MODULATION OF COGNITIVE CONTROL SIGNALS IN PREFRONTAL CORTEX BY RHYTHMIC TRANSCRANIAL MAGNETIC STIMULATION

STUDY CODE: PATRC

Funding Mechanism:	*N/A*
Principal Investigator:	*Flavio Frohlich, PhD*
Co-Investigator:	*N/A*
Protocol Number:	*18-1789*
Version Number:	*5.0*
Date:	*10 June 2019*

Initial version: 13 July 2018

TABLE OF CONTENTS

LIST OF ABBREVIATIONS

AE	Adverse Event/Adverse Experience
ANOVA	Analysis of Variance
aMFG	Anterior Middle Frontal Gyrus
BIS/BAS	Behavioral Inhibition System / Behavioral Approach System
CFR	Code of Federal Regulations
Co-I	Co-Investigator
CRF	Case Report Form
DC	Direct current
DHHS	Department of Health and Human Services
DMV	Department of Motor Vehicles
DSM-5	Diagnostic and Statistical Manual of Mental Disorders (Version 5)
DSMB	Data and Safety Monitoring Board
dPM	Dorsal Premotor cortex
eCRF	Electronic Case Report Form
EEG	Electroencephalogram
EMG	Electromyography
EOG	Electrooculography
FDA	Food and Drug Administration
GCP	Good Clinical Practice
Hz	Hertz
ICF	Informed Consent Form
ICH	International Conference on Harmonization
IDE	Investigational Device Exemption
IFJ	Inferior Frontal Junction
IRI	Interpersonal Reactivity Index
LAR	Legally Authorized Representative
M1	Primary motor cortex
NAMI	National Alliance on Mental Illness
NIH	National Institutes of Health
NIMH	National Institute of Mental Health
NRB	Neurosciences Research Building
OHRE	Office of Human Research Ethics
OHRP	Office for Human Research Protections
PHI	Protected Health Information
PI	Principal Investigator
SAE	Serious Adverse Event/Serious Adverse Experience
SMC	Safety Monitoring Committee
SPI	Serial Peripheral Interface
SOP	Standard Operating Procedure
TMS	Transcranial Magnetic Stimulation
UE	Unexpected Event
UNC	University of North Carolina
UNC-CH	University of North Carolina at Chapel Hill
US	United States

STUDY SUMMARY

Title	*Modulation of cognitive control signals in prefrontal cortex by rhythmic transcranial magnetic stimulation*
Short Title	*PATRC*
Protocol Number	*18-1789*
Phase	*Pilot*
Methodology	*Four-session, crossover design, active sham controlled*
Study Duration	*This study is expected to last eighteen months.*
Study Center(s)	*This is a four-session and single-site study performed at The University of North Carolina at Chapel Hill.*
Objectives (Purpose)	*The purpose of this pilot study is to modulate oscillatory activity in the prefrontal cortex using rhythmic transcranial magnetic stimulation during performance of a cognitive control task.*
Number of Participants	*100 (until 48 are completed)*
Diagnosis and Main Inclusion Criteria	*Eligible participants will be healthy adults between the ages of 18-35 with no history of mental or psychiatric disorder.*
Description of Intervention (Procedures/methods)	*The participants will perform a cognitive control task. During the task, rhythmic trains of transcranial magnetic stimulation will be delivered. Electroencephalography will be collected throughout every session.*
Related IRB Applications	*17-0149*

1 KEY ROLES

1.1 INDIVIDUALS

Principal Investigator Flavio Frohlich, PhD

Medical Monitor *N/A*

Flavio Frohlich, PhD – Principal Investigator
UNC – Chapel Hill Department of Psychiatry
Email: flavio_frohlich@med.unc.edu
Office: 4109F Neuroscience Research Building
Phone: (919) 966-4584

Justin Riddle, PhD – Study Coordinator
UNC – Chapel Hill Department of Psychiatry
Email: justin_riddle@med.unc.edu
Office: Medical School Wing C 245A
Phone: (919) 523-3127

Trevor McPherson, BS – Research Assistant
UNC – Chapel Hill
Email: trevor_mcpherson@med.unc.edu
Office: Medical School Wing C – Room 245A
Phone: (919) 445-0227

1.2 INSTITUTIONS

The University of North Carolina at Chapel Hill

1.3 OPTIONAL

IRB
The University of North Carolina – Chapel Hill
Medical School Building 52
Mason Farm Road
CB #7097
Chapel Hill, NC 27599-7097
(919) 966-3113

1.4 FUNDING SOURCES

Please list below the funding sources for this project:

Sponsor Name	UNC Ramses Number	Sponsor Type	Prime Sponsor Name	Prime Sponsor Type	Sponsor/Grant Number
N/A	N/A	N/A	N/A	N/A	N/A

External Funding: This project is not externally funded.

UNC-CH Funding: This project is not funded through UNC-CH.

Classified: This project is not classified.

2 INTRODUCTION

This document is a protocol for a human research study. This study is to be conducted according to U.S. and international standards of Good Clinical Practice (FDA Title 21 Part 312 and International Conference on Harmonization guidelines), applicable government regulations and Institutional research policies and procedures.

2.1 BACKGROUND

Neural oscillations are proposed to be a mechanism of coordinating information processing across distributed regions of cortex (Fries 2015). Different neural oscillations may correspond to different underlying neural computations (Wang 2010). Noninvasive brain stimulation allows experimenters to modulate specific neural oscillations by targeting particular frequency bands (Romei et al. 2016). By collecting simultaneous electroencephalography (EEG), rhythmic transcranial magnetic stimulation (TMS) has been previously demonstrated to entrain neural oscillations at the frequency of stimulation (Thut et al. 2011; Hanslmayr et al. 2014; Albouy et al. 2017). Furthermore, when the frequency of entrained neural oscillations is matched to the frequency of endogenous activity in a cognitive task, the brain stimulation improves behavioral performance (Albouy et al. 2017). Therefore, noninvasive brain stimulation is a promising tool for improving cognition by inducing optimal neural activity via externally applied electromagnetic fields; e.g. cognitive control improvements (Alekseichuk et al. 2016; Albouy et al. 2017; Reinhart 2017; Wolinski et al. 2018).

Previous evidence has implicated neural activity in the alpha band (8-12 Hz) in information suppression and activity in the theta band (4-7 Hz) in information processing (Klimesch et al. 2007; Dé et al., 2014). Cognitive control task paradigms have been shown to elicit distinct activity in both of these bands (Popov er al., 2018). In this task, the presented stimuli can be lateralized to the right and left visual field during encoding, while a cue informs participants which stimuli (right or left) are relevant to be remembered for testing later. In this paradigm, alpha activity in parietal cortex is found contralateral to irrelevant stimuli—supporting the role of alpha in information suppression—while theta activity in frontal cortex increases with the number of stimuli to be remembered—supporting the role of theta in active information processing (Wallis et al., 2015).

For the current study, we propose to deliver rhythmic trains of TMS in either alpha frequency, theta frequency, or an arrhythmic control to modulate neural processing during a cognitive control task. By collecting simultaneous EEG with TMS, we will be able to measure the entrained oscillations from rhythmic TMS. The goal of this experiment is to boost the observed theta and alpha activity that is seen with the successful prioritization and suppression of information during the task.

To provide causal evidence that parietal cortex generates alpha activity and frontal cortex generates theta activity, we will apply rhythmic TMS stimulation to two scalp location, the anterior middle frontal gyrus and inferior intraparietal sulcus. By applying alpha frequency, theta frequency, and arrhythmic TMS at each location, we will be able to examine the causal relationship of frontal theta oscillations to information prioritization and parietal alpha oscillations to information suppression.

2.2 DOSE RATIONALE

TMS is a safe, non-invasive, widely-used tool that applies focal electric fields to the brain using magnetic coils placed on the scalp (Rossi et al. 2009). On the first session of the study, participants will receive a motor thresholding procedure in which electrodes are attached to the first dorsal interosseous muscle of the right hand. The contralateral motor cortex will be targeted by single pulses of TMS until a motor evoked potential (MEP) is generated: defined as a near-instantaneous increase muscle activity greater than 200 microvolts. Next, the intensity of single pulse TMS will be lowered until an MEP is generated on five out of ten pulses. This is defined as the participant's motor threshold (Rossi et al. 2009). For the subsequent stimulation days, participants will receive TMS at 120% of their motor threshold. Participants will receive trains of 5 pulses at 5 hertz, 10 hertz, or arrhythmic randomized pulse intervals not to exceed an inter-pulse-interval of 20 milliseconds. Between every train will be at least 5 seconds. An inter-train-interval of 5 seconds or greater allows for any residual effects of stimulation to return to baseline. This intensity, inter-pulse-interval, and inter-train-interval is well within the safety guidelines set forth for repetitive TMS (Wassermann, 1998; Rossi et al., 2009) and has been used in similar paradigms as the one described here (Thut et al. 2011; Hanslmayr et al. 2014; Albouy et al. 2017). EEG will be collected using the Geodesic 400 system (EGI INC., Eugene, OR, USA). Collecting simultaneous EEG and TMS does not pose any additional risk over TMS on its own. EEG does not involve brain stimulation and is used purely for neuroimaging (minimal risk).

