be compared between the intervention and placebo groups. | For all analyses, change scores will be computed comparing V9 with V0 or V1, therefore the time frame is expected to be approximately 4-8 weeks. |

# STUDY DESIGN

This study is a double-blind, placebo-controlled, randomized trial investigating the efficacy of photobiomodulation (PBM) therapy for chronic TMD pain. A total of 130 participants with chronic TMD, age 18 years and older, will be randomized at the University of Florida, College of Dentistry. Recruitment will include community-based (e.g., advertising across multiple forms of print and electronic media, including posted flyers, radio, newspaper, social media, etc.) and clinic-based strategies. Individuals who express interest in the study will be contacted by trained study staff to complete the pre-screening computer assisted telephone interview (CATI). Initial eligibility criteria will include reporting facial pain for at least 3 months and an average pain intensity rating for the week preceding CATI of ≥30 on a numerical rating scale (0-100). Potential participants who meet the initial eligibility screening criteria will be invited to an in-person visit in which they will undergo consent procedures and a baseline clinical visit (V0) to confirm eligibility and verify their TMD status via clinical examination using the Diagnostic Criteria for Temporomandibular Disorder (DC/TMD) (28, 29). Pressure pain threshold (PPT) and inflammatory markers will also be measured at baseline (V0). Enrolled participants will complete an electronic Daily Symptom Diary (DSD) for at least one week prior to the first intervention visit (V1). At V1, participants with an average pain level from the DSD ≥30 will be randomized to either active or sham PBM, with the first intervention treatment applied at V1. This will be followed by seven more intervention visits, such that all participants will complete two to three intervention visits per week for up to four weeks. A blood draw, PPT, and DC/TMD will be repeated at the intervention midpoint (V5), the post-intervention visit (V9), scheduled between four and seven days after the last intervention visit (V8), and at visit 10 (V10), scheduled about six months after the last intervention visit (V8). Likewise, DSDs will be completed throughout the intervention period and for one week leading up to V10. Also, at one and three months after V9, follow-up data regarding pain intensity and interference will be collected from the participant electronically or over the telephone. The final visit (V10) will occur six months post-intervention to investigate the effectiveness of PBM reducing pain related to TMD up to 6 months after completion of the intervention. The expected duration of subject participation is around 220 days ± 30 Days.

## Photobiomodulation Therapy Intervention

Our PBM intervention aligns with a comprehensive meta-analysis of existing studies, which recommends the use of multiple wavelengths and PBM light sources to be delivered to multiple craniofacial regions, including the TMJ, masticatory muscles, and cervical muscles (26) (see Table 2, Section 6.2). We will first apply PBM to painful musculoskeletal tissue associated with palpable tender points at the temporalis, masseter, sternocleidomastoid and trapezius (2 points per muscle bilaterally). Then we will apply PBM to the nerve pathway at the spinal cord and trigeminal nucleus by treating the C2 to C5 area and peripherally targeting the trigeminal area around the temporomandibular joint (TMJ) bilaterally. Finally, we will target the lymphatic system in the pre-auricular area, cervical chain, and spinal nerve chain bilaterally. We hypothesize that this multimodal PBM intervention that targets different pathophysiologic mechanisms will produce significant and long-lasting analgesic effects. The PBM therapy will be delivered two to three times per week over three to four weeks.

## Sham PBM

When applying PBM therapy, the active intervention device includes heating elements that are perceptible to the participant and the interventionist. The THOR® LX2.3 PBM control unit with multiple probes includes a placebo condition feature that mimics the heating activity of the active intervention and serves to decrease the likelihood of unblinding the participant and interventionist.

The intervention will be delivered by a trained operator who will be blind to the type of intervention to be delivered.

## Randomization

The following criteria will be required for trial eligibility, assessed after the week prior to randomization (at V1): (1) a minimum of 4 of 7 diary entries completed, and (2) the mean average pain score for the week was ≥30 of 100. Participants who meet these criteria will then be block randomized to active versus sham PBM. A permuted block randomization will be used with random block sizes of 4 and 8 and with sex and baseline pain level from the DSD [moderate (30-59) vs. severe (60-100)] treated as stratification factors.

# STUDY POPULATION

## Participant Inclusion Criteria

To be eligible to participate in this study, an individual must meet all of the following criteria (See Table 1 for inclusion criteria by session):

* Male or female, aged 18 years and older
* Meets the CATI pre-screening criteria during pre-screening visit [participant-reported facial pain for at least 3 months and an average pain intensity rating for the week preceding CATI of ≥30 on a numerical rating scale (NRS, 0-100)].
* Willing to provide signed and dated informed consent
* Willing to comply with all study procedures and to be available for the duration of the study
* Meets diagnostic criteria for TMD (Masticatory Muscle disorder, 1 A: Myalgia) during Visit 0
* Completes a minimum of 4 of 7 daily symptom diary (DSD) entries prior to Visit 1 (Randomization visit), and the weekly average pain score on this DSD is ≥30 of 100

|  |  |  |  |
| --- | --- | --- | --- |
| **Table 1: INCLUSION CRITERIA BY SESSION** | | | |
|  | Prescreening | Visit 0 | Visit 1 |
| Medical History | X | X | X |
| Facial Pain for at least 3 months preceding CATI | X |  |  |
| Average NRS pain intensity rating ≥30 for the week preceding CATI | X |  |  |
| Willing to sign Informed consent |  | X |  |
| Meets DC/TMD criteria for myalgia or myalgia and arthralgia |  | X |  |
| Completes a minimum of 4 of 7 Daily Symptom Diary |  |  | X |
| Weekly average pain score on this DSD is ≥30 of 100 |  |  | X |

## Participant Exclusion Criteria

An individual who meets any of the following criteria will be excluded from participation in this study:

* Diagnosis of Rheumatoid Arthritis
* Has a medical condition, laboratory finding, or physical exam finding (e.g., renal failure or dialysis, uncontrolled diabetes mellitus, or uncontrolled seizures) that precludes participation as determined by the investigator
* Has been diagnosed with fibromyalgia, and the most painful area is any part of the body other than the orofacial region.
* Initiated occlusal appliance therapy within 30 days prior to CATI
* Initiated non-pharmacologic therapy, such as acupuncture, biofeedback, and/or TENS within 30 days prior to CATI
* Is in active orthodontic treatment
* Received any injection therapy (e.g., tender or trigger point injections, steroid injections) for the management of pain within 14 days prior to the CATI
* Has a history of facial trauma or orofacial surgery within 6 weeks prior to CATI
* Has a history of psychiatric hospitalization within one year prior to CATI
* Currently pregnant or lactating
* Has a known skin hypersensitivity to light therapy
* Currently being treated with chemotherapy or radiation therapy
* Is undergoing treatment with another investigational drug or treatment initiated within 30 days prior to the Screening and Baseline Visit
* Initiated a new daily prescribed or over-the counter medication for the management of pain within 30 days prior to Screening and Baseline Visit.
* Anything that would place the individual at increased risk or preclude the individual’s full compliance with or completion of the study as determined by the investigator

## Strategies for Recruitment and Retention

Over a 48-month recruitment period, we propose to randomize 130 individuals. To achieve our recruitment goal, we need to randomize 2.7 participants per month. As in our previous studies, we will implement a multimodal recruitment strategy to ensure timely achievement of our planned enrollment. This will include community-based recruitment as well as clinic-based recruitment. Our recruitment strategy has been developed and implemented with support from the Recruitment Center of our Clinical and Translational Science Institute (CTSI). The CTSI Recruitment Center assists with drafting recruitment plans, developing recruitment materials (e.g., flyers, online ads), using social media for recruitment and linking with other local and national recruitment resources.

Community-Based Recruitment: Alachua County, Florida has a population of 269,956 people, 72% of which are 18 years and older. Racial/ethnic diversity is substantial, 20.6% of residents are African American, 10.3% Hispanic, and 60.8% non-Hispanic white. The University of Florida has a large footprint in the community, which facilitates recruitment into research studies. Our community-based recruitment methods will include several strategies. First, we will advertise around our local institutions as well as throughout the local communities, including advertisements in local retail establishments, and in local print media. Second, we will implement a coordinated online and social media recruitment strategy, including Facebook ads. Third, we will participate in community health fairs and education programs sponsored by entities within our University. We have participated in many such events in the past, which have been highly successful. Fourth, we will leverage existing CTSI recruitment resources, including HealthStreet and UFHealth Study Listings. HealthStreet is a community portal of entry for linking and navigating underrepresented populations to opportunities to collaborate with the research community through town halls, focus groups, individual interviews, library use, individual health assessment, and navigation to appropriate research. HealthStreet’s Community Health Workers engage with residents to assess health needs and enroll interested individuals in a research registry. The HealthStreet registry has more than 10,000 individuals from the local area who have agreed to be contacted for research studies. Our protocol will also be listed on UFHealth Study Listings, where potential participants search for appropriate research projects. Individuals performed more than 15,000 searches for research opportunities in 2018. Finally, we will contact individuals from the Pain Research & Intervention Center of Excellence Research Registry. This registry includes more than 1,000 participants who have expressed interest in research participation.

