PROTOCOL TITLE:

Tracking Brain Biomarkers and Renormalization Associated with Antidepressant TMS Therapy

PRINCIPAL INVESTIGATOR:

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1.0 Objectives / Specific Aims

Study Aim: Quantify the phenotypic-related brain states of MDD patients compared to healthy individuals using a novel INSCAPE neuroimaging method and identify dynamic biomarkers that can track brain recovery of MDD patients over an antidepressant course of TMS therapy.

In this study, we plan to apply our novel "Individualized Network-based Single-frame Coactivation Pattern Estimation" (INSCAPE) method in 20 patients with Major Depressive Disorders (MDD) and 20 healthy controls to identify MDD-related brain states. We will then track changes in MDD-related brain states while MDD patients are undergoing 6 weeks of transcranial magnetic stimulation (TMS) therapy. Three separate neuroimaging visits will be conducted at the MUSC 30 Bee Street imaging center before, during, and after 30 sessions of the standard-of-care TMS treatment from the MUSC Brain Stimulation Service (~6 weeks in total). Additionally, healthy participants will undergo 3 times of MRI scanning with a 3-week interval between each two MRI sessions.

Hypothesis: *INSCAPE method can identify MDD specific brain states and track brain circuits recovery during TMS therapy.* We intend to establish INSCAPE as an effective neuroimaging technique to track antidepressant response in MDD patients undergoing 6 weeks of TMS therapy. We hypothesize that a) our INSCAPE method will be able to quantify phenotypic differences between MDD patients before TMS therapy and compared to healthy individuals. We further hypothesize that we can b) track the renormalization of brain state during the course of TMS treatment, as patients transition from "depressed brain states" to more "healthy" brain states. Lastly, we c) identify whether baseline brain state dynamic measures can predict response to TMS based on behavioral measures of response to TMS treatment.

2.0 Background

Major depressive disorder (MDD) is a prevalent psychiatric disorder characterized by impairments in affect, behavior, and cognition. Individuals with MDD persistently suffer from feelings of sadness and worthlessness and experience less enjoyment in social interactions (1-3). Over 21 million adults have had at least one major depressive episode, which leads to job absenteeism and reduces productivity, and has been one of the highest causes of disability worldwide that creates a tremendous economic burden in society (3-5). Aside from conventional antidepressant medications, transcranial magnetic stimulation (TMS) has emerged as an effective second-line intervention for medication-resistant depression (6, 7). Although TMS delivered to the left dorsolateral prefrontal cortex (dlPFC) is used clinically for the treatment of depression, it remains limited by large heterogeneity in clinical response between patients (8). One factor contributing to this wide range of individual response rates is a limited understanding of the changes induced in neural circuits that drive TMS response. Thus, investigating the functional circuits underlying depression at the individual level can help lead to more effective treatments for depression.

Resting-state functional magnetic resonance imaging (rs-fMRI) is a promising technique for assessing functional connectivity (FC) between brain regions that exhibit spontaneous synchronous fluctuations (9). Our previous rs-fMRI studies have revealed depressive-related functional connectivity (FC) changes in the default mode network (DMN) track with the severity of depression symptoms, including the anterior cingulate cortex, angular gyrus, and inferior parietal lobe (3, 10). Additionally, recent rs-fMRI advances have enabled the delineation of the spatial configuration of networks at the individual level (11). This individual-level analysis, rather than group-based averaging, has led to the discovery of new subnetworks in individuals that were missed in the group-average maps (12) and achieved higher accuracy in predicting phenotypes and disease symptoms based on functional measures (13). Therefore, exploring subnetworks of DMN at the individual level may help discover new biomarkers for depression treatment.