2.3 STUDY AIMS/HYPOTHESES

2.3.1 PRIMARY OUTCOME – WORKING MEMORY CAPACITY

RATIONALE. Participants make a button press on a keyboard to indicate if the probed items are matched or non-matched to the items held in memory. The investigators will calculate the accuracy for these responses and convert this to Pashler's working memory capacity metric for each condition. Theta oscillations are known to be generated in frontal cortex when cued to the contralateral visual field. Alpha oscillations are known to be generated in parietal cortex when cued to the ipsilateral visual field.
ALTERNATIVE HYPOTHESIS. When the frequency of TMS to either frontal or parietal cortex is compatible with the endogenous activity, working memory capacity should increase relative to TMS that is incompatible. Therefore, we will report the TMS compatibility effect as the average of "theta minus alpha TMS to frontal cortex for retro-cue right" and "alpha minus theta TMS to parietal cortex for retro-cue left". We will report the mean and standard deviation of the TMS compatibility effect across participants. This effect will be investigated using repeated-measures two-way ANOVA, and an interaction between TMS site and TMS frequency is hypothesized.
NULL HYPOTHESIS. There is no systematic effect of frequency specific TMS on working memory capacity.

2.3.2 PRIMARY OUTCOME – SPECTRAL POWER

RATIONALE. The electrical activity of the brain is recorded during performance of the task and during brain stimulation. The investigators will perform Morelet wavelet convolution on the recorded electrical signal to calculate the spectral power of theta (5-8 hertz) and alpha (8-12 hertz). Theta oscillations are known to be generated in frontal cortex when cued to the contralateral visual field. Alpha oscillations are known to be generated in parietal cortex when cued to the ipsilateral visual field.
ALTERNATIVE HYPOTHESIS. When the frequency of TMS to either frontal or parietal cortex is compatible with the endogenous activity, spectral power in the stimulated frequency band should increase relative to TMS that is incompatible. Therefore, we will report the TMS compatibility effect as the average of "theta power for theta minus alpha TMS to frontal cortex for retro-cue right" and "alpha power for alpha minus

theta TMS to parietal cortex for retro-cue left". We will report the mean and standard deviation of the TMS compatibility effect across participants. This effect will be investigated using repeated-measures two-way ANOVA, and an interaction between TMS site and TMS frequency is hypothesized.
NULL HYPOTHESIS. There is no systematic effect of frequency specific TMS on spectral power.

2.3.3 SECONDARY OUTCOME – RESPONSE TIME

RATIONALE. Participants make a button press on a keyboard to indicate if the probed items are matched or non-matched to the items held in memory. The investigators will calculate the response time for each condition. Theta oscillations are known to be generated in frontal cortex when cued to the contralateral visual field. Alpha oscillations are known to be generated in parietal cortex when cued to the ipsilateral visual field.
ALTERNATIVE HYPOTHESIS. When the frequency of TMS to either frontal or parietal cortex is compatible with the endogenous activity, response time should decrease relative to TMS that is incompatible. Therefore, we will report the TMS compatibility effect as the average of "theta minus alpha TMS to frontal cortex for retro-cue right" and "alpha minus theta TMS to parietal cortex for retro-cue left". We will report the mean and standard deviation of the TMS compatibility effect across participants. This effect will be investigated using repeated-measures two-way ANOVA, and an interaction between TMS site and TMS frequency is hypothesized.
NULL HYPOTHESIS. There is no systematic effect of frequency specific TMS on response time.

3 PARTICIPANT SELECTION AND WITHDRAWAL

A total of 48 participants will complete this study and all data will be collected at UNC-CH. No specific plans have been made to enroll participants from vulnerable populations.

3.1 INCLUSION CRITERIA

In order to be eligible to participate in this study, a participant must meet all of the following criteria:

- Between the ages of 18 and 35
- Right handed
- Able to provide informed consent
- Have normal to corrected vision
- Willing to comply with all study procedures and be available for the duration of the study
- Speak and understand English

3.2 EXCLUSION CRITERIA

A potential participant who meets any of the following criteria will be excluded from participation in the study:

- ADHD (currently under treatment)
- Neurological disorders and conditions, including, but not limited to:
    - History of epilepsy
    - Seizures (except childhood febrile seizures)
    - Dementia
    - History of stroke
    - Parkinson's disease
    - Multiple sclerosis
    - Cerebral aneurysm
    - Brain tumors
- Medical or neurological illness or treatment for a medical disorder that could interfere with study participation (e.g., unstable cardiac disease, HIV/AIDS, malignancy, liver or renal impairment)
- Prior brain surgery
- Any brain devices/implants, including cochlear implants and aneurysm clips
- History of current traumatic brain injury
- (For females) Pregnancy or breast feeding
- Anything that, in the opinion of the investigator, would place the participant at increased risk or preclude the participant's full compliance with or completion of the study

Justifications for any exclusions based on race, gender, or ethnicity: There are no exclusion criteria based on race, gender, or ethnicity. However, non-English speaking individuals are excluded because the ability to accurately and completely communicate study information, answer questions about the study, and obtain consent are necessary.
Justification for excluding women or women who become pregnant: Pregnant participants will be excluded despite the fact that theoretical risk to mother or fetus is exceedingly small, since no safety data for pregnancy is known to exist for TMS studies. Female participants will be asked if there is a possibility that they are pregnant at every stimulation session. If the participant says yes or is unsure, then we will verify pregnancy status via a urine pregnancy test. Only upon a verbal confirmation that pregnancy is not possible or a negative finding will we proceed with the experiment.

3.3 STRATEGIES FOR RECRUITMENT AND RETENTION

3.3.1 RECRUITMENT

The target population of this study includes healthy individuals between the ages of 18 to 35 recruited from UNC Chapel Hill and the surrounding area, including approximately 30,000 candidates for participation. Recruitment will occur via two methods: flyers to be distributed at major locations on the campus (libraries, gyms, classroom buildings, etc.) and a mass email to all individuals with a UNC-affiliated email address.

3.3.2 RETENTION

Our retention strategy includes a payment schedule of one time per session per participant. The payment amount increases with each session to encourage study completion. The participant will receive payment at the end of each session. The research staff will also give each participant a reminder call or email for upcoming sessions. Each research staff member will be available for the participants to contact via email or phone.

4 BASIC STUDY DESIGN

This study is a pilot, four-session study with TMS and EEG to understand the neural oscillatory basis of cognitive control. Participants will perform a cognitive control task in which a retro-cue is used to distinguish between relevant and irrelevant lateralized stimuli that need to be accurately recalled for testing in each trial. The rhythmic TMS manipulation targets the previously localized EEG frequencies and previously localized regions of the brain from MEG (Wallis et al., 2015). The frequencies of stimulation will be theta (5 hertz), alpha (10 hertz), and arrhythmic. The regions of stimulation will be anterior-MFG, inferior-IPS. The study consists of an initial screening session, a baseline session, and two counterbalanced stimulation sessions with TMS delivered during task performance. During the initial screening session, participants will earn and perform the cognitive control task at varying levels of difficulty in order to titrate their performance accuracy. During the baseline session, participants will perform the titrated cognitive control task while EEG is recorded. The motor threshold (MT) for each participant will also be calculated either on this day or during the Stimulation 1 session. MT is calculated by placing the TMS coil at a 45-degree angle from the midline along the central sulcus (just above the ears). An electrode is attached to the contralateral first dorsal interosseous muscle. Then the primary motor cortex is stimulated with single pulse TMS until a motor evoked potential (MEP) is generated in the contralateral hand muscle, defined as a near-instantaneous increase in electrical activity above 200 microvolts. Then the intensity of TMS is lowered until an MEP is evoked on five out of ten pulses. Additionally, visible hand twitches may be used as instead of visual twitches to determine MT. The MT allows for TMS to be calibrated to the particular sensitivity level of the individual participant's brain. On the subsequent TMS session we will stimulate at 110% of MT. We ensure that TMS is driving neural activity but that this activity is within the range of endogenous activity and hence is at an appropriately safety level according to guidelines established for TMS (Rossi et al. 2009). On the third and fourth session (Stimulation 1 and Stimulation 2) the anterior-MFG and inferior-IPS is targeted by rhythmic TMS during performance of the cognitive control task. The sequence of TMS targets will be randomized and counterbalanced across participants. However, it is not possible to double-blind the study because the location of the TMS coil needs to be aligned to the targeted brain region with neuro-navigation software (Localite, Sankt Augustin, Germany). In other words, the region targeted by TMS is necessarily visible to the experimenter for accurate targeting. The study will be single blinded as the participant will not know the anatomical differences between the targeted regions and the position on the scalp differs by only a few centimeters between each site. Critically, the manipulation of interest is the frequency of stimulation. On each trial, the frequency will be randomly selected, counter-balanced, and inter-mixed. The effects of rhythmic TMS do not last for more than a few cycles beyond stimulation itself (Thut et al. 2011; Hanslmayr et al. 2014; Albouy et al. 2017). Therefore, the experimental design randomly intermixes the frequency of stimulation within every task block. The critical analysis is the within participant comparison of frequency specific TMS effects for a single site. The sequence order of target sites will have no impact because all analyses will be run as a difference from our active control (arrhythmic TMS) from within the same session. In addition, a brief resting-state EEG recording will be collected at the beginning of the

baseline and stimulation sessions.