Clinic-Based Recruitment: Potential participants will be recruited from the UF College of Dentistry Faculty Practice and Student Clinics, where individuals with TMD are often evaluated and treated. This includes Dr. Dasilva’s practice. In addition, we will identify potential participants through the CTSI’s Integrated Data Repository, which contains data from the electronic health record. This allows for cohort identification, and UFHealth has implemented consent-to-share, such that a large proportion of participants seen in our clinics have provided consent to be contacted about research. This enables us to request a list of all individuals with a TMD-related diagnosis, whom we can then contact directly to determine their eligibility and interest in study participation.

Retention: We plan to randomize an additional 20% participants to account for attrition. Several approaches to promote retention are incorporated into the study. First, participants are carefully screened to ensure they have availability for multiple study visits, and study staff will clearly communicate the participant’s responsibilities as dictated by the protocol. Also, a written copy of the visit schedule will be provided to each participant after their first visit, which will also include a map to the facility and contact information for study staff. Further, study staff will provide reminder phone calls 24-48 hours before each study visit, or an in-person reminder if the participant has another visit within this window. Also, contact information is confirmed with the participant at each study visit, and multiple modes of contact are always requested. In addition, the following strategies will be used as needed to enhance retention:

* + Emphasize developing strong rapport with each participant by expressing appreciation for their participation and showing personal interest in them as people.
  + Study staff will practice positive communication skills and provide encouragement as participants complete components of the protocol.
  + Maintain consistency in the staff members working and interacting with the participant throughout their participation in the study.
  + After each visit, thank them for their participation and ask whether they have any questions or concerns regarding the next visit.

## Treatment Assignment Procedures

### Randomization Procedures

Participants will be block randomized to active PBM intervention or sham PBM, which involves application of the probes in the absence of any light from the probes used. A permuted block randomization will be used with random block sizes of 4 and 8 and with sex and baseline pain level [moderate (30-59) vs. severe (60-100)] treated as stratification factors.

The statistician will maintain the randomization code. Randomization will be implemented in REDCap at V1. Once a randomization code is generated from REDCap, , the study coordinator and or research staff will record the code on the participants’ record in REDCap, and the study coordinator will set the machine prior to the randomization visit for each participant with the randomization code given. Also, it will be the responsibility of the study coordinator to set the randomization code in the laser machine prior to each subsequent intervention visit.

### Masking Procedures

The statistician will maintain the randomization code list. All other study personnel will be blinded to participants’ intervention assignments throughout the data collection period. The lab manager will be the only one other than the statistician to be unblinded in case of emergency.

The laser device to be used in the study (THOR® LX2.3) includes a Randomizing Switch Box that is preprogrammed with numeric codes for placebo and active intervention. The Study Coordinator and/or research staff will enter the code without knowledge about whether the code is assigned to active or placebo treatment. Also, protective eyewear that absorbs radiation will be worn by intervention staff as well as by participants during both active and placebo therapy. Specifically, the manufacturer provides the participant Thor goggles also emit LED light inside (behind the lenses), which prevents participants from discerning placebo versus active intervention. There is a lead which plugs into a socket on the Randomizing Switch Box to power the LED when the treatment device turns on. Therefore, only the THOR goggles will be worn. These goggles meet the necessary laser safety requirements, and they prevent potential “unblinding” of intervention assignment. Unblinding of a participant before the study is completed will occur only if a participant’s well-being is threatened and is deemed necessary for participant safety. Study participants will be provided with instructions to contact the lab manager and will receive a card with the study manager contact information for emergency situations at Visit 1. For more information refer to section 5.4.1 of the MOP. If oversight boards, such as the IRB or the Medical Monitor, request an unblinded data report during the data collection period, the unblinded biostatistician will generate the report.

## Participant Withdrawal or Discontinuation from Study Procedures/Intervention

### Reasons for Participant Withdrawal or Discontinuation from Study Procedures/Intervention

Participants are free to withdraw from the study at any time upon request.

Participants may choose to discontinue the intervention or study procedure but continue to be followed.

An investigator may discontinue an individual’s participation in an intervention or withdraw an individual from the study if:

* Any clinical adverse event (AE), laboratory abnormality, or other medical condition or situation occurs such that continued participation in the study would not be in the best interest of the participant.
* The participant meets an exclusion criterion (either newly developed or not previously recognized) that precludes further study participation.
* For female participants, reporting becoming pregnant during the study. Female participants must have a negative pregnancy test at the enrollment visit (V0) to meet eligibility criteria for study participation.
* One of the PIs determines it is in the best interest of the study participant to discontinue the intervention or be withdrawn from the study. Participants who undergo surgery requiring general anesthesia and/or hospitalization and that has an expected recovery period of more than a month will be withdrawn from the study.

Investigators will use discretion in making these determinations. If a participant does not return for a scheduled visit, every effort should be made to contact the participant and document the outcome. If the participant withdraws consent, no further evaluations will be performed, and no attempts will be made to collect additional data.

### Handling of Participant Withdrawals from Study or Participant Discontinuation of Study Intervention

Participants who withdraw from the study voluntarily or are withdrawn by an investigator for the reasons described in 5.5.1 after randomization will not be replaced. Study staff will complete a study disposition form, indicating the reason for withdrawal. If unblinding is required, the investigator will follow the unblinding procedures described in the MOP Section 5.4.1. If the study intervention is discontinued for a participant, the participant will continue to complete other study procedures until their participation in the study is completed.

Any participant with an AE that is ongoing at the time of withdrawal will be followed until the event returns to baseline, resolves, or stabilizes. If the AE does not meet these outcomes within 30 days after withdrawal or the end of the study, the participant will be referred to an appropriate practitioner for continued care.

## Premature Termination or Suspension of Study

This study may be suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be provided by the suspending or terminating party to the investigator, funding agency (NIDCR) and any other relevant regulatory authorities. The principal investigator will also promptly inform the IRB and will provide the reason(s) for the termination or suspension.

Circumstances that may warrant termination include, but are not limited to:

* Determination of unexpected, significant, or unacceptable risk to participants.
* Insufficient adherence to protocol requirements.
* Data that are not sufficiently complete and/or evaluable.
* Determination of futility.

If the study is prematurely suspended or terminated, the PI will provide written notification to the investigators, the NIDCR program official, the IRB, and the Medical Monitor.

# STUDY INTERVENTION

## Study Intervention Description

THOR® laser system active intervention arm uses both high irradiance laser and low irradiance LED light. Three types of active probes will be used in this investigation. Laser A (Single Diode Laser) is designed for isolated trigger points and superficial muscles. Laser B (Laser Cluster Laser) is designed for a larger intervention area, targeting nerves for analgesia, deep inflammation (>1 cm below the skin), and deep tissue repair (>1 cm below the skin). Laser C (LED Cluster) is designed for superficial inflammation (<1cm deep).

The THOR® laser device was chosen because it offers all three probes needed for the intervention, and it incorporates a built-in placebo condition, which maintains blinding of both participants and interventionists. The investigational THOR® laser system will be supplied by THOR Photomedicine Ltd. This device has been determined to be non-significant risk by the UF IRB.

## Dosage, Preparation and Administration of Study Intervention

The interventionist will first apply PBM to painful musculoskeletal tissue associated with palpable tender points at the temporalis, masseter, sternocleidomastoid, occipital, and trapezius (2 points per muscle bilaterally). Then we will apply PBM to the nerve pathway at the spinal cord and trigeminal nucleus by treating the C2 to C5 area and peripherally targeting the trigeminal area around the TMJ bilaterally. Also targeting the sternocleidomastoid muscle and trapezius muscle. Finally, we will target the lymphatic system in the pre-auricular area, and cervical chain, bilaterally. The PBM therapy will be delivered two to three times per week, ensuring a total of eight sessions within three to four weeks.

A screenshot of a medical chart

AI-generated content may be incorrect.

Table 2. Summary of treatment parameters and locations.

First, target areas for the intervention will include trigger points on temporalis, masseter, sternocleidomastoid, and trapezius bilaterally using the single laser (Laser A) probe. Next, the neuronal tissues in the trigeminal area, spinous process of C2-C5, broad muscle trapezius and occipital areas will be targeted using the laser cluster (Laser B). Lastly, the blood and lymphatics of preauricular will be targeted using the LED cluster (Laser C), as detailed in Table 2. These intervention targets are based on the most common locations of pain reported by participants with TMD and the putative underlying neuronal and inflammatory mechanisms, and they are consistent with recent evidence-guided recommendations.9

## Modification of Study Intervention Administration for a Participant

If a participant demonstrates high sensitivity to the laser therapy, the intervention will be paused, and the participant will determine whether they are able to continue. If a specific site of stimulation becomes too sensitive to continue, the interventionist will continue treatment with the probe ~15mm from the skin surface or move the probe to an adjacent site in the same area, if feasible. If this is not feasible or the newly chosen site also shows sensitivity, the interventionist will move on to the next stimulation site, as tolerated by the participant. In the next session, the previously sensitive area will be probed to determine if sensitivity is still present. If so, the interventionist will follow the same procedures as in the prior session. Any modifications of the intervention due to sensitivity will be recorded on the Intervention Form eCRF. The same procedure will be followed when a participant presents with a scar, tattoo, or wounded area. For each intervention session, the interventionist will complete the Intervention Documentation Form to document the intervention that was delivered and to note any modifications that were necessary.