An advancement in rs-fMRI is a novel dynamic analytic approach which examines absolute coactivation patterns of BOLD signal across the brain at each time point (14). Using this method, we have already found brain states of psychiatric phenotypes in schizophrenia and psychotic experience patients, which can track with psychotic symptom severity (15). Based on this, our group has further developed a new method of detecting robust, dynamic brain states at the individual level, called INSCAPE – Individualized Network-based Single-frame Coactivation Pattern Estimation. INSCAPE can detect phenotypic differences amongst different individuals based on brain state (16). Our

findings indicated that the occurrence rate of brain states related to brain laterality could predict language lateralization derived from a semantic decision task and distinguish genders and handedness. Additionally, we also found a brain state that can track longitudinal brain recovery from stroke over six months – the occurrence rate of this brain state continuously reduced to the normal level during inter-hemispheric communication restoration. Thus, INSCAPE is a novel and advanced method that can allow us to begin understanding functional brain state changes that occur over the course of antidepressant treatment track recovery.

Ultimately, this study begins to optimize antidepressant treatments by identifying individual differences in brain states amongst patients. These individual differences likely play a large role in response rate, as well as network restoration in MDD patients. Thus, if successful, neuroimaging can become an additional tool that can be used before undergoing TMS treatment to better predict and improve treatment outcomes.

3.0 Intervention to be studied (if applicable)

This study is a neuroimaging study, thus, there is no intervention need to be managed for this study. Although, participants will be patients receiving the standard-of-care TMS therapy as part of their normal treatment plan in the MUSC Brain Stimulation Service, this intervention is not part of the study procedures.

4.0 Study Endpoints (if applicable)

Primary Outcome: *Dynamic brain states changes relative to MDD.*

The main outcomes of this study are neuroimaging related dynamic parameters derived from our INSCAPE approach, including occurrence rate, dwell time, transition matrix, etc. These outcomes will be compared between depression patients and healthy individuals. Moreover, we will also estimate the renormalization of brain states by quantifying the continuous changes of functional imaging measures as patients improve over the course of TMS treatment, moving towards the level of healthy controls. Lastly, the psychiatric-related imaging measures at baseline will then be verified whether they can predict symptom improvement and response to TMS therapy.

5.0 Inclusion and Exclusion Criteria/ Study Population

Twenty patients with MDD receiving standard-of-care TMS treatment at the MUSC Brain Stimulation Service will be recruited to participate in this prospective neuroimaging study. No preference will be given based on race, gender, or ethnicity. Pregnant females and children under the age of 18 will be excluded for safety reasons. No vulnerable populations or special classes of subjects will be considered for participation.

<u>Major Depression Disorder (MDD) Patients</u>

Inclusion Criteria

- Age 18-65
- Have the capacity and ability to provide one's own consent in English and sign the informed consent document.
- DSM-IV diagnosis of MDD

Exclusion Criteria

- Unable to speak English.
- Contraindicated for MRI.

- Any current or recent untreated medical, neurological, or psychiatric conditions other than MDD that would preclude candidacy for TMS.
- Metal implant devices in the head, heart, or neck.
- History of brain surgery.
- History of cortisol medication use or electroconvulsive therapy.
- History of myocardial infarction or arrhythmia, bradycardia.
- Personal history of recent head injury, concussion, or self-report of moderate to severe traumatic brain injury.
- Individuals suffering from frequent/severe headaches.
- Moderate to severe alcohol or substance use disorder.
- Pregnancy

Healthy Individuals:

Additionally, 20 demographic-matched healthy individuals will also be recruited as a control group.

Inclusion Criteria

- Age 18-65
- Have the capacity and ability to provide one's own consent in English and sign the informed consent document.

Exclusion Criteria

- Unable to speak English.
- Contraindicated for MRI.
- Any current or recent untreated medical, neurological, or psychiatric conditions
- Metal implant devices in the head, heart, or neck.
- History of brain surgery.
- History of cortisol medication use or electroconvulsive therapy.
- Comorbidity with other psychiatric/neurological illnesses or personality disorders
- History of myocardial infarction or arrhythmia, bradycardia.
- Personal history of recent head injury, concussion, or self-report of moderate to severe traumatic brain injury.