4.1 TREATMENT ASSIGNMENT PROCEDURES

The three frequencies of interest (theta, alpha, and arrhythmic) are randomly intermixed within every task block. The randomization is uniquely generated for each participant and session. The order of regions targeted (a-MFG and i-IPS) will be randomized and counterbalanced across participants. The primary analysis of the experiment is to analyze the frequency specific effect of rhythmic TMS versus arrhythmic TMS within the same session. Therefore, the experiment is designed such that sequence order is irrelevant as every analysis is performed as a within session difference. Using neuro-navigation software, precludes a double-blinding as the experimenter must be aware of the targeted region. The participant will not be keen to the anatomical differences between these regions that are separated by only a few centimeters.

5 STUDY SCHEDULE

In order to increase data quality, the assessments for an individual participant will be administered by the same researcher. It is important to note that consent, scales, and experiments will all take place in a private room. Any phone calls will take place in a private lab environment as well.

5.1 SCREENING

The phone screening allows researchers to screen out participants based on self-report responses and for potential participants to become familiar with the study schedule, including procedures. During the telephone screening, researchers will provide a brief background about TMS. The timeline of visits will be explained, including the number of visits and the time commitment required. The participant will be informed of compensation, both amount and payment schedule. The participant will be asked if they have any questions. Once all questions have been answered, the participant will be asked if he/she is still interested in participating in the study. If yes, the researcher will ask if the participant will provide verbal consent to begin the initial phone screening which will determine eligibility for the stimulation session. *A telephone script, which includes the screening questions, is provided in Appendix G.* If the participant meets initial criteria with these two assessments, the stimulation session will be scheduled and a reminder call or email will be given at least 24 hours before the stimulation session.

5.2 STIMULATION SESSIONS

Participants will undergo four sessions: one screening, one baseline, and two active sessions. During all four sessions, participants will be guided through the consent form. To ensure that all aspects of the research are understood, participants may be asked a series of questions about the research they are about to take part in (*Appendix E*). Once it is clear that the participant understands the consent form they may sign the form.

Session 1: Screening

- In this initial session, participants will be explained the procedure and reminded of their expectations and rights (informed consent). After obtaining consent, a pregnancy screening will be administered. Following this the participant will practice and perform cognitive control task. Participants have a self-paced break between blocks of the task.
- Participant will receive $20 in compensation

Session 2: Baseline

- The following questionnaires will be administered during the session or will be completed remotely by the participant through an online survey weblink before the session - These measures are listed below under “self-report measures”
- Preparation - The EEG net will be applied. Resting state EEG – Participant will be asked to relax with eyes open and eyes-closed while EEG is collected simultaneously. Following this the participant will practice and perform cognitive control task. Motor thresholding using single pule TMS for use in future sessions (performed once for each participant, either on Baseline or Stimulation 1 day). Electrodes and net will be removed and participant can wash their hair and face Participant will receive $30 in compensation

Session 3 and 4: Stimulation 1 and 2

- The procedure for these sessions will follow the same timeline as the second, with the exclusion of informed consent, questionnaires Motor threshold may be performed on Stimulation 1 day if it was not done during the Baseline session.
- Before practice and performance of the cognitive control task, the TMS coil will be position over the targeted region. During performance of the task, stimulation will be delivered on every trial. At the end of the session, participants will receive $40, and $60 in compensation for sessions 3, and 4, respectively.

6 STUDY PROCEDURES/EVALUATIONS

6.1 SELF-REPORT MEASURES

Study coordinator will use a standard questionnaire to screen TMS candidates. The following questions include the standard contraindications for TMS research.

1. Do you have epilepsy or have you ever had a convulsion or a seizure?
2. Have you ever had a fainting spell or syncope? If yes, please describe in which occasion(s)
3. Have you ever had severe (i.e., followed by loss of consciousness) head trauma?
4. Do you have any hearing problems or ringing in your ears?
5. Are you pregnant or is there any chance that you might be?
6. Do you have metal in the brain/skull (except titanium)? (e.g. splint, fragments, clips, etc.)
7. Do you have cochlear implants?
8. Do you have an implanted neurostimulator? (e.g., DBS, epidural/subdural, VNS)
9. Do you have a cardiac pacemaker or intracardiac lines or metal in your body?
10. Do you have a medication infusion device?
11. Are you taking any medications? (Please list)
12. Did you ever have a surgical procedure to your spinal cord?
13. Do you have spinal or ventricular derivations?
14. Did you ever undergo TMS in the past?

Affirmative answers to one or more of questions 1–13 do not represent absolute contraindications to TMS, but the risk/benefit ratio should be carefully balanced by study coordinator.

In addition, several self-report measures will be collected throughout this study. These measures are listed below and can be found in the attached documents.

A. THE STATE-TRAIT ANXIETY INVENTORY (STAI) is a 20-item self-report assessment that assesses either temporary or chronic anxiety. For the purposes of this study, the state version will be used to measure anxiety as a result of the stress condition. The STAI is commonly used to assess both types of anxiety, and has applications in both clinical and research settings (Spielberger et al. 1983).
B. THE BEHAVIORAL INHIBITION AND BEHAVIORAL APPROACH SYSTEM SCALES (BIS/BAS) are a set of 24 questions used to assess an individual's sensitivity to approach vs. inhibition in motivating behavior. This scale is commonly used to measure behavior and has been demonstrated to be reliable (Carver and White 1994).

C. The Positive and Negative Affect Schedule (PANAS) is a 20-item self-report assessment using a 5-level Likert scale to assess the positive and negative affect of an individual over the past week. This scale has been shown to be a reliable measure of both positive and negative affect with little participantivity to demographic variables (Watson et al. 1988).
D. The Edinburgh Handedness Inventory is a 20-item scale to assess the hand dominance of a person in everyday activities (Oldfield 1971).
E. The Big Five Personality Questionnaire is a 50-item self-report assessment using a 5-level Likert scale to assess five dimensions of personality (John and Srivastava 1999).
F. Repetitive Thinking Questionnaire is a 31-item self-report assessment using a 5-level Likert scale to assess levels of rumination (McEvoy et al. 2010).
G. Montgomery-Asberg Depression Rating Scale Self-report is a 9-item self-report assessment using a 7-level Likert scale to assess level of depression (Montgomery and Asberg 1979)
H. Magical Ideation Scale is a 30-item self-report assessment using a 5-level Likert scale to assess traits of schizotypy (Eckblad and Chapman 1983).

6.2 SPECIAL ASSAYS OR PROCEDURES

Each participant will receive stimulation on all four sessions, separated by a week. The baseline session will involve single pulse TMS for calculating the motor threshold. The other three sessions will involve train of rhythmic TMS during performance of a cognitive control task. For stimulation day, the rhythmic TMS trains are delivered on every trial of the task (there are 600 trials per session). Each train of TMS consists of 5 pulses. There will be a total of 2400 pulses delivered per session. Each TMS train will be separated by a minimum of 5 seconds. There are three possible trains: alpha frequency with a 100 milliseconds inter-pulse interval, theta frequency with a 200 milliseconds inter-pulse interval, and arrhythmic with a variable inter-pulse interval (not to exceed a minimum of 20 milliseconds inter-pulse interval). The inter-pulse interval, inter-train interval, number of trains, and TMS intensity are all under the safety guidelines set forth by Rossi et al. 2009 and is typical for experiments that deliver online TMS (Hanslmayr et al. 2014; Albouy et al. 2017).

6.3 SAFETY MEASURES

We will be monitoring the safety of our participants throughout the study with the following measures. These measures are listed below and can be found in the attached documents.

A. A stimulation adverse effects questionnaire used in previous studies (IRB #14-1622, #14-3285, and #14-0600) will be administered at the end of each stimulation session. This questionnaire will be used as a safety measure and to collect data on participant experience. Please see *9.1 Safety Parameters* for more information.