## Concomitant Medications/Treatments

In addition to the daily pain level recorded in the DSD, participants will record information pertaining to pain medication intake, including rescue and concomitant medication(s), total daily dose, start date, stop date (if applicable), and primary reason for use. Information about treatment with injection therapy, acupuncture, biofeedback, TENS, and occlusal splint therapy will also be collected for all participants during the study.

### Rescue Medication

Rescue medications described below may be used to alleviate pain and will be recorded in the DSD. Episodic use of a rescue medication will be defined as use for no more than 3 consecutive days and for no more than 18 total days from Visits 1 to 9.

#### Over-the-counter medications

NSAIDs are most often used to manage pain in TMD participants. If a participant is taking an over-the-counter medication daily for pain management for more than 30 days before CATI screening, the participant will be encouraged to continue the same usage of that medication throughout the study. The usage of short-acting, non-prescription analgesics (e.g., NSAIDs, aspirin, and/or acetaminophen) by participants during the study will be considered rescue medication and it will be recorded on the DSD and quantified at each visit, and the usage will be classified as either daily or episodic.

#### Prescription medications

Participants who enter the study on a daily regimen of a prescription medication for pain, more than 30 days before CATI screening, will be encouraged to continue that regimen throughout the study. The usage of all prescription medications for pain during the study will be recorded on the DSD and quantified at each visit.

### Nonpharmacologic Therapy

During the course of the trial, participants will be instructed not to start new nonpharmacologic therapies. If a participant initiates a new therapy, this information will be recorded on the Concomitant Therapies eCRF in REDCap.

## Administration of Intervention

The intervention will be delivered by a trained operator who will be blind to the type of treatment to be delivered. Each participant will undergo 8 intervention sessions, at a frequency of 2-3 times per week, such that intervention sessions will be conducted with intervals of 2-5 days between visits. Each intervention session is expected to last approximately 30 minutes. If a participant intervention visit cannot be rescheduled within 10 days of the prior session, the session will be considered missed. This will be noted as a protocol deviation. The only visit that cannot be missed is visit 5. If more than 10 days have elapsed since visit 4, then the procedures of visit 5 will be performed at the next visit and visit 6 will be skipped.

## Procedures for Training Interventionists and Maintaining Consistency with Delivering Procedural Intervention

PBM Interventionist Certification: Before delivering the intervention, each interventionist (Study coordinator, Dr. Migliorati and Dr. Dasilva) will complete the training offered by the manufacturer (Thor Photomedicine) to ensure thorough understanding of safe use of the device. This training will serve as the initial certification procedure for all interventionists. Then, in collaboration with James Carroll (consultant), the Multiple Principal Investigators (MPIs) will create a training video for delivery of the intervention procedures. The Study Coordinator and any future interventionists who may be added to the study team will study the Manual of Procedures (MOP) and the training video, and they will complete at least three practice sessions under supervision of Dr. Dasilva or Dr. Migliorati. In order to be certified, the interventionist must complete an intervention session demonstrating correct placement and delivery of all intervention components that will be observed by Dr. Dasilva or Dr. Migliorati and recorded in the training log maintained in Sharepoint. To reduce drift, each interventionist will complete a retraining session annually to demonstrate they are delivering the intervention in a consistent manner across all study participants.

DC/TMD Exam Training: Before enrollment starts, Dr. Dasilva, who is a DC/TMD calibrated examiner, will be responsible for training the Study coordinator on the clinical exam. The training will comprise two steps.

The first step will entail training on the following:

1. Calibrating the finger pressure to apply 0.5kg and 1 kg force during palpation
2. Correctly identifying the location of the orofacial muscles and TMJ points to be accessed with the palpation
3. Correctly measuring the free mouth opening, maximum opening, and assisted maximum opening.
4. Being familiar with the sequence of the DC/TMD exam and form

The second part of the training will involve assessing the accuracy of the DC/TMD exam delivered by the study coordinator. To certify that the study coordinator is doing the exam correctly, Dr. Dasilva and the study coordinator will complete the DC/TMD exam in 10 different participants (5 controls and 5 TMD). The goal is to reach 100% concordance of TMD classification between the study coordinator and Dr. Dasilva. If needed, more participants will be recruited to reach the goal.

Dr. Ohrbach (consultant) will assist with the DC/TMD training as needed.

# STUDY SCHEDULE

Below we provide a description and planned timing of each visit. The proposed visit windows are expressed relative to Day 0, which is the date of Visit 1. The goal is to schedule intervention visits two to three times per week. Because participant schedules can be impacted by expected or unexpected events, we anticipate that deviations from the planned visit windows may occur. If a visit falls outside the planned visit window, and subsequent visits also deviate, for reporting purposes, only the first deviated visit will be reported as a single protocol deviation. The subsequent visit windows will be referenced to the date of the deviated visit, and no further protocol deviations will be reported.

## Prescreening (Day -49 to Day-7)

Prescreening may occur by telephone or at a clinic visit, and it may be combined with the Screening and Baseline Visit (Visit 0). After obtaining verbal consent, a brief prescreening interview script using the CATI will be administered to verify basic eligibility information, including eligibility criteria related to the participant’s medical history, and to obtain their contact information. If a participant expresses interest in the study and is eligible based upon initial prescreening, the participant will be scheduled for a Screening and Baseline Visit (Visit 0; Day -21 to -7) within 6 weeks.

**Participants may be re-screened if exclusion criteria are likely to resolve, such as transient or treatable conditions, or temporary use of pain management medications or therapies.**

## Baseline Visit (Visit 0; Day -21 to Day-7)

The following procedures and assessments will be conducted at Visit 0. Items 1 – 7 are collected first, as they are used to determine eligibility. Once eligibility is confirmed (item 8), the remaining procedures are conducted as baseline assessments.

1. Obtain consent for study participation
2. Collect demographic information
3. Obtain medical history
4. Record concomitant medications and therapies
5. Perform a urine pregnancy test in female participants of childbearing potential
6. Administer the TMD-Jaw Functional Limitation Scale
7. Perform TMD examination
8. Review eligibility criteria\*\*, if participant still eligible, items 9 to 15 will be performed. If not participants will be considered screening failures.
9. Administer Questionnaires

* Pain, Enjoyment, General Activity (PEG) Scale;
* Somatic Symptoms (PHQ15) Questionnaire;
* Perceived Stress (Perceived Stress Scale);
* Pain catastrophizing (Pain Catastrophizing Scale);
* Depression (PROMIS Anxiety & Depression short forms);

1. Administer the Pressure Pain Threshold test
2. Collect physical measurements and collect skin tone as described on the MOP section 5.5.8.2
3. Collect vital signs
4. Dispense new Daily Symptom Diaries.
5. Schedule next visit

*\*\*Eligibility criteria*: For women, not pregnant. For all participants: Meet DC/TMD criteria for Myalgia or Myalgia and Arthralgia and meet no exclusion criteria.

## Intervention Visits

### Randomization Visit (Visit 1; Day 0)

The following procedures and assessments will be conducted at Visit 1:

1. Review medical history
2. Record concomitant medications and therapies
3. Collect Daily Symptom Diaries if in paper form
4. Assess compliance with Daily Symptom Diaries. If the number of entries fall below the cut off or the pain score is very close to 30, another week of DSD can be given to the participant prior to excluding them from participation.
5. Perform a urine pregnancy test in female participants of childbearing potential
6. Review eligibility criteria\*\*, if participant still eligible, items 7 to 14 will be performed. If not participants will be withdrawn.
7. Randomize participant in REDCap
8. Perform Blood draw
9. Administer Intervention Expectations Questionnaire.
10. Administer the Pre-Intervention Questionnaire
11. Assess and record AEs
12. Deliver the intervention (code previously sent by statistician)
13. Administer the Post-Intervention Questionnaire
14. Dispense new Daily Symptom Diaries
15. Schedule next visit

*\*\*Eligibility criteria*: For women, not pregnant. For all participants: At least 4 entries in the DSD must be completed satisfactorily within the week preceding this visit for the participant to meet the eligibility criterion for randomization. The participant must also have an average pain intensity score of ≥ 30 as calculated from the DSD entries, to meet the eligibility criteria.

### Intervention Visits 2 (Visit 1+4 Days ± 3 Days), 3 (Visit 2+4 Days ± 3 Days), and 4 (Visit 3+4 Days ± 3 Days)

1. Perform a urine pregnancy test in female participants of childbearing potential
2. Review Medical History
3. Review study continuation criteria\*\*\*\*
4. Administer the Pre-Intervention Questionnaire
5. Record concomitant medications and therapies
6. Assess and record AEs
7. Collect Daily Symptom Diaries
8. Assess compliance with Daily Symptom Diaries
9. Deliver intervention
10. Administer the Post-Intervention Questionnaire
11. Review DSD
12. Dispense new Daily Symptom Diaries
13. Schedule next visit

*\*\*\*\*Study continuation criteria*: No new health conditions listed in the exclusion criteria that pose a safety risk or threat to study validity (e.g., initiation of chemotherapy, facial trauma).