- Individuals suffering from frequent/severe headaches.
- Moderate to severe alcohol or substance use disorder.
- Pregnancy

6.0 Number of Subjects

We will enroll 40 total participants into this study (n=20 patients with depression and n=20 healthy adults) for this study.

7.0 Setting

Volunteers with Major Depressive Disorder who are candidates for TMS will be consented and educated about the study in a private screening area at either the MUSC Brain Stimulation Service or the MUSC 30 Bee Center for Biomedical Imaging, whatever is most convenient for the participant. Healthy volunteers will be consented at MUSC 30 Bee Street Center for Biomedical Imaging.

All neuroimaging conducted in this study, for both healthy volunteers and depressed volunteers, will be completed at the MUSC 30 Bee Street Center for Biomedical Imaging.

Depressed volunteers will complete the additional standardized mood assessments at the MUSC Brain Stimulation Service during their scheduled standard-of-care TMS treatment visits at the MUSC Brain Stimulation Service. Depressed volunteers will receive standard-of-care TMS treatment at the MUSC Brain Stimulation Service.

8.0 Recruitment Methods

The Brain Stimulation Service will be recruiting depressed volunteers who are seen in consultation for clinical services. Patients who are found to be appropriate candidates for TMS for MDD will be offered to be screened for enrollment into the study protocol by clinical care team members prior to being contacted by members of the study team. Healthy participants will be recruited from the MUSC community (students and staff) as well as general population from the greater Charleston region. We will recruit via flyers around campus and word of mouth.

9.0 Consent Process

Consent procedures will be conducted in a private, quiet room at the MUSC 30 Bee Street Center for Biomedical Imaging or within the MUSC Brain Stimulation Service. Approved study personnel will walk through the consent procedures with the participant. Furthermore, after the consenting procedures, the consent form will be printed and provided to all participants to independently review to ensure understanding, including describing the laboratory measures, study duration, and equipment and materials. The study team will describe confidentiality/privacy measures, participant right to withdraw, risks/benefits, and that $50 compensation will be paid using Clincards. In addition, participants will be prompted to ask questions throughout consenting to further ensure understanding. After signing the consent form, they will also be offered a hard copy.

10.0 Study Design / Methods

1. *Study Overview:* After determining eligibility and interest, written informed consent will be obtained from participants at the MUSC Center for Biomedical Imaging. Enrolled MDD patients (n=20) will be involved in neuroimaging before, during, and after 30 sessions of the standard-of-care TMS treatment MUSC brain stimulation service (~6 weeks in total). For each neuroimaging visit, participants will be placed in the MRI scanner and undergo four runs of resting-state fMRI (6 min/run). During the scanning, participants will be

asked to stay still in the scanner. Additionally, enrolled healthy control participants (n=20) will undergo 3 sessions of MRI scanning over a 6-week interval (Baseline, week 3, week 6). Upon completion of the 30 minutes neuroimaging visit, participants will be compensated $50. A timeline and group assignment overview are demonstrated in **Figure 1**.