6.4 LABORATORY EVALUATIONS

6.4.1 SCREENING LABORATORY EVALUATIONS

A urine pregnancy test will be performed for all female participants.

Instructions for specimen preparation, handling, and storage. For this evaluation, an FDA approved commercial home-based kit will be used (HCG Urine Pregnancy Test Strip). Results are available after only a few minutes. Once the results are clear, the researcher will make a note and the sample will be disposed. All samples will be handled using single-use disposable medical gloves.

6.5 COGNITIVE TASKS

6.5.1 COGNITIVE CONTROL TASK

The cognitive control task used in this paradigm consists of the presentation of colored squares on the left and right side of a computer screen. Stimuli are presented briefly, followed by a brief delay. After this, an arrow pointing to the left, right, or both left and right will briefly appear on the screen. This informs the participants which set of squares (left or right) they must remember and will be tested on. The cue is followed by another delay, and finally a new set of squares is presented. Participants press a button to indicate whether the squares are the same or different from the ones they originally saw. Between presentation of stimuli there is a small centrally presented fixation cross upon which participant maintain their gaze. Participants will complete 10 blocks of the task, each composed of 20-30 trials each. Participants will have a self-paced break between each of the blocks.

7 STUDY INVESTIGATIONAL PRODUCT

We will use the MagPro X100 system (MagVenture Inc., Alpharetta, Georgia, USA) for transcranial magnetic stimulation. The MagPro X100 is an advanced, high performance magnetic stimulator designed primarily for research purposes. It is a high-quality tool for researchers with a large choice of stimulating parameters and has stimulation rates up to 100 pulses per second at high intensities and the possibility to combine waveforms and pulse modes.

The simulator has several features:

- 3 waveforms: Biphasic, Biphasic Burst and Monophasic.
- Selectable current direction.
- Stimulation rates up to 100 pulses per second.
- Easily connects to external equipment via programmable input/output triggers.
- System operation control via a built-in computer, eliminating the need for an external computer to set up and control the timing of stimulus sequences.
- Controllable from an external device.

7.1 SAFETY FEATURES

In the USA, federal law regulates the sale of Medical Devices through the US Food and Drug Administration (FDA). This is done to ensure safety and effectiveness. Devices which are permitted to be marketed for their intended use must either have a 510(k) or PMA clearance.
MagPro® stimulators R20, R30, R30 with MagOption, X100, and X100 with MagOption are all FDA 510(k) cleared (k160280, k061645, k091940 and k150641).
k150641: The intended use is treatment of Major Depressive Disorder in adult patients who have failed to receive satisfactory improvement from prior antidepressant medication in the current episode.
k160280, k061645, k091940: The intended use is for stimulation of peripheral nerves for diagnostic purposes.
The use of devices for other than their FDA cleared intended use is considered as investigational. Such use is only permitted if the Investigational Device Exemption (IDE) guidelines have been followed. For full information on this procedure, please consult FDA's website (www.fda.gov).
All investigational devices must be labeled in accordance with the labeling provisions of the IDE regulation (§ 812.5) and must bear a label with this statement:
"CAUTION Investigational Device. Limited by Federal (or United States) law to investigational use."

7.2 PREPARATION AND ADMINISTRATION OF STUDY INVESTIGATIONAL PRODUCT

After participants have completed the questionnaire, they will be comfortably seated. The study coordinator and/or the research assistant will be thoroughly trained and have trainings documented on the transcranial magnetic stimulation device and will be present during all stimulation sessions. To monitor side effects of stimulation, a questionnaire will be administered after each stimulation.

7.3 ASSESSMENT OF PARTICIPANT COMPLIANCE WITH STUDY INVESTIGATIONAL PRODUCT

Compliance for this study is determined as completion of all four sessions.

8 POTENTIAL RISKS AND BENEFITS

8.1 BENEFITS TO PARTICIPANTS AND SOCIETY

This research has the potential to provide critical data for understanding the neural basis of cognitive control. The current study has no immediate benefit to the healthy participants that participate. However, the results of this study may be used to develop novel tools for the treatment of psychiatric mood-disorders that involve a detriment in cognitive control. Furthermore, simultaneous neuro-imaging with brain stimulation will provide increased understanding of the neural mechanisms that underlie the efficacy of brain stimulation.

8.2 POTENTIAL RISKS

8.2.1 PSYCHOLOGICAL

Risk of Embarrassment: Self-report questionnaire contains questions regarding personal information. This risk is necessary in order to assess symptomology and associated psychopathology. Participants will be assured upon intake that only study personnel will see any answers.

Risk of Confidentiality Breach. To avoid breaches in confidentiality, study documents that contain personal information, including the informed consent, and the document that links study ID numbers to personal identifying information are kept in locked filing cabinets in locked rooms, separate from any source documents containing participating dummy identifiers. All data is stored in locked cabinets inside locked offices; electronic data will be stored only on password-protected computers, and data encryption methods will be used during communication between investigators. Only study personnel will have access to these data. All study staff participate in annual human participating training that includes education about responsibilities to minimize the risk of confidentiality breach. In the unlikely event of a breach of confidentiality, people might discover that an individual was involved in this research study and some people might not agree with the principle of participating in research or of changing natural brain activity.

8.2.1 PHYSICAL

Risk of Injury and Discomfort: Transcranial magnetic stimulation has been cleared for use in the USA by the FDA. TMS (the methodology used in this experiment) has nothing to do with electroconvulsive therapy that applies many orders of magnitude higher stimulation current. The level of electrical stimulation produced by TMS is within the range of activity that is endogenous to the brain. Furthermore, the intensity of stimulation is calibrated to the sensitivity of the individual participant such that the level of stimulation is matched to that of naturally occurring activity. In order to monitor side-effects, we will be administering an adverse effects stimulation questionnaire after each stimulation session to determine whether these effects were experienced. Research personnel present during these sessions will also check with the participant periodically during the stimulation to see whether they are comfortable. If any side-effect occurs (rated by the participant as stronger than “moderate”) or the participant is experiencing severe discomfort, the stimulation will be immediately stopped.

8.3 REFERRALS FOR MEDICAL FOLLOW-UP OR PSYCHOLOGICAL COUNSELING

There is a purely theoretical likelihood that stimulation of neuronal circuits can lead to epileptic discharge. To minimize this occurrence, we screen and exclude participants with personal and family history of neurological conditions from the study. If abnormalities or a seizure is witnessed during the course of the study, a referral will be made to the UNC Department of Neurology for follow-up. In the theoretical event that a seizure is witnessed that involves the loss of consciousness, the participants will be told not to operate a motor vehicle until cleared by the DMV.

To ensure participant comfort, a study coordinator or research assistant will periodically check in with the participant about any side-effects he/she may be experiencing during each stimulation session. Following the conclusion of the stimulation session, the participant will receive an Adverse Effects Questionnaire to report on any of the side-effects he/she may have experienced. This questionnaire reports side-effects on a Likert scale (1=Absent, 2=Mild, 3=Moderate, 4=Severe). If the participant reports side-effects of Moderate to Severe intensity, a study coordinator or research assistant will discuss the side-effects experienced and note this response. The medical monitor will be contacted based on the reported intensity on the Adverse Events Questionnaire and the participant's verbal confirmation of intensity.

9.3.1 PREGNANCY FOLLOW-UP

Pregnancy tests will be administered on the first session of the experiment to all female participants. After testing negative, the experiment can continue. On subsequent sessions, female participants will be asked if it is possible that they may have become pregnant since the previous session. If the participant responds yes, or is unsure, then a pregnancy test will be administered. After testing negative at a given session (meeting inclusion criteria), if a participant reports becoming pregnant during the course of the study, she will be withdrawn from further participation. There are no plans to follow participants who become pregnant while enrolled in the study.

9 DATA AND SAFETY MONITORING

9.1 FROHLICH LAB MONITORING PLAN

The purpose of this monitoring plan is to present the Frohlich Lab's approach to monitoring clinical trials. The plan facilitates compliance with good clinical practice (GCP):

a. The rights and well-being of human participants are protected.
b. The reported trial data are accurate, complete, and verifiable from source documents
c. The conduct of the trial is in compliance with the currently approved protocol/amendment(s) with GCP, and with applicable regulatory requirement(s)

This section identifies key monitoring activities and specifies the data to be reviewed over the course of a clinical trial. This is a single site, investigator-initiated, clinical trial, so there will be no site monitoring plan in place.
The latest version of the approved IRB application for this clinical trial will be followed at all times. This responsibility falls into the hands of the study coordinator and research assistants. If at any time there is a deviation from protocol, the deviation from protocol log will be filled out. All team members will be trained on how and when to use this log. The most up-to-date IRB application will be on file in the Clinical Trials office in Room 233 of the Medical School Wing C. Deviations will be sent to the IRB every 4-6 weeks (if necessary).
Periodically, study staff should review 3 randomly selected informed consent forms to ensure that (1) these forms have been filled out appropriately, and (2) the consent form process was followed and properly documented. Should any consent form be in violation, the research team will perform and document a complete review of all consent forms.
AE and SAE are clearly defined in this document. Documents of AE and SAE can be found in the study binder on file in the Clinical Trials office in Room 233 of the Medical School Wing C. It is the responsibility of the study coordinator to report all events to the PI in a timely manner. All AEs and SAEs will be discussed with the PI. For our practices, we have adapted the decision tree provided by the UNC-CH IRB to assist with reporting of such events.