### Mid-Intervention Visit (Visit 5; Visit 4+4 Days ± 3 Days)

The following procedures and assessments will be conducted at the Mid-Intervention visit:

1. Perform a urine pregnancy test in female participants of childbearing potential
2. Review Medical History
3. Review study continuation criteria\*\*\*\*
4. Administer the Pre-Intervention Questionnaire
5. Administer the Pain, Enjoyment, General Activity (PEG) Scale Questionnaire
6. Administer the TMD-Jaw Functional Limitation Scale
7. Perform TMD examination
8. Administer the Pressure Pain Threshold
9. Record concomitant medications and therapies
10. Assess and Record AEs
11. Blood draw
12. Deliver intervention
13. Administer the Post-Intervention Questionnaire
14. Review DSD
15. Dispense new Daily Symptom Diaries
16. Schedule next visit

*\*\*\*\*Study continuation criteria*: No new health conditions listed in the exclusion criteria that pose a safety risk or threat to study validity (e.g., initiation of chemotherapy, facial trauma).

### Intervention Visits 6 (Visit 5+4 Days ± 3 Days), 7 (Visit 6+4 Days ± 3 Days), and 8 (Visit 7+4 Days ± 3 Days)

1. Perform a urine pregnancy test in female participants of childbearing potential
2. Review Medical History
3. Review study continuation criteria\*\*\*\*
4. Administer the Pre-Intervention Questionnaire
5. Record concomitant medications and therapies
6. Assess and record AEs
7. Collect Daily Symptom Diaries
8. Assess compliance with Daily Symptom Diaries
9. Deliver intervention
10. Administer the Post-Intervention Questionnaire
11. Review DSD
12. Dispense new Daily Symptom Diaries
13. Schedule next visit

*\*\*\*\*Study continuation criteria*: No new health conditions listed in the exclusion criteria that pose a safety risk or threat to study validity (e.g., initiation of chemotherapy, facial trauma).

### Visit 9 – Post Intervention Assessment Visit (Visit 8+5 Days ± 9 Days)

The following procedures and assessments will be conducted at the Post-Intervention assessment visit:

1. Review medical history
2. Assess and record AEs
3. Administer the Pain, Enjoyment, General Activity (PEG) Scale Questionnaire
4. Administer Jaw Functional Limitation Scale
5. Administer the Intervention Experiences Questionnaire.
6. Record concomitant medications and therapies
7. Perform TMD examination
8. Dispense new Daily Symptom Diaries (DSD)
9. Review DSD
10. Administer the Pressure Pain Threshold
11. Blood draw

## 1- and 3-Month Post-Intervention Follow-Up Assessments (Remote Data Collection timepoint 1 – Visit 9+30 Days ± 15 Days and Remote Data Collection timepoint 2 – Remote Data Collection timepoint 1+60 Days ± 15 Days)

At 1 and 3 months after V9, participants will complete the following assessments via computer or telephone.

1. Administer the Pain, Enjoyment, General Activity (PEG) Scale Questionnaire
2. Record concomitant medications and therapies
3. Dispense new electronic or paper Daily Symptom Diaries (DSD)
4. Review Daily Symptom Diaries
5. Assess and record AEs

## Final Study Visit – Visit 10 (Visit 9+180 Days ± 30 Days)

The following procedures and assessments will be conducted at the final follow up visit 10:

1. Administer the Pain, Enjoyment, General Activity (PEG) Scale Questionnaire
2. Record concomitant medications and therapies
3. Review Daily Symptom Diaries
4. Assess and record AEs
5. Administer the TMD-Jaw Functional Limitation Scale
6. Perform TMD examination
7. Administer the Pressure Pain Threshold
8. Blood draw

Since significant life changes (e.g., relocation, job changes) may occur within six months after the last in-person treatment, some participants may be unable to attend the final study visit (V10). To mitigate this, we will collect questionnaires remotely. If this is not possible, the visit will be marked as missed and reported as a protocol deviation.

## Withdrawal Visit

If a participant is withdrawn before study completion, prior to the withdrawal of consent and with the participant’s permission, an Early Termination Visit may be conducted, either during a scheduled or unscheduled visit. Each of the procedures and assessments described for Visit 9 (Section 5) will be conducted at the Early Termination Visit. The Intervention Experiences Questionnaire will also be completed.

## Unscheduled Visit

An unscheduled visit may occur at the discretion of the investigator. An unscheduled visit would be completed if needed to collect data related to a safety event. All unscheduled visits will be documented in the participant’s study record.

# STUDY PROCEDURES/EVALUATIONS

## Study Procedures/Evaluations

Additional details for all study procedures are described in the MOP, which will be provided to study staff prior to study start-up. Study procedure overviews are provided below. Data collected during these procedures will be entered into electronic case report forms within REDCap.

## Clinical Visit Procedures

### Medical History

The medical history for each participant will be obtained by questionnaire supplemented with an interview and entered onto the Medical History eCRF in REDCap at Baseline (Visit 0). Medical history will be reviewed and updated at each visit, up to V9. Medical history information to be collected is primarily related to inclusion/exclusion criteria, including: current medical conditions, active treatments for TMD, history of facial trauma or orofacial surgery, history of psychiatric hospitalization within the past year, pregnancy, hypersensitivity to laser therapy, current chemotherapy or radiation therapy. Current treatment with another investigational drug or treatment, new daily prescription medication or treatment for the management of pain and duration of treatment will be collected to assess participant eligibility. At each visit after the randomization visit (V1), concomitant medications and/or therapies will be recorded in the DSD and classified as daily or episodic (see Section 6.4).

### Vital Signs and Physical Measurements

A physical examination will be performed at the Baseline Visit (Visit 0) and will include height and weight measurements. Vital signs will be also collected at Visit 0. After the participant has rested in a chair for 10 minutes, trained personnel will take 3 readings of the participant’s systolic blood pressure, diastolic blood pressure, and heart rate at 1-minute intervals.

### Pregnancy Test

Because the effect of PBM is unknown in pregnant women and fetus, female participants of childbearing potential will undergo a urine pregnancy test (instant type) at V0 and at all intervention visits. The results will be read by a member of the study staff. The urine will be discarded after the test.

### Blood Draw

Staff trained in phlebotomy will obtain up to 40 milliliters of blood by venipuncture at Visits 1, 5, 9, and 10. All blood tubes will be processed to obtain plasma. Plasma for cytokine, and cortisol analyses will be divided into aliquots and placed in storage at -80ºC at UF.

### Skin tone

Due to potential differences in sensitivity to PBM based on skin tone, we will record skin tone using the [Monk Skin Tone Scale](https://skintone.google/get-started#:~:text=MST%20Orbs%20are%2010%20colored,real%20world%20and%20in%20images.). The Monk Scale will be used to record the participant’s skin tone at two sites (right cheek, and right side of the neck halfway between the occipital bone and the trapezius). This will be entered on the V0 eCRF in the REDCap system.

For more detail, see MOP section 5.5.8.2

### Temporomandibular Disorder Examination

Trained and calibrated study staff will perform a TMD examination at the Baseline Visit (V0), V5, V9 and V10 based on the Diagnostic Criteria for Temporomandibular Disorder (DC/TMD) (29). The examination assesses pain in masticatory muscles and the TMJ in response to palpation and maneuvering of the jaw. The examiner will bilaterally palpate the extraoral masticatory muscles (temporalis and masseter) and TMJ. For eligibility, the participant must meet the following criteria on the DC/TMD exam: 1) reported pain or ache in the jaw, temple, face, preauricular area, or inside the ear that is changed by jaw function; and 2) finding(s) of TMD myalgia, or myalgia and arthralgia classified according to the DC/TMD criteria during the examination.

### Pressure Pain Threshold Measurements

Trained study staff will perform the PPT measurements at V0, V5, V9 and V10. The PPT will be assessed bilaterally over the temporalis, masseter, and trapezius muscles, the TMJs, and the lateral epicondyles with a pressure algometer as described in the MOP section 5.6.12. The PPT will be defined as the amount of pressure at which the participant first perceives the stimulus to be painful. Repeated assessments will be performed at each site until 3 trials differing by less than 30 kPA are obtained, or 5 assessments are administered. The mean of the 2 closest values will be recorded as the threshold estimate. Pressure stimuli will be delivered at an approximate rate of 30 kPA/s. The cutoff pressure for all sites will be 600 kPA. The values from the right and left sides will be averaged to obtain a single PPT value per anatomical site.

### Daily Symptom Diary (DSD)

Participants will be asked to complete the DSD electronically at the end of each day starting at Visit 0 and ending at Visit 9, and for one week prior to 1-month follow up and 3-month follow up and V10. Participants who complete at least 4 out of the 7 entries per week, and report a pain rating of ≥30 by Visit 1 will be considered eligible for study participation (inclusion criterion). The diary requests information about the participant’s pain intensity (reported on a 0-100 numeric rating scale) and duration (reported on a 0-100 percentage scale), fatigue, sleep, and somatic symptoms. Compliance with the DSD will be defined as completion of at least 4 daily entries within a 7-day time period. Any noncompliance found during Visits 2 through 9 will be reported as protocol deviations.

### Pain, Enjoyment, General Activity (PEG) Questionnaire

This self-administered questionnaire comprises 3 separate numerical scales. Each scale has ratings ranging from 0-10. The first scale asks individuals to rate their pain, on average, over the past week. The second scale asks individuals to rate how pain has interfered with their enjoyment of life in the past week. The third scale asks individuals to rate how pain has interfered with their general activities (30). The PEG will be administered at V0, V5, V9, 1- and 3-month follow-up, and at V10.