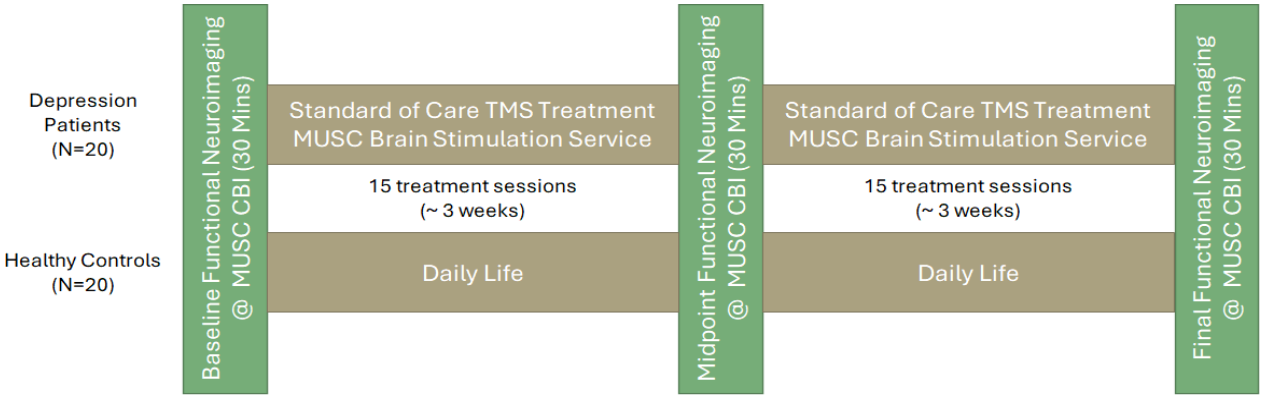

Figure 1. Study Overview.

2. *Recruitment, Screening, and Consent Procedures:* MDD patients will be recruited from the Brain Stimulation Service at MUSC by the clinical care team. Patients who are found to be appropriate candidates for TMS for MDD will be offered to be screened for enrollment into the study protocol.

   Healthy participants will be recruited from the MUSC community (students and staff) as well as general population from the greater Charleston region. We will recruit via flyers around campus and word of mouth. If interested in learning more about the study, potential subjects will be phone screened and educated regarding the study's details. If interested in participating in the study, they will be consented and enrolled into the study's protocol. After consent, the participant will complete a demographics and intake form.

3. *Pregnancy Test Procedures:* If participants are individuals of childbearing potential, they will be provided with a pregnancy test strip. After completing the pregnancy test, participants with negative results will be asked to continue to the remaining study procedures, whereas those with a positive result will be debriefed and released.

4. *Neuroimaging Procedures:* All MRI imaging will be performed using a Siemens 3.0 T Prisma scanner equipped with a 32-channel head coil. All MRI scan visits will be held at the MUSC Center for Biomedical Imaging (CBI; see the Facilities and Resources section). Each MRI scan session will include a high-resolution T1-weighted structural scan (whole brain sagittal acquisition, 224 slices, TR=2530ms, TE=3.65ms, TI=1100ms, 1mm thick slices, FOV=256, 256x176, flip angle = 7 degrees.), followed by four runs of resting-state fMRI scans using a gradient-echo echo-planar pulse sequence (TR = 2000 ms, TE = 36 ms, FA = 80°, 2.2 × 2.2 × 2.2-mm voxels).

5. *Depression/Anxiety Assessments:* MDD participants will complete several standardized assessments performed prior to starting TMS treatment, and repeated on weekly intervals. The assessments to be performed are the Beck Depression Inventory II (BDI-II), Hamilton Anxiety Rating Scale (HAM-A), Hamilton Depression Rating Scale (HAM-D), and Patient Health Questionnaire 9 (PHQ-9). These assessments will be used to describe more accurately clinical changes in depression over the course of TMS, and will be used alongside the collected MRI data to potentially identify changes to the brain associated with clinical improvement or lack thereof.

12.0 Data Management

All data will be stored in the Redcap database as well as in paper documentation. Information about the participant (including their identifiable private information) may have all their identifiers removed and used for future research

studies or distributed to other researchers for future research without additional informed consent. After participation, RedCAP data will be downloaded in excel format to the secure MUSC server. Paper documentation will be stored in locked cabinet within a locked office of the study team. In terms of publication, data will be published in aggregate form, so individual participants will not be identifiable in the final manuscript. No identifying information will be published.

Most of the data collected in this study are imaging data. These data will be collected from the 3T MRI scanner at 30 Bee Street. All imaging data will be automatically transferred to and stored on a password-protected, encrypted secure server that limits data access to personnel directly involved with the study. The data will be analyzed using standard imaging analysis software packages (such as FSL and SPM) and lab developed programs (such as INSCAP and Individual Parcellation).