9.2 SAFETY OVERSIGHT

Safety oversight will be under the direction of the PI, who will review AEs in real time and make decisions regarding a participant's continuation of the clinical trial.

9.3 EARLY WITHDRAWAL OF PARTICIPANTS

9.3.1 REASONS FOR WITHDRAWAL

A study participant will be discontinued from further participation if:

- The participant meets any exclusion criteria (either newly developed or not previously recognized).
- Anything that, in the opinion of the investigator, would place the participant at increased risk or preclude the participant's full compliance with or completion of the study.

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

9.3.2 DATA COLLECTION FOR WITHDRAWN PARTICIPANTS

We will collect safety data on any participant discontinued because of an AE or SAE. In any case, every effort will be made to undertake protocol-specific follow-up procedures. If voluntary withdrawal occurs, the participant will be asked to continue scheduled evaluations and complete an end-of-study evaluation.

If an AE has been reported, researchers will help the participant seek the medical care they need and a follow-up will be performed by the PI In the case of an early withdrawal, the researcher will make a note to file indicating this.

9.4 TERMINATION OF STUDY

If a seizure occurs at the time of a study visit, a temporary hold will be placed over the study and further investigation will ensue. This could lead to stopping the study prematurely or continuing on with further safety measures in place. If two seizures are witnessed during the study visits, the entire study will be stopped prematurely. These individuals would be referred for further medical attention. It is very unlikely that a seizure will occur.

The study will also be stopped (at least temporarily) if studies provide evidence that transcranial current/magnetic stimulation caused brain damage or other harmful effects on participants, either short-term or long-term.

Examples of findings that might trigger a safety review are the number of SAEs overall, the number of occurrences of a particular type of SAE, severe AEs/reactions, or increased frequency of events.

The IRB will also be informed promptly and provided the reason(s) for the termination of suspension of by the investigator, as specified by the applicable regulatory requirement(s).

10 SAFETY & REPORTING

It is important to assess safety over the course of this study. This section describes in detail how safety is assessed, reporting of Adverse Events, Serious Adverse Events, and Unanticipated Problems. This section is a reference for internal use.

10.1 SAFETY PARAMETERS

STIMULATION SIDE EFFECTS. A stimulation adverse effects questionnaire used in previous studies will be administered at the end of each stimulation. This questionnaire will be used as a safety measure and to collect data on participant experience. The adverse effects questionnaire asks participants to respond on a 4 point Likert scale on the severity of symptoms experienced during the stimulation session (1 = absent, 2 = mild, 3 = moderate, 4 = severe). The side effects listed are headache, neck pain, scalp pain, tingling, itching, ringing/buzzing noise, burning sensation, local redness, sleepiness, trouble concentrating, improved mood, worsening of mood, dizziness, flickering lights, and other (specify). Participants are also asked to rate on a 5 point Likert scale how related they believe the side effects to be to stimulation (1 = no relation, 2 = remote, 3 = possible, 4 = probable, 5 = definite).

10.2 METHODS AND TIMING FOR ASSESSING, RECORDING, AND ANALYZING SAFETY PARAMETERS

10.2.1 ADVERSE EVENTS

All AEs, including local and systemic reactions not meeting the criteria for "serious adverse events", will be captured on the appropriate CRF. In addition, the AE Report Form will be completed by the study coordinator (Appendix B). The AE Report Form includes the following:

- What is known about the therapy
- What is known about previous reported side effects
- If the AE occurred in temporal relation to the therapy
- Whether or not the AE improves or disappears when treatment is stopped
- Whether the AE is worsening of baseline symptoms
- Whether the AE is related to concurrent medical condition or medication use

Once complete, this form will be given to the PI, who will review, comment, and sign this form. Completed forms will be placed in the participant's folder.

In addition, the study coordinator will document any AE occurrence on the AE log (*Appendix D*), which includes information such as the date of the AE, severity, relationship to the treatment (assessed by the PI*), actions taken, and outcome(s). The log will be reviewed and initialed by the PI 72 hours after being completed. All AEs will be followed to adequate resolution and will be graded for severity and relationship to study. Any medical condition noted at the stimulation session will be considered at baseline and not reported as an AE.

***Relationship to Study Products**: The PI will determine whether an AE is associated with the study treatment. The event will be labeled associated if the event is temporally related to the administration of a therapy and no other factors can explain the event. The event will be labeled as not associated if the event is temporally independent of the study treatment and can be explained by external factors, such as major life events.

10.2.1 SERIOUS ADVERSE EVENTS

Serious Adverse Event (SAE): An SAE, as defined by the NIH, consists of adverse events that result in death, require either inpatient hospitalization or the prolongation of hospitalization, are life-threatening, result in a persistent or significant disability/incapacity or result in congenital anomaly/birth defect. Other

important medical events, based upon appropriate medical judgment, may also be considered Serious Adverse Events if a trial participant's health is at risk and intervention is required to prevent an outcome mentioned.

All SAEs will be recorded on the Serious Adverse Events Form (Appendix B), documented in the UE/SAE log and reported to the IRB. The SAE Form will be completed by the study coordinator, and includes information relating to the onset and nature of the SAE, relationship to the study treatment, seriousness of the SAE, treatment required as a response to the SAE, and outcome. This form will be filed in the participant's folder at the resolution of the event. The study coordinator will complete the UE/SAE log (Appendix D) which includes information such as the date of the event, time at which the study team was informed of the event, details, when the IRB was notified, and the date that the SAE form was completed.

10.2.2 UNANTICIPATED PROBLEMS

Unexpected Events (UE) will be recorded on the UE/SAE log (Appendix D) and will include information such as the date of the event, when the study team was informed of this event, details of the event, when the IRB was notified, and whether the SAE form was completed. The IRB will be notified of each UE that may occur during the study.

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

- 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;
- related or possibly related to participation in the research (in the guidance document, 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
- 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.

If an UE occurs the IRB will be notified and the study will be adjusted as needed to protect the health and safety of the participants. Depending on the nature of the UE, the research protocol, inclusion/exclusion criteria, and informed consent will be changed to reflect the possibility of this event reoccurring. During this time, no new participants will be recruited and the research procedures for currently enrolled participants will be stopped. Each UE will be recorded and reported throughout the study.

10.3 REPORTING PROCEDURES

We will be adopting the following table for reporting procedures:

What Event is Reported	When is Event Reported	By Whom the Event is Reported	To Whom the Event is Reported
Fatal or life-threatening unexpected, suspected serious adverse reactions	Within **24 hours** of initial receipt of information	Investigator	Local/internal IRBs

Non-fatal, non-life threatening unexpected, suspected serious adverse reactions	Within **48 hours** of initial receipt of information	Study Coordinator	Local/internal IRBs/Institutional Officials, DSMB
Unanticipated adverse device effects	Within **10 working days** of investigator first learning of effect	Investigator	Local/internal IRBs
Unanticipated problem that is not an SAE	Within **7 days** of the investigator becoming aware of the problem	Investigator	Local/internal IRBs/Institutional officials
All Unanticipated Problems	Within **30 days** of the IRB's receipt of the report of the UP from the investigator	IRB	OHRP
		Investigator	External IRBs

11.3.1 REPORTING OF PREGNANCY

Pregnancy tests will be administered on the first session of the experiment to all women of child-bearing potential (CBP). After testing negative, the experiment can continue. On subsequent sessions, female participants will be asked if it is possible that they may have become pregnant since the previous session. If the participant responds yes, or is unsure, then a pregnancy test will be administered. Within the study target population (ages 18-35), menstruating females, regardless of age are considered "CBP". Women who have had a hysterectomy or a bilateral oophorectomy are not considered of "CBP". There are no studies that suggest TMS would interfere with pregnancy. However, should a participant become pregnant during the study, her participation will be immediately terminated.

10.4 TYPES AND DURATION OF FOLLOW-UP OF PARTICIPANTS AFTER ADVERSE EVENTS

Medical monitors and PI will follow up with participants within one week of an AE.

11 STATISTICAL PLAN

The statistician for this study is Dr. Justin Riddle.