### Jaw Functional Limitation Scale

The Jaw Functional Limitation Scale (JFLS) will be administered electronically to assess jaw function disability and it is incorporated into Axis II of the Diagnostic Criteria for Temporomandibular disorder (DC/TMD). The JFLS will be administered at V0, V5 (prior to intervention delivery), V9 and V10. The instrument comprises 3 constructs for assessing functional status of the masticatory system; the 3 scales exhibit properties that are ideal for both research and patient evaluation in patient groups with a range of functional limitations of the jaw.( [Ohrbach R et al, 2008](https://pubmed.ncbi.nlm.nih.gov/18780535/)).

### Intervention Expectations  Questionnaire

The Intervention Expectations Questionnaire will be administered electronically to access participant expectations regarding outcomes of the intervention. This questionnaire will be administered in the Visit 1 before the randomized treatment is delivered.

### Pre-Intervention Questionnaire

The Pre-Intervention Questionnaire will be administered electronically to assess current facial pain and screen for AEs. Trained study staff will review participant responses to questions about the occurrence and severity of expected AEs known to be associated with PBM. The Pre-Intervention Questionnaire will be administered at all intervention visits, as described in the MOP (section 5.6.17.3)). If a symptom has emerged or worsened, study personnel will evaluate the symptom for a possible AE.

### Post-Intervention Questionnaire

The Pos-Intervention Questionnaire will be administered electronically to assess facial pain and screen for AEs post PBM treatment. Trained study staff will review participant responses to questions about the occurrence and severity of expected AEs known to be associated with PBM. The Pos-Intervention Questionnaire will be administered at all intervention visits, as described in the MOP (section 5.6.17.3). If a symptom has emerged or worsened, study personnel will evaluate the symptom for a possible AE

### Intervention Experiences Questionnaire

The Intervention Experiences Questionnaire will be completed by the participant electronically at Visit 9 to assess the participant’s experiences of the intervention. This measure includes questions about participant satisfaction with the intervention experience, global impressions of change, perceived treatment allocation, and whether they would repeat the treatment given the opportunity.

### Psychological Questionnaires

The questionnaires that will be utilized are listed in the Schedule of Events ([Appendix A](#AppendixA)) and are briefly described below. Copies of the questionnaires are reproduced in the attachments. All questionnaires are self-administered electronically via REDCap. The questionnaires will be administered at V0.

Somatic Symptoms (PHQ15)is a self-administered version of the PRIME-MD diagnostic instrument for common mental disorders. The PHQ-15 comprises 15 somatic symptoms from the PHQ, each symptom scored from 0 (“not bothered at all”) to 2 (“bothered a lot”)(31).

Perceived Stress Scale. This self-administered questionnaire assesses symptoms and perceptions of stress and produces an overall perceived stress score (32).

Pain Catastrophizing Scale. This scale is a 13 item self-report and self-administered measure designed to assess catastrophic thinking related to pain among adults with or without chronic pain (33).

PROMIS Anxiety & Depression short forms. Each of these measures includes 8 items that assess symptoms of anxiety and depression experienced over the past seven days. This questionnaire will be self-administered (34).

## Laboratory Procedures/Evaluations

### Special Assays or Procedures

The Luminex xMAP multiplexing (Bio-Rad Laboratories, CA, USA) 8-plex (MCP1, IL-1ra, IL-8, IL6, IL-1β, IL10, and TNF-α) will be used to measure circulating cytokines in whole blood, following manufacturer’s guidelines. Samples will be analyzed in duplicate to evaluate intersample differences. Cortisol will be measured by immunoassay and used as a control variable.

### Specimen Preparation, Handling, and Storage

Blood draws will occur as described above in Section 8.2.4. Standard operating procedures for specimen collection, preparation, storage, and analysis will be provided in the MOP section 9. Briefly, after collecting the blood sample, trained staff will process the blood and store the samples in a -80 degrees freezer. The blood specimens will be used to assess cortisol and cytokine levels, and specimens will be analyzed in batches. The storage of all samples will be tracked by a web-based specimen tracking system in REDCap.

# ASSESSMENT OF SAFETY

## Specification of Safety Parameters

Safety monitoring for this study will focus on unanticipated problems involving risks to participants, including unanticipated problems that meet the definition of a serious adverse event (SAE). In addition, adverse events (AEs) including SAEs will be recorded, and the MPIs will monitor these events to grade severity, relationship to the study intervention, and assess whether the nature, severity, or frequency is unexpected. Safety data will be collected at all study visits through clinical examination by study personnel and solicitation of adverse events from participants at each study visit. Study personnel will carefully monitor participant safety throughout the study. Known study risks are described in [Section 2.3](#_Potential_Risks_and)

### Unanticipated Problems

The Office for Human Research Protections (OHRP) considers unanticipated problems (UP) involving risks to participants or others to include, in general, any incident, experience, or outcome that meets all of the following criteria:

1. Unexpected in terms of nature, severity, or frequency given (a) the research procedures that are described in the protocol-related documents, such as the IRB-approved research protocol and informed consent document; and (b) the characteristics of the participant population being studied;
2. Related or possibly related to participation in the research (“possibly related” means there is a reasonable possibility that the incident, experience, or outcome may have been caused by the procedures involved in the research); and
3. Suggests that the research places participants or others at a greater risk of harm (including physical, psychological, economic, or social harm) than was previously known or recognized.

### Adverse Events

An adverse event is any untoward or unfavorable medical occurrence in a human subject, including any abnormal sign (for example, abnormal physical exam or laboratory finding), symptom, or disease, temporally associated with the subject’s participation in the research, whether or not considered related to the subject’s participation in the research.

### Serious Adverse Events

A serious adverse event (SAE) is one that meets one or more of the following criteria:

* Results in death
* Is life-threatening (places the subject at immediate risk of death from the event as it occurred)
* Results in participant hospitalization or prolongation of existing hospitalization
* Results in a persistent or significant disability or incapacity
* Results in a congenital anomaly or birth defect
* An important medical event that may not result in death, be life threatening, or require hospitalization may be considered an SAE when, based upon appropriate medical judgment, the event may jeopardize the subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

## Time Period and Frequency for Event Assessment and Follow-Up

One of the MPIs or their designee will record all events with start dates occurring any time after informed consent is obtained until 7 (for non-serious AEs) or 30 days (for SAEs) after the last day of study participation. At each study visit, the investigator will inquire about the occurrence of AE/SAEs since the last visit. Events will be followed for outcome information until resolution or stabilization. If the AE does not meet these outcomes within 30 days after discontinuation or the end of the study, the participant will be referred to an appropriate practitioner for continued care.

## Characteristics of an Adverse Event

Each event will be recorded on an Adverse Event eCRF that includes assessment of the characteristics defined below. These characteristics, along with the frequency of an event’s occurrence, will be considered in determining if the event is a UP.

### Relationship to Study Intervention

The MPIs will make the determination of the relationship of an AE to the study intervention according to the following guidelines:

1. Related (Possible, Probable, Definite)
   1. The event is known to occur with the study intervention, and/or
   2. There is a temporal relationship between the intervention and event onset and/or
   3. The event abates when the intervention is discontinued, and/or
   4. The event reappears upon a re-challenge with the intervention.
2. Not Related (Unlikely, Not Related)
   1. There is no temporal relationship between the intervention and event onset, and/or
   2. An alternate etiology has been established.

### Expectedness

The Study MPIs will determine whether an AE is expected or unexpected. An AE will be considered unexpected if the nature, severity, or frequency of the event is not consistent with the risk information previously described for the intervention.

### Severity of Event

The severity of each AEwill also be graded as follows:

* **Mild:** Awareness of signs or symptoms, but easily tolerated and are of minor irritant type causing no loss of time from normal activities. Symptoms do not require therapy or a medical evaluation; signs and symptoms are transient.
* **Moderate**: Events introduce a low level of inconvenience or concern to the participant and may interfere with daily activities, but are usually improved by simple therapeutic measures; moderate experiences may cause some interference with functioning
* **Severe**: Events interrupt the participant’s normal daily activities and generally require systemic drug therapy or other treatment; they are usually incapacitating

## Reporting Procedures

### Unanticipated Problem Reporting

Incidents or events that meet the OHRP criteria for UPs require the creation and completion of an unanticipated problem report form. OHRP recommends that investigators include the following information when reporting an adverse event, or any other incident, experience, or outcome as an unanticipated problem to the IRB:

* appropriate identifying information for the research protocol, such as the title, investigator’s name, and the IRB project number;
* a detailed description of the adverse event, incident, experience, or outcome;
* an explanation of the basis for determining that the adverse event, incident, experience, or outcome represents an unanticipated problem;
* a description of any changes to the protocol or other corrective actions that have been taken or are proposed in response to the unanticipated problem.

To satisfy the requirement for prompt reporting, UPs will be reported to the IRB via the IRB’s online Reportable Events application using the following timeline:

* Unanticipated problems that are serious adverse events will be reported to the IRB within five (5) working days of the investigator becoming aware of the event.
* Any other unanticipated problem will be reported to the IRB within five (5) working days of the investigator becoming aware of the problem.