If desired, participants may request a deidentified CD of their imaging data the day the scan occurs, so they may have it for their records, free of cost. Additionally, if any incidental findings occur during the scan, research staff and PI will follow the Center for Biomedical Imaging incidental finding procedures and have the scan read by a CBI affiliated neurologist or neuroradiologist for report.

Confidentiality and Quality Control: All study personnel will complete Social-Behavioral-Educational research CITI training, and also complete in-lab training regarding data security practices. Study personnel will be trained in the IRB protocol. The investigator, and co-investigators will be available to monitor data collection to ensure quality, confidentiality, and adherence to the IRB protocol.

The study's procedures will take place at the MUSC 30 Bee Street Neuroimaging Center. Regarding documentation, participant names will appear only on the IRB-approved Consent and HIPAA.

13.0 Provisions to Monitor the Data to Ensure the Safety of Subjects

There are three areas in which safeguards to protect subjects from undue risk require discussion. These Include: 1) procedures used to obtain informed consent, 2) procedures used to ensure confidentiality of the subject data, and 3) procedures used to minimize possible risks associated with the laboratory procedures. Regarding informed consent, participants are fully advised on the research procedures to be used, the amount of time required of them, the possible risks and benefits of the procedures, their right to refuse participation in the study without prejudice, their right to terminate participation at any moment without prejudice, and the name and telephone number of the principal investigator. All subjects will be required to have capacity to consent. Regarding confidentiality, subjects are informed that the information they provide will be kept strictly confidential, with access limited to the research staff. Participation in the study will be treated as confidential, as will all records. The identity of subjects will be protected with alphanumeric codes. All data will be kept in locked file cabinets file cabinets or on secure servers designed for use and access by study team only.

The PI and Co-Is of this study will meet every two months, for the duration of this project and review all the data related to this project.

We do not anticipate any adverse events to occur in this study, however the experienced research team has a long-standing record of recording and reporting unanticipated adverse events to the IRB. We will report any adverse event within 48 hours to the IRB.

14.0 Withdrawal of Subjects

Participants will be informed during consenting that they are free to withdraw from the study at any time. They will be informed that they are not obligated to participate once the study is initiated and in particular will be reminded prior to neuroimaging that they may discontinue the experiment at any point. Resting-state fMRI has been a very safe procedure and we do not anticipate subject withdrawal due to fMRI side effects.

15.0 Risks to Subjects

Magnetic Resonance Imaging (MRI):
MRI tests are non-invasive and painless. There are no known risks or side effects associated with conventional MRI procedures except to those people who have electrically, magnetically, or mechanically activated implants (such as cardiac pacemakers) or to those who have clips on blood vessels in their brain. However, an MRI may cause participants to feel claustrophobic (uncomfortable in a small space) or anxious from the noises made by the machine.

Unknown Risks:
There is always the possibility of other risks for a relatively new image processing technique. The Study team will let the participant know if they learn anything that might make the participant change their mind about participating in the study.

Loss of Confidentiality:
There is a risk of a loss of confidentiality of personal information. Subjects are informed that the information they provide, as well as participation in the study, will be kept strictly confidential, with access limited to the research staff. The identity of subjects in databases will be protected with alphanumeric codes. All data will be kept in locked file cabinets or on secure servers designed for use and access by Study Team members only.

16.0 Potential Benefits to Subjects or Others

There will be no direct benefit to the participant in the study. Data from this study, however, will benefit society by improving the understanding of the functional neurocircuitry underlying depression and whether there are MDD-related dynamic brain states that can track clinical antidepressant TMS treatment. MRI is an FDA-approved diagnostic tool.

17.0 Sharing of Results with Subjects

There is no plan to inform subjects of the results of the study, but they can always contact the research staff and ask. If there are significant new findings during the course of the study, they will be notified.

If desired, participants may request a deidentified CD of their imaging data the day the scan occurs, so they may have it for their records, free of cost.

18.0 Drugs or Devices

There is no plan to use any drugs or devices in this study.

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