11.1 STATISTICAL ANALYSIS STRATEGIES

To control for non-specific effects of rhythmic TMS, we will use an active control: arrhythmic TMS (randomized inter-pulse-interval) that is matched to the target stimulation in number of pulses and temporal duration. All analyses will be run on the difference from condition-matched arrhythmic TMS. The key analysis is a two-way repeated-measures ANOVA with factors TMS frequency (theta vs alpha) and location (inferior-IPS vs anterior-MFG). Post-hoc t-tests will be run on a significant main effect of TMS frequency, TMS site, or interaction. We will use Tukey's adjusted significance for post-hoc testing to correct for multiple comparisons.

Primary outcome: working memory capacity. Two-way repeated-measures ANOVA run on the difference from arrhythmic TMS with factors (TMS site and TMS frequency). The direction of the retro-cue is held consistent for each site: for TMS to frontal cortex (anterior-MFG) the retro-cue to the right is used, and for TMS to parietal cortex (inferior-IPS) the retro-cue to the left is used. We hypothesize that theta frequency rhythmic TMS will improve behavioral performance when applied to anterior-MFG; i.e. a main effect of TMS frequency and/or an interaction between TMS frequency and cue. We hypothesize that alpha frequency rhythmic TMS will improve behavioral performance for retro-cue trials when applied to inferior-IPS; i.e. an interaction between TMS frequency and cue.

Primary outcome: Spectral power. We collected EEG recording during the delivery of rhythmic TMS. We hypothesize, based on previous studies with identical rhythmic TMS trains, that rhythmic TMS will entrain neural oscillations in the targeted frequency. We will quantify entrainment from rhythmic TMS by analyzing the spectral decomposition of the EEG signal during TMS and immediately after TMS. We will run a six-cycle Morlet wavelet convolution with the EEG signal to estimate spectral density (microvolts per hertz). We hypothesize that the amplitude of theta activity will be increased following theta rhythmic TMS relative to theta activity following arrhythmic TMS. Similarly, alpha amplitude will be increased following alpha rhythmic TMS relative to alpha amplitude following arrhythmic TMS. The direction of the retro-cue is held consistent for each site: for TMS to frontal cortex (anterior-MFG) the retro-cue to the right is used, and for TMS to parietal cortex (inferior-IPS) the retro-cue to the left is used. We hypothesize an interaction between TMS frequency and cue.

Primary outcome: response time. Two-way repeated-measures ANOVA run on the difference from arrhythmic TMS with factors (TMS site and TMS frequency). The direction of the retro-cue is held consistent for each site: for TMS to frontal cortex (anterior-MFG) the retro-cue to the right is used, and for TMS to parietal cortex (inferior-IPS) the retro-cue to the left is used. We hypothesize that theta frequency rhythmic TMS will improve behavioral performance (decreased response time) when applied to anterior-MFG; i.e. a main effect of TMS frequency and/or an interaction between TMS frequency and cue. We hypothesize that alpha frequency rhythmic TMS will improve behavioral performance (decreased response time) for retro-cue trials when applied to inferior-IPS; i.e. an

interaction between TMS frequency and cue.

11.2 SAMPLE SIZE DETERMINATION

Based on a previous study with the current behavioral paradigm and similar TMS manipulation, the effect size of the interaction between TMS site and TMS frequency on WM capacity was 0.532 and used 21 participants. A power analysis of this effect suggests that to reach 95% statistical power we would need to recruit 48 participants. Therefore, we will run our final analysis on 48 participants. Previous studies using a similar methodology of concurrent rhythmic TMS and EEG to look for changes in oscillatory power of the frequency of rhythmic TMS have used 10 participants for alpha frequency TMS (Thut et al., 2011) and 17 participants for theta rhythmic TMS (Albouy et al. 2017). Here, we propose to collect 48 participants. Therefore, our statistical power exceeds that of previous studies published in high-impact journals using a nearly identical methodology. Furthermore, we utilize a repeated-measures inter-mixed design in order to maximize our ability to control for confounding variables and increase statistical power. Based on a previous study by Thut et al., 2011 that found an increase in alpha oscillatory power from alpha rhythmic TMS relative to arrhythmic TMS with an effect size of 0.83 and 10 participants, we would need 21 participants to reach 95% statistical power for this comparison. Based on a previous study by Albouy et al., 2017 that found an increase in theta oscillatory power from theta rhythmic TMS relative to arrhythmic TMS with an effect size of 0.56 and 17 participants, we would need 44 participants to reach 95% statistical power for this comparison. Therefore, we estimate that based on these previously reported effect sizes for the impact of rhythmic TMS on behavior and oscillatory power measured by EEG in similar studies and our proposed sample size of 48 participants, we conclude that the proposed study has sufficient statistical power to answer the posed hypotheses.

11.3 DATA MANAGEMENT

Data will be stored in a password-protected cloud-based data system that does not contain any participant information. Individual records are referred to by dummy identifiers that cannot be traced back to the study participants except with the master code list that is stored separately in a secured location.

Information about study participants will be kept confidential and managed according to the requirements of the Health Insurance Portability and Accountability Act of 1996 (HIPAA). Those regulations require a signed participant authorization informing the participant of the following:

-What protected health information (PHI) will be collected from the participants in this study

-Who will have access to that information and why

-Who will use or disclose that information

-The rights of a research participant to revoke their authorization for use of their PHI.

In the event that a participant revokes authorization to collect or use PHI, the investigator, by regulation, retains the ability to use all information collected prior to the revocation of participant authorization. For participants that

have revoked authorization to collect or use PHI, attempts should be made to obtain permission to collect at least vital status (i.e., that the participant is alive) at the end of their scheduled study period.

The responsibilities designated to each member of the research team are documented on the Delegation of Authority Form. The study coordinator and research assistants will be responsible for the informed consent process, review for eligibility, questionnaire administration, data entry, device administration, EEG administration, and CRF entries. The study coordinator will be responsible for AE/SAE documentation and reporting, while the PI will be responsible for the AE assessment, review of the AE documentation forms, and overview of the research staff.

All data (including AEs, concomitant medications, and expected adverse reactions data) will be entered into a data capture system provided by REDCap. The data system includes password protection and internal quality checks, such as automatic range checks, to identify data that appear inconsistent, incomplete, or inaccurate. Clinical data will be entered directly from the source documents.

All assessments completed by the participant at home will be completed via REDCap as well, ensuring participant security and confidentiality.

All deviations from the protocol will be addressed in study participant source documents. The researcher will complete a Protocol Deviation Log using the participant code as the identifier. This form will collect information such as the date the deviation occurred, details of what the deviation consisted of, any corrective and preventative actions that were taken as a result of the deviation, and the date that the PI and IRB were notified. The PI will review the information and initial once approved. A completed copy of the Protocol Deviation Form will be maintained in the regulatory file, as well as in the participant's source document. Protocol deviations will be sent to the IRB per their guidelines. The site PI/study staff will be responsible for knowing and adhering to their IRB requirements.

According to the University of North Carolina at Chapel Hill's Archives and Record Management Services schedule for General Records Retention and Disposition Schedule 6.10, records will be kept for 5 years after the completion of the study or grant end date, whichever is later.

12 DATA HANDLING AND RECORD KEEPING

The study coordinator and research assistants are responsible for the accuracy, completeness, legibility, and timeliness of the data reported.

12.1 PHI AND HIPAA

Information about study participants will be kept confidential and managed according to the requirements of the Health Insurance Portability and Accountability Act of 1996 (HIPAA). Those regulations require a signed participant authorization informing the participant of the following:

- What protected health information (PHI) will be collected from the participants in this study
- Who will have access to that information and why
- Who will use or disclose that information
- The rights of a research participant to revoke their authorization for use of their PHI.

In the event that a participant revokes authorization to collect or use PHI, the investigator, by regulation, retains the ability to use all information collected prior to the revocation of participant authorization. For participants that have revoked authorization to collect or use PHI, attempts should be made to obtain permission to collect at least vital status (i.e., that the participant is alive) at the end of their scheduled study period.

12.2 CONFIDENTIALITY

12.2.1 ACCESS TO SOURCE DOCUMENTS

The research coordinator, research assistants, and PI will have access to all of the source documents collected over the course of the study. The medical monitor will have access to files upon request, as they will need access to the locked filing cabinets and rooms in which these documents are located.
Data will stay on a password-protected computer. Subsequently, a copy will be processed on a separate, password-protected desktop computer in the Frohlich Lab (Neuroscience Research Building 4129).

12.2.3 OTHER

Please note that there is no significant risk of deductive disclosure in this study. In addition, none of the groupings or subgroupings used in analysis will be small enough to allow individuals to be identified.

12.3 SOURCE DOCUMENTS

Source data is all information, original records of clinical findings, observations, or other activities in a study necessary for the reconstruction and evaluation of the trial. Source data are contained in source documents. Source data include:

PARTNERS HUMAN RESEARCH COMMITTEE (IRB).