All unanticipated problems will be reported to NIDCR concurrently with reporting to the IRB. These reports will be made to NIDCR’s centralized reporting system via Rho Product Safety:

* Product Safety Fax Line (US):  1-888-746-3293
* Product Safety Fax Line (International):  919-287-3998
* Product Safety Email: [rho\_productsafety@rhoworld.com](mailto:rho_productsafety@rhoworld.com)

General questions about UP reporting can be directed to the Rho Product Safety Help Line (available 8:00AM – 5:00PM Eastern Time):

* US: 1-888-746-7231
* International: 919-595-6486

### Serious Adverse Event Reporting

Any AE meeting the specified Serious Adverse Event criteria will be submitted on an SAE form to NIDCR’s centralized safety system via Rho Product Safety. This report may be sent by fax or email. Once submitted, Rho Product Safety will send a confirmation email to the investigator within 1 business day. The investigator should contact Rho Product Safety if this confirmation is not received. This process applies to both initial and follow-up SAE reports.

SAE Reporting Contact Information:

* Product Safety Fax Line (US):  1-888-746-3293
* Product Safety Fax Line (International):  919-287-3998
* Product Safety Email: [rho\_productsafety@rhoworld.com](mailto:rho_productsafety@rhoworld.com)

General questions about SAE reporting can be directed to the Rho Product Safety Help Line (available 8:00AM – 5:00PM Eastern Time):

* US: 1-888-746-7231
* International: 919-595-6486

The study’s clinically responsible individual will complete a Serious Adverse Event Form and submit via fax or email within the following timelines:

* All deaths and immediately life-threatening events, whether related or unrelated, will be recorded on the Serious Adverse Event Form and submitted to Product Safety within five (5) working days of site awareness.
* Serious adverse events other than death and immediately life-threatening events, regardless of relationship, will be reported by fax within five (5) working days of site awareness.

All SAEs will be followed until resolution or stabilization.

### Events of Special Interest

Any clinically significant worsening of a medical condition (other than TMD) established at baseline or the development of any new clinically significant medical condition during the study will be considered an AE and will be reported as described previously.

### Reporting of Pregnancy

Pregnancy will be recorded on a case report form within the REDCap system if it begins any time during the study. Pregnancy will not be regarded as an SAE unless there is suspicion that the study intervention may have interfered with the effectiveness of a contraceptive medication and the event meets the criteria for an unanticipated problem. Results of all positive pregnancy tests will be given to the participant and/or her legally authorized representative, as appropriate.

Participants who become pregnant before randomization will be withdrawn from the study. If a pregnancy occurs after randomization, the intervention will be discontinued and the participant will be referred for appropriate care. If the participant is willing, data collection will continue, and the participant will be followed for safety until a pregnancy outcome is reached. If the pregnancy results in an outcome other than a normal birth or elective abortion of a healthy fetus, it will be reported as an SAE.

## Halting Rules

The study may be temporarily halted due to safety events. Circumstances that may results in halting the trial include, but are not limited to, the following:

* More than one AE graded as severe that is determined to be possibly related to the intervention.
* More than one occurrence of the same SAE determined to be possibly related to the intervention.
* Increased frequency of safety events over the course of the trial.

After complete assessment of the occurrence of the safety event(s), a determination may be made to terminate the trial or to continue the trial, as determined by the PI, the NIDCR program official or medical monitor, or the IRB.

# STUDY OVERSIGHT

In addition to the PI’s responsibility for oversight, study oversight will be under the direction of a Medical Monitor. The PI will submit a report every 6 months to the NIDCR Medical Monitor for review. This report will include data regarding enrollment and retention, unanticipated problems and protocol deviations, disposition of biospecimens, outcome measures, quality management findings and other relevant parameters. If necessary, additional steps may be taken to ensure data integrity and protocol compliance.

# CLINICAL SITE MONITORING

No outside clinical site monitoring will be employed for this study. The Principal Investigator(s) and staff will closely monitor the subjects as they progress through the study. They will monitor and evaluate study processes and documentation based on the International Council for Harmonisation (ICH), E6: Good Clinical Practice guidelines (GCP), and internal quality management plans. The NIDCR reserves the right to conduct independent clinical site monitoring as necessary.

# STATISTICAL CONSIDERATIONS

## Study Hypotheses

### ***Primary Hypothesis***

Our primary hypothesis is that, compared to a credible placebo condition, the multimodal PBM intervention will significantly reduce TMD pain immediately following the intervention.

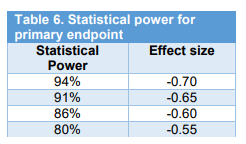
### Secondary Hypothesis

Our secondary hypothesis is that, compared to the placebo condition, participants who receive multimodal PBM will have more favorable secondary endpoints at up to 6 months after completion of the intervention as follows: a) less pain intensity & interference 1,3 and 6 months after intervention, and b) lower DSD pain intensity ratings 6 months after intervention.

### Exploratory hypothesis

Our exploratory hypotheses are that our multimodal PBM will increase mechanical pain thresholds and jaw function measure via a standardized movement task (immediately and 6 months after intervention); decrease levels of circulating pro-inflammatory cytokines (immediately after intervention); and that PBM-induced reductions in mechanical pain sensitivity and inflammation midway through and immediately after intervention will predict greater improvements in clinical pain immediately following PBM.

## Sample Size Considerations

For the primary endpoint (average daily pain) we assume the standardized effect size is -0.55 based on findings reported in a recent meta-analysis (26). We plan to randomize 130 participants (65/group). Assuming a 20% attrition rate, we will have 104 subjects (52/group) for the analysis at the end of the study. The statistical power with two-sided type I error rate of 0.05 is 80% based on a two-sample T test. We also provide the statistical power for different possible effect sizes in Table 6. A 30% reduction in pain has been reported as the minimally clinically significant effect (35). Assuming an average pre-intervention pain rating of 5/10, and a standard deviation of 2.5 (as in our pilot study), a 30% reduction in pain would be equal to a standardized mean difference of 0.6. Thus, we have more than adequate power for detecting a clinically significant reduction in pain. Because limited data exist data to determine an effect size for the secondary and tertiary objectives, we have powered the study on our primary objective. However, given our power to detect a medium effect size (see Table 6), we have sufficient power to detect changes that are of practical significance for all secondary (i.e., PEG scores, pain intensity from DSD at 6-month follow-up) and exploratory (i.e., jaw function, PPT, inflammatory markers) outcomes.

Randomization: A permuted block randomization will be used with random block sizes of 4 and 8 and with sex and baseline pain level [moderate (30-59) vs. severe (60-100)] treated as stratification factors. The intervention will then be delivered by a trained operator that will be blind to the type of intervention to be delivered.

## Planned Interim Analyses (if applicable)

No interim analyses are planned.

## Final Analysis Plan

Intent-to-treat (ITT) analysis will be performed for all analyses. The ITT sample includes all the subjects that received randomization regardless of non-compliance. A per-protocol (PP) analysis will also be considered as a complementary to the ITT analysis. The PP sample includes those who were compliant with the protocol (i.e., completed at least 75% of treatment visits).

Data transformation: The distributions of all variables will be examined before entering the analyses models. Transformations such as natural-logarithm or Box-Cox transformations will be applied to continuous variables as needed to comply with normality assumptions. All binary variables will be coded 1/0 to indicate the two categories, and categorical variables will be converted into dummy variables. Generalized linear models or generalized linear mixed effects models will be used for non-continuous outcomes.

Covariates adjustment: Since the randomization will be stratified by sex and baseline pain level [moderate (30-59) vs. severe (60-100)], those two variables will always be adjusted in all regression models to match the design so that the advantages of stratification can be achieved in the analysis results. In theory, we may not need to adjust for any other potential confounders because of the randomized design, which is supposed to balance their distributions across the two treatment arms. However, we will examine the distributions of potential confounders such as race/ethnicity, age, psychological factors including Pain Catastrophizing (Pain Catastrophizing Scale Score), Anxiety (PROMIS Anxiety Score), Depression (PROMIS Depression Score), Perceived Stress Scale Score and Treatment Expectations Score, assessed via questionnaire at V0 Those with significantly different distributions between the two arms will be adjusted in the regression models. In addition, we will also adjust for those variables (if any) that are associated with the outcome variable in the regression models to increase the efficiency of the statistical estimation for the treatment effects.

**Primary Objective**: We hypothesize that compared to a credible placebo condition, the PBM intervention will significantly reduce TMD pain. For this primary endpoint, to evaluate the change in TMD related pain immediately after PBM/placebo intervention, the dependent variable will be the 0 to 100 average of daily pain ratings on the numerical rating (0 = no pain, 100 = worst pain imaginable) from the Daily Symptom Diary. Daily pain ratings averaged over one week prior to V1 will be treated as the baseline variable. The average daily pain one-week prior to V9 will be treated as the endpoint. Transformations such as natural-logarithm will be applied on the outcome variable if needed to comply with normality assumptions. A linear regression model will be employed to test the association between average daily pain at V9 and intervention group (PBM vs placebo), adjusting for average daily pain at baseline. This approach will allow us to control for potential confounders (e.g., demographic variables, psychological factors, intervention expectations). We will also consider linear mixed effect models (LMM) as the primary approach to handle repeated measurements. An advantage of LMM is that it can provide valid estimates if data are missing at random. An alternative to LMM is GEE to analyze the repeated measurements in relation to the intervention groups. GEE requires data missing completely at random. If there are no or very few missing data, GEE will be used as the primary method for the longitudinal data analysis.