- All IRB correspondences are documented
- The study staff is IRB approved prior to performing any study procedures
- Adverse events and deviations are reported to the IRB per current guidelines and stored appropriately
- All versions of the IRB protocols and informed consent forms are on file

INFORMED CONSENT.

- Ensure that participant identification is not recorded on the ICF (i.e., no participant ID)
- There is documentation that the participant is given a copy of the consent form
- The participant and study representative signed and dated the consent form for him/herself
- The participant initialed and dated all appropriate pages on the informed consent form

- Note to file (Appendix F) made for any informed consent deviations
- Ensure a valid (current version date) copy of the consent form was used

PROTOCOL DEVIATIONS.

- Any and all protocol deviations (exceptions and violations) are documented in the participant folder and reported to the IRB as required

OTHER SOURCE DOCUMENTS.

- Each participant folder will contain a checklist to ensure that all source documents are administered and collected properly. The checklist will be dated by the researcher for each time an assessment is administered
- Review participant folders to ensure the accuracy, completeness, and legibility of the data.
- Any correction made to the source documents is dated, initialed, and explained. The original entry should not be obscured.
- The protocol-specific source documents are on file.
- Source documents are completed in ink.
- Note to files (Appendix F) are made for missing or incomplete data and to explain any discrepancies or additional comments.

12.4 DATA MANAGEMENT RESPONSIBILITIES

The responsibilities designated to each member of the research team are documented on the Delegation of Authority Form. The study coordinator and research assistants will be responsible for the informed consent process, review for eligibility, questionnaire administration, data entry, device administration, EEG administration, and CRF entries. The study coordinator will be responsible for AE/SAE documentation and reporting, while the PI will be responsible for the AE assessment, review of the AE documentation forms, and overview of the research staff.

12.5 DATA CAPTURE METHODS (REDCAP)

All data (including AEs, concomitant medications, and expected adverse reactions data) will be entered into a data capture system provided by REDCap. The data system includes password protection and internal quality checks, such as automatic range checks, to identify data that appear inconsistent, incomplete, or inaccurate. Clinical data will be entered directly from the source documents.

All assessments completed by the participant at home will be completed via REDCap as well, ensuring participant security and confidentiality.

12.6 PROTOCOL DEVIATIONS

All deviations from the protocol will be addressed in study participant source documents. The researcher will complete a Protocol Deviation Log using the participant code as the identifier. This form will collect information such as the date the deviation occurred, details of what the deviation consisted of, any corrective and preventative actions that were taken as a result of the deviation, and the date that the PI and IRB were notified. The PI will review the information and initial once approved. A completed copy of the Protocol Deviation Form will be maintained in the regulatory file, as well as in the participant's source document. Protocol deviations will be sent to the IRB per their guidelines. The site PI/study staff will be responsible for knowing and adhering to their IRB requirements.

12.7 RECORD RETENTION

According to the University of North Carolina at Chapel Hill's Archives and Record Management Services schedule for General Records Retention and Disposition Schedule 6.10, records will be kept for 5 years after the completion of the study or grant end date, whichever is later.

13 ETHICAL CONSIDERATIONS

13.1 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 Participants of Research, as drafted by the US National Commission for the Protection of Human Participants of Biomedical and Behavioral Research (April 18, 1979) and codified in 45 CFR Part 46 and/or the ICH E6; 62 Federal Regulations 25691 (1997).

13.2 INSTITUTIONAL REVIEW BOARD (IRB)

The Office of Human Research Ethics is responsible for ethical and regulatory oversight of research at UNC-Chapel Hill that involves human participants. The OHRE administers, supports, and guides the work of the Institutional Review Boards and all related activities. Any research involving human participants proposed by faculty, staff, or students must be reviewed and approved by an IRB before research may begin, and before related grants may be funded. OHRE and the IRBs are critical components of the coordinated Human Research Protection Program, which serves to protect the rights and welfare of human participants. All components of this program must work together to ensure institutional compliance with ethical principles and regulatory requirements. The following is a mission statement for the coordinated Human Research Protection Program:

The University of North Carolina at Chapel Hill is committed to expanding and disseminating knowledge for the benefit of the people of North Carolina and the world. An important part of that commitment to knowledge is research of the highest quality on all aspects of the health and behavior of people, and such research is only possible through the participation of humans as research participants. Human participants are partners in research and a precious resource to the university. At UNC-Chapel Hill, human participant research is a privilege, but not a right. Consistent with that philosophy, it is the mission of the UNC-Chapel Hill Human Research Protection Program to ensure that:

a. The rights and welfare of human participants are paramount in the research process;
b. The highest standards of ethical conduct are employed in all research involving human participants;
c. Research investigators are properly trained in the ethical and regulatory aspects of research with human participants;
d. Research investigators deal honestly and fairly with human participants, informing them fully of procedures to be followed, and the risks and benefits of participating in research; and
e. Research using human participants at UNC-CH conforms to applicable local, state, and federal laws and regulations and the policies of the university.

13.3 INFORMED CONSENT PROCESS

Informed consent is a process that is initiated prior to the individual's agreeing to participate in the study and continues throughout the individual's study participation. Extensive discussion of risks and possible benefits of TMS will be provided to the participants and their families. Consent forms describing, in detail, the study intervention, device, procedures, and risks are given to the participant and written documentation of informed consent is required prior to the administration of any treatment or assessments used in this study. All consent forms will be IRB-approved and updated with any new information as modifications are made throughout the study.

Together, the researcher and potential participants will review the study in its entirety by reviewing the consent form together in a private location. At several intervals during the consent review, the researcher will ask the participant questions that will assess the comprehension of the information in the consent. If the participant is unsure or does not know, the researcher will return to that section and more carefully explain the information. Participants must sign the informed consent document prior to any procedures taking place. If needed, the participants will have the opportunity to discuss the study with their surrogates or think about it prior to agreeing

to participate. Participants may withdraw consent at any time throughout the course of the trial. A copy of the informed consent document will be given to the participants for their records. The rights and welfare of the participants will be protected by emphasizing to them that the quality of their medical care will not be adversely affected if they decline to participate in this study.

13.4 EXCLUSION OF WOMEN, MINORITIES, AND CHILDREN (SPECIAL POPULATIONS)

Individuals that do not speak English are excluded because the ability to accurately and complete communicate study information, answer questions about the study, and obtain consent is necessary. Pregnant participants will be excluded despite the fact that theoretical risk to mother or fetus is exceedingly small, since no safety data for pregnancy is known to exist for transcranial magnetic/electrical stimulation studies.

13.5 PARTICIPANT CONFIDENTIALITY

Participant confidentiality is strictly held in trust by the participating investigators, their staff, and the research team. This confidentiality is extended to cover testing of biological samples and genetic tests in addition to the clinical information relating to participants.

All data will only be referenced by dummy identifier code. Data will be stored on a password protected computer. A key connecting names and code numbers will be kept in a locked cabinet, accessible only by research personnel. All data will be stored and analyzed on password protected computers, also only accessible by research personnel. Participants will not be identified in any report or publication about this study. See *10 Data Handling and Record Keeping* for more information on source documentation storage and security.

13.6 STUDY DISCONTINUATION

In the event that the study is discontinued, participants who have completed or who are still enrolled in the study will be notified. Any new information gained during the course of the study that might affect participant's willingness to continue will be communicated within 2 days of the PI learning this information.

14 PUBLICATION POLICY

There are no restrictions on publications since this is an investigator-initiated study funded by a grant agency that has no influence on the publications resulting from this study.

15 LITERATURE REFERENCES

Albouy P, Weiss A, Baillet S, Zatorre RJ. Selective Entrainment of Theta Oscillations in the Dorsal Stream Causally Enhances Auditory Working Memory Performance. Neuron. Elsevier Inc; 2017 Mar 21;94(1):1–14.

Alekseichuk I, Turi Z, de Lara GA, Antal A, Paulus W. Spatial Working Memory in Humans Depends on Theta and High Gamma Synchronization in the Prefrontal Cortex. Current Biology. Elsevier Ltd; 2016 May 23;26(12):1–10.

Antzoulatos EG, Miller EK. Increases in Functional Connectivity between Prefrontal Cortex and Striatum during Category Learning. Neuron. Elsevier Inc; 2014 Jul 2;83(1):216–25.

Badre D. Cognitive control, hierarchy, and the rostro–caudal organization of the frontal lobes. Trends in Cognitive Sciences. 2008 May;12(5):193–200.

Badre D, D'Esposito M. Functional Magnetic Resonance Imaging Evidence for a Hierarchical Organization of the Prefrontal Cortex. Journal of Cognitive Neuroscience. 2007 Nov 12;:1–27.