**Secondary Hypotheses**: We hypothesize that, compared to the placebo intervention arm, participants in the PBM treated arm will have more favorable secondary endpoints at up to 6 months after completion of the intervention as follows: a) lower pain intensity & interference collected by the PEG questionnaire, b) lower jaw functional disability collected by JFLS questionnaire c) longer analgesia based on pain ratings from the DSD for the week prior to V10. Total PEG Scores will be computed per the instructions for the instrument. For this hypothesis, in one model PEG Scores measured at 1, 3 and 6 months after the intervention will be analyzed as the outcome variable. For this analysis, a linear regression model will be used to test the intervention effect at 1, 3 and 6 months separately where baseline PEG scores and potential confounders (e.g., demographic variables, psychological factors, intervention expectations) will be adjusted. LMM and GEE (if needed) will also be employed to test the overall long-term effect by analyzing the measurements at 1, 3 and 6 months in the same model. Similar analyses will be performed using the JFLS score as the dependent variable, to assess intervention effects on jaw functional disability. In another model, pain intensity ratings from the DSD averaged over the week prior to the 6-month follow-up visit will be analyzed as the outcome variable. Again, a linear regression model will be used to test the intervention effect at 6 months where baseline DSD pain intensity and potential confounders (e.g., demographic variables, psychological factors, intervention expectations) will be adjusted.

**Exploratory hypothesis**: We hypothesize that multimodal PBM will increase mechanical pain thresholds and jaw function (immediately after and 6 months after intervention) and decrease levels of circulating pro-inflammatory cytokines (immediately after intervention); and PBM-induced reductions in mechanical pain sensitivity and inflammation measured midway through and immediately after intervention will predict greater improvements in clinical pain immediately following PBM. For this hypothesis, cytokine levels measured in blood collected at Visit 1 will be treated as the baseline variable. The cytokine level at Visit 5 will be treated as midpoint and cytokine levels at Visit 9 will be treated as the endpoint. A linear regression model will be employed to test the level of cytokines at V1, V5 and V9 separately (PBM vs placebo) and potential confounders (e.g., demographic variables, psychological factors, intervention expectations) will be controlled in the model. We will use a potential outcomes (PO, i.e., counterfactual-outcomes) mediation analysis approach to estimate and test whether changes in inflammation and pain sensitivity measured at V5 (half-way through intervention) mediate the effect of the intervention condition on average daily pain at V9. Mediation analyses that use mediator variables ascertained at intermediate time points support stronger causal inferences. However, we will also consider changes in pain sensitivity and inflammation from V0 and V1, respectively, to V9, as we recognize that intervention effects on these potential mediators may not fully emerge until the full dose of the intervention has been delivered. PO mediation analysis is more flexible than traditional mediation analysis approaches in terms of allowing interaction between the independent variable and mediators and non-linear indirect effects. In the mediation model, the intervention is the independent variable, inflammation and pain sensitivity will be the mediators, and the outcome variable is average daily pain from the DSD. Baseline values and potential confounders will be adjusted in the mediation model.

**Missing data:** We will test for differences in baseline characteristics between participants lost to follow-up versus those who are not using the t-test and Fisher’s exact test. Missing data patterns will also be evaluated including the frequency and percentage of those missing for each variable and the distribution of the number of variables missing. In addition, data collected to the point of lost to follow-up will be compared to the data of those who complete the study to examine possible missing data mechanisms, e.g., missing completely at random, missing at random, or missing not at random. In situations where missing data occurs, we will document the reasons for the missing data whenever possible. While data may not be missing completely at random, it may be reasonable to assume that data are missing at random. If this is the case, LMM or GLMM approaches can generate valid results. Multiple model-based imputation methods will be applied to account for the missing data, if needed.

# SOURCE DOCUMENTS AND ACCESS TO SOURCE DATA/DOCUMENTS

Data will be collected on electronic CRF (eCRFs) entered directly into REDCap, and consequently the eCRF will serve as the source documents, with exception of laboratory data. Data will include clinical examination data, safety event data, and questionnaire responses. Blood biospecimens will undergo laboratory analyses, which will be recorded in electronic form (e.g., in an Excel spreadsheet) to be used in data analyses.

Study staff will maintain appropriate medical and research records for this study, in compliance with ICH E6, Section 4.9 and regulatory and institutional requirements for the protection of confidentiality of participants. Study staff will permit authorized representatives of NIDCR and regulatory agencies to examine (and when required by applicable law, to copy) research records for the purposes of quality assurance reviews, audits, and evaluation of the study safety, progress, and data validity.

# QUALITY CONTROL AND QUALITY ASSURANCE

Quality control is the ongoing, concurrent review of data collection forms for completion and logic. Quality assurance is a comprehensive, retrospective review of all components of research records to assess adherence to protocol, standard operation procedures, and regulations and to evaluate the accuracy of the records. Quality management is the process of assessing the quality of processes within a system and encompasses quality assurance and quality control.

The quality management processes will include, but will not be limited to, the following:

* A study start-up list of tasks that must be completed prior to the first participant visit
* Training of staff on the protocol, study procedures, and use of the eCRFs and REDCap
* Documentation and tracking of training for each staff member on GCP, Human Subjects Protections (HSP), and all aspects of the MOP
* A Clinical Quality Management Plan that describes the processes and activities that will be used to monitor and facilitate quality protocol execution following study initiation
* A Data Management Plan that describes procedures for data entry and completion, data security, and data quality control and validation

**14.1** Other types of QA/QC (detailed in the MOP) include:

* All study personnel training on GCP and study protocol/procedures
* Quality checks of consent documents to ensure completion and documentation of the consent process will be checked by the study coordinator after each enrollment visit
* Completion of data collection for each study visit, quality checks to ensure data entered into REDCap is complete and accurate
* Any QC related to biospecimens, such as logs to ensure biospecimens have been received/processed
* Programmed checks implemented in REDCap to ensure biospecimen data will be entered within a specified period of time after collection

# ETHICS/PROTECTION OF HUMAN SUBJECTS

## Ethical Standard

The investigator will ensure that this study is conducted in full conformity with the principles set forth in The Belmont Report: Ethical Principles and Guidelines for the Protection of Human Subjects of Research, as drafted by the US National Commission for the Protection of Human Subjects of Biomedical and Behavioral Research (April 18, 1979) and codified in 45 CFR Part 46 and/or the ICH E6.

## Institutional Review Board

The protocol, informed consent form(s), recruitment materials, and all participant materials will be submitted to the IRB for review and approval. Approval of both the protocol and the consent form(s) must be obtained before any participant is enrolled. Any amendment to the protocol will require review and approval by the IRB before the changes are implemented in the study.

## Informed Consent Process

Informed consent is a process that is initiated prior to the individual agreeing to participate in the study and continues throughout study participation. A narrated powerpoint presentation video will be presented to the participants, offering an extensive discussion of risks and possible benefits of study participation will be provided to participants. In addition, a consent form describing in detail the study procedures and risks will be given to the participant ahead of time before the first visit so they have sufficient time to review it. An IRB-approved electronic consent (e-consent) form in REDCap will also be provided and the participant is required to read and review the document or have the document read to him or her. The investigator or designee will explain the research study to the participant and answer any questions that may arise. The participant will sign the e-consent form in REDCap prior to any study-related assessments or procedures. Participants will be given the opportunity to discuss the study with their surrogates or think about it prior to agreeing to participate. They may withdraw consent at any time throughout the course of the study. A copy of the signed informed consent document will be given to participants for their records. The rights and welfare of the participants will be protected by emphasizing to them that the quality of their clinical care will not be adversely affected if they decline to participate in this study.

The consent process will be documented in the clinical or research record.

## Exclusion of Women, Minorities, and Children (Special Populations)

Children below the age of 18 will be excluded. While TMD can affect children, the manifestations of the condition can be quite different in children than in adults. Therefore, it is not advisable to study children in this trial, particularly given the use of an experimental intervention. This will be the only age group excluded.

Based on our use of both clinic- and community-based recruitment strategies, we anticipate enrolling a diverse cohort of individuals with TMD. Due to sex differences in the prevalence and severity of TMD, we expect to recruit substantially more women than men. Given the demographics of the local community, we expect to recruit a sample with significant racial and ethnic diversity.

## Subject Confidentiality

Subject confidentiality is strictly held in trust by the investigators, study staff, and the study sponsor(s) and their agents. This confidentiality is extended to cover testing of biological samples and genetic tests in addition to any study information relating to participants.

The study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study or the data will be released to any unauthorized third party without prior written approval of the study sponsor.

The study monitor or other authorized representatives of NIDCR may inspect all study documents and records required to be maintained by the investigator, including but not limited to, medical records (office, clinic, or hospital) for the study participants. The clinical study site will permit access to such records.

Certificate of Confidentiality

To further protect the privacy of study participants, the Secretary, Health and Human Services (HHS), has issued a Certificate of Confidentiality (CoC) to all researchers engaged in biomedical, behavioral, clinical, or other human subjects research funded wholly or in part by the federal government. Recipients of NIH funding for human subjects research are required to protect identifiable research information from forced disclosure per the terms of the NIH Policy (<https://humansubjects.nih.gov/coc/index>). As set forth in [45 CFR Part 75.303(a)](https://www.ecfr.gov/cgi-bin/text-idx?SID=f3e9328bbbd5aabe8e639ca48dcbcc7f&mc=true&node=se45.1.75_1303&rgn=div8) and [NIHGPS Chapter 8.3](https://grants.nih.gov/grants/policy/nihgps/HTML5/section_8/8.3_management_systems_and_procedures.htm), recipients conducting NIH-supported research covered by this Policy are required to establish and maintain effective internal controls (e.g., policies and procedures) that provide reasonable assurance that the award is managed in compliance with Federal statutes, regulations, and the terms and conditions of award. It is the NIH policy that investigators and others who have access to research records will not disclose identifying information except when the participant consents or in certain instances when federal, state, or local law or regulation requires disclosure. NIH expects investigators to inform research participants of the protections and the limits to protections provided by a Certificate issued by this Policy.

NIH Data Sharing Policies

As described in section 17, it is NIH policy that the results and accomplishments of the activities that it funds should be made available to the public (see <https://grants.nih.gov/policy/sharing.htm>). PIs and funding recipient institutions will ensure that all mechanisms used to share data include proper plans and safeguards to protect the rights and privacy of individuals who participate in NIH-sponsored research.

# DATA HANDLING AND RECORD KEEPING

The study MPIs are responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported. Most source documents will be completed electronically, and all source documents will be completed in a manner to ensure accurate interpretation of data. The investigators will maintain adequate case histories of study participants, including accurate eCRFs, and source documentation. Study staff will be trained to review all collected data and to identify and reconcile missing data.

## Data Management Responsibilities

Data collection and accurate documentation are the responsibility of the study staff under the supervision of the MPIs. During the study, the PIs will maintain complete and accurate documentation for the study. All source documents and laboratory data will be reviewed by the study team, who will ensure that all recorded information is accurate and complete. Safety events must be reviewed by the MPIs or designee. The biostatistician will be responsible for data management, data quality review, and data analyses.

## Data Capture Methods

Study staff will complete case report forms online via web-based REDCap, an electronic data capture system that has been validated and includes multiple security features. The data will be stored remotely at a central database managed by University of Florida REDCap team. Data quality will be ensured through the data collection system’s continuous monitoring of data and real-time detection and correction of errors. All elements of data entry (i.e., time, date, verbatim text, and the name of the person performing the data entry) will be recorded in an electronic audit trail to allow all changes in the database to be monitored and maintained in accordance with federal regulations.

## Types of Data

Study data include clinical examination findings and clinical pressure pain results, which will be obtained by trained study staff and recorded directly into REDCap. Additional data will include demographic and medical history data and questionnaire data obtained through study participant self-report and entered into REDCap by the participant. All data will be recorded electronically directly in the RedCap system. A trained phlebotomist will collect the blood draw samples, which will be processed. Biospecimens will be analyzed in batches under the supervision of Co-Investigator Gibson. The results of biospecimen analyses will be provided in electronic form for statistical analysis.

## Schedule and Content of Reports

The study team will produce study progress reports (i.e., screening, enrollment, missing data and study progress reports) as needed for monitoring purposes, as determined by the NIDCR program official and the MPIs. Study enrollment/retention reports will be provided to NIDCR monthly. The REDCap system will generate quality management reports to monitor measures of data quality, such as the number of outstanding queries and data completion rates. Study reports will be provided to the Medical Monitor and IRBs prior to their periodic meetings. No interim statistical analyses are planned.

Prior to database lock, the REDCap team will continuously perform the automated edit checks (Data Quality Rules) implemented in REDCap and will perform manual checks for all the study data until all queries are resolved and the data are determined to be clean. This process will include the detection of missing data, incorrect data type, data out of range, and invalid values in multiple choice fields or in calculated fields.

## Study Records Retention

Study records will be maintained for at least three years from the date that the last grant federal financial report (FFR) is submitted to the NIH.

## Protocol Deviations

A protocol deviation is any noncompliance with the clinical study protocol or Good Clinical Practice requirements. The noncompliance may be on the part of the participant, the investigators, or study staff. Protocol deviations will be assessed for their impact on safety, study operations, and data integrity. Appropriate corrective and preventive actions will be implemented, if warranted.

Consistent with the investigator obligations in the ICH E6 Guideline for GCP, the study staff will document in study source documents and explain any deviation from the IRB-approved protocol. The PIs will report to the IRB any deviations or changes made to eliminate immediate hazards to participants and any changes that increase risk to participants and/or significantly affect the conduct of the study.

Protocol deviations will be reported to the UF IRB per its requirements for annual review.

# PUBLICATION/DATA SHARING

This study will comply with all applicable NIH Data Sharing Policies. See <https://grants.nih.gov/policy/sharing.htm> for policies and resources.

NIH Public Access Policy

The NIH [*Public Access Policy*](https://publicaccess.nih.gov/index.htm) requires scientists to submit final peer-reviewed journal manuscripts that arise from NIH funds to [*PubMed Central*](https://www.ncbi.nlm.nih.gov/pmc/) immediately upon acceptance for publication. This ensures that the public has access to the published results of NIH funded research.

NIH Policy on the Dissemination of NIH-Funded Clinical Trial Information

The study is a clinical trial and will comply with the NIH policy that establishes the expectation that all investigators conducting clinical trials funded in whole or in part by the NIH will ensure that these trials are registered at ClinicalTrials.gov, and that results of these trials are submitted to ClinicalTrials.gov.

Food and Drug Administration Amendments Act of 2007 (FDAAA) and the Final Rule for Clinical Trials Registration and Results Information Submission

This study is an applicable clinical trial and will comply with [U.S. Public Law 110-85](http://frwebgate.access.gpo.gov/cgi-bin/getdoc.cgi?dbname=110_cong_public_laws&docid=f:publ085.110.pdf) (Food and Drug Administration Amendments Act of 2007 or FDAAA), Title VIII, Section 801 and [42 CFR Part 11](https://www.federalregister.gov/documents/2016/09/21/2016-22129/clinical-trials-registration-and-results-information-submission) (HHS Final Rule for Clinical Trials Registration and Results Information Submission), which mandate that a "responsible party" (i.e., the sponsor or designated principal investigator) register and report results of "applicable clinical trials."

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APPENDICES

APPENDIX A: SCHEDULE OF EVENTS

| Procedures | Pre-Screening (Day –49 to -7) | Baseline (Day -21 to -7) – Visit 0 | Randomization Visit 1 (Day 0) | Visits 2 (V1+ 4 days ± 3 days), 3 (V2 + 4 days ± 3 days), 4 (V3 + 4 days ± 3 Days), | Mid- intervention Visit 5 (Visit 4 + 4 days ± 3 Days) | Visits 6 (V5+ 4 days ± 3 Days),7 (V6+ 4 days ± 3 Days), 8 (V7+ 4 days ± 3 days), | Visit 9 – Post Intervention Assessment Visit *(*V8+ 5 days ± 9 Days ) | 1- and 3-Month Post-Intervention Follow-Up Assessments | Final Study Visit - Visit 10 (V9+ 180 days ± 30 Days) | Premature Discontinuation |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Verbal consent to pre-screen | X |  |  |  |  |  |  |  |  |  |
| Obtain written consent for study participation |  | X |  |  |  |  |  |  |  |  |
| Collect demographic information |  | X |  |  |  |  |  |  |  |  |
| Obtain and review health history |  | X | X | X | X | X | X |  |  | X |
| Record concomitant medications and therapies |  | X | X | X | X | X | X | X | X | X |
| Perform a urine pregnancy test in female participants of childbearing potential |  | X | X | X | X | X |  |  |  |  |
| TMD Jaw Functional Limitation Scale |  | X |  |  | X |  | X |  | X | X |
| Perform DC/TMD examination |  | X |  |  | X |  | X |  | X | X |
| Review eligibility/continuation criteria |  | X | X | X | X | X |  |  |  |  |
| Administer Psychological Questionnaires (PHQ15, Perceived Stress Scale, Pain Catastrophizing Scale, PROMIS Anxiety/Depression) |  | X |  |  |  |  |  |  |  |  |
| Administer PEG |  | X |  |  | X |  | X | X | X | X |
| Administer the Pressure Pain Threshold test (PPT) |  | X |  |  | X |  | X |  | X | X |
| Collect physical measurements and Skin Tone Measurement |  | X |  |  |  |  |  |  |  |  |
| Collect vital signs (blood pressure) |  | X |  |  |  |  |  |  |  |  |
| Dispense new electronic or paper Daily Symptom Diaries (DSD) |  | X | X | X | X | X | X | X | X | X |
| Collect DSD if in a paper form and review DSD |  |  | X | X | X | X | X | X | X | X |
| Blood draw |  |  | X |  | X |  | X |  | X | X |
| Intervention Expectations Questionnaire |  |  | X |  |  |  |  |  |  |  |
| Pre- and Post-Intervention Questionnaire |  |  | X | X | X | X |  |  |  |  |
| Intervention Experiences Questionnaire |  |  |  |  |  |  | X |  |  | X |
| Assess and record AE |  |  | X | X | X | X | X | X | X | X |
| PBM Intervention |  |  | X | X | X | X |  |  |  |  |