Badre D, Hoffman J, Cooney JW, D'Esposito M. Hierarchical cognitive control deficits following damage to the human frontal lobe. Nat Neurosci. 2009 Mar 1;12(4):515–22.

Badre D, Nee DE. Frontal Cortex and the Hierarchical Control of Behavior. Trends in Cognitive Sciences. Elsevier Ltd; 2017 Dec 8;22(2):1–19.

Carver C, White T. Behavioral inhibition, behavioral activation, and affective responses to impending reward and punishment: the BIS/BAS Scales. Journal of Personality and Social Psychology. 1994;67:319–33.

Eckblad M, Chapman L. Magical Ideation as an Indicator of Schizotypy. Journal of Consulting and Clinicial Psychology. 1983;51(2):215–25.

Fries P. Rhythms for Cognition: Communication through Coherence. Neuron. Elsevier Inc; 2015 Oct 7;88(1):220–35.

Hanslmayr S, Matuschek J, Fellner M-C. Entrainment of Prefrontal Beta Oscillations Induces an Endogenous Echo and Impairs Memory Formation. Current Biology. Elsevier Ltd; 2014 Apr 14;24(8):904–9.

John O, Srivastava S. The big five trait taxonomy: history, measurement, and theoretical perspectives. Handbook of personality Theory and research. 1999. pp. 102–38.

McEvoy PM, Mahoney AEJ, Moulds ML. Are worry, rumination, and post-event processing one and the same?: Development of the repetitive thinking questionnaire. Journal of Anxiety Disorders. Elsevier Ltd; 2010 Jun 1;24(5):509–19.

Montgomery SA, Asberg M. A new depression scale designed to be sensitive to change. The British Journal of Psychiatry. 1979 Apr 1;134(4):382–9.

Oldfield R. The assessment and analysis of handedness: the Edinburgh inventory. Neuropsychologia. 1971;9:97–113.

Reinhart R. Disruption and rescue of interareal theta phase coupling and adaptive behavior. Proceedings of the National Academy of Sciences. 2017 Oct 24;114(43):11542–7.

Romei V, Thut G, Silvanto J. Information-Based Approaches of Noninvasive Transcranial Brain Stimulation. Trends in Neurosciences. Elsevier Ltd; 2016 Nov 1;39(11):782–95.

Rossi S, Hallett M, Rossini PM, Pascual-Leone A, Group TSOTC. Safety, ethical considerations, and application guidelines for the use of transcranial magnetic stimulation in clinical practice and research. Clinical Neurophysiology. International Federation of Clinical Neurophysiology; 2009 Dec 1;120(12):2008–39.

Spielberger C, Gorsuch R, Lushene R, Vagg P, Jacobs G. Manual for the State-Trait Anxiety Inventory. Palo Alto, CA: Consulting Psychologists Press; 1983.

Thut G, Veniero D, Romei V, Miniussi C, Schyns P, Gross J. Rhythmic TMS Causes Local Entrainment of Natural Oscillatory Signatures. Current Biology. Elsevier Ltd; 2011 Jul 26;21(14):1176–85.

Voytek B, Kayser AS, Badre D, Fegen D, Chang EF, Crone NE, et al. Oscillatory dynamics coordinating human frontal networks in support of goal maintenance. Nat Neurosci. 2015 Jul 27;18(9):1318–24.

Wang X-J. Neurophysiological and Computational Principles of Cortical Rhythms in Cognition. Physiological Reviews. 4 ed. 2010 Jul;90(3):1195–268.

Watson D, Clark L, Tellegen A. Development and validation of brief measures of positive and negative affect: the PANAS scales. Journal of Personality and Social Psychology. 1988;54(6):1063–70.

Wolinski N, Cooper NR, Sauseng P, Romei V. The speed of parietal theta frequency drives visuospatial working memory capacity. Jensen O, editor. PLoS Biol. 2018 Mar 14;16(3):e2005348–17.

Wutz A, Loonis R, Roy JE, Donoghue JA, Miller EK. Different Levels of Category Abstraction by Different Dynamics in Different Prefrontal Areas. Neuron. Elsevier Inc; 2018 Jan 23;97(3):1–27.

APPENDIX A: SCHEDULE OF EVENTS

Procedures	Telephone Screening Interview	Session 1: Screening	Session 2: Baseline	Sessions 3 & 4: Stimulation
Provide Verbal Consent	x			
Signed Consent Form		x		
Assessment of Eligibility Criteria	x	x		
Urine Pregnancy Test		x		
Baseline Self-Report Assessments			x	
Baseline EEG (Resting state)			x	x
Cognitive control task		x	x	x
Task EEG (during cognitive control task)			x	x
Stimulation during task				x
Stimulation Questionnaire				x

APPENDIX B: AE REPORT FORM

Adverse Effects Report:

Reasons for Report (adverse event, time, date and place of occurrence if available):

1. What do we already know about the stimulation?
    a.
2. What is the temporal relationship of the AE to the stimulation?
    a.
3. Does the AE improve or disappear when the stimulation is stopped?
    a.
4. Is the AE a worsening of baseline symptom(s)?
    a.
5. Is the AE a result of an underlying concurrent medical condition(s) or concurrent medication(s)?
    a.
6. Additional Information provided by research team
    a.

Research team member signature _____ Date_____

Co-Investigator:

_____
_____
_____
_____
_____

Steps to be taken (if applicable)

_____
_____
_____
_____

Co-I signature _____Date_____

PI Comments:

_____
_____
_____
_____
_____

Steps to be taken (if applicable)

_____
_____
_____
_____

PI signature _____Date_____

APPENDIX C: IRB AMENDMENT TRACKING LOG

Change Initiated By:		Description of IRB: Type and Brief Summary	Date Submitted to IRB	Date of IRB Response	Requires Stipulations? (Y/N)	Requires Updated Consent Form? (Y/N)	Stipulation Submission Date	IRB Approval Date
Initials	Reference ID							

APPENDIX D: AE REPORT FORM

Participant ID	**√ if AE meets definition of serious***	**Grade/Intensity** 1. Asymptomatic 2. Mild 3. Moderate 4. Severe	**Date of Incident**	**Relationship to study device** 1. Related 2. Possibly 3. Not Likely 4. Not Related	**Was Action Taken?**	**Action(s) Taken:**	**Outcome:** **1. Recovered** **2. Not Recovered** **3. Recovered w/Sequel** **4. Fatal** **5. Unknown**	**PI Initials / Date**
					Yes / No			
					Yes / No			
					Yes / No			
					Yes / No			
					Yes / No			
					Yes / No			
					Yes / No			
					Yes / No			
					Yes / No			
					Yes / No			

APPENDIX E: INFORMED CONSENT QUIZ

Name of Research Study:

You have been asked to be in a research study. This sheet will help you think of questions to ask but you may have other questions. This is not a test. We want to be sure you understand what it means to be in this research study. You should understand the research before you decide whether or not to participate.

1. What is the purpose of the research?

2. What are the possible benefits of the research?

3. What are the possible risks of the research?

4. Will everyone receive the same treatment?

5. How is this research different than the care or treatment I would get if I wasn't in the research study?

6. Does in the research cost me anything extra?

7. Can you stop being in the research once you've started?

8. Who will view your medical records?

9. Who do you call if I have questions about being a research participant?

10. Any questions?

APPENDIX F: NOTE TO FILE

IRB#: 18-1789 PI: Flavio Frohlich

Study Title: [Insert Short Name]

Researcher: _____ Date of Occurrence: _____

Participant ID: _____

Reason for Note:

_____

_____

Note:

_____

_____

_____

_____

_____

_____

_____

_____

_____

_____

_____

Corrective action (if applicable):

_____

_____

_____

Signature: _____ Date: _____

APPENDIX G: TELEPHONE SCRIPT

Telephone Consent and Screening (TMSEEG)

Date: _____ ParticipantID: _____ Criteria fulfilled: ❒ Yes ❒ No

Hello, my name is _____. I'm calling in regards to your interest in our study on the effects of brain stimulation on brain activity. Do you have about 10 minutes now to hear about the study, answer a few screening questions, and possibly schedule your visit?

(If 'No', ask for a good time to call back)
(If 'Yes', proceed)

First, I need to ask for your verbal consent to conduct the screening interview. I will ask questions about your age, medication and drug use, and family and personal health history. You may decline to answer any questions, but please keep in mind that this may affect our ability to determine if you qualify for the study. Of course, the information you provide is strictly confidential, and will not be used for any purpose other than eligibility. Do you consent to participate in the screening interview?

(If 'No', thank them for their time and hang up.)
(If 'Yes', proceed)

Great! This study is looking at how brain activity responds to magnetic stimulation. In this study, a magnetic field will be applied to your scalp. Some participants have reported mild muscle twitching and headache, but no other side effects have been found. It is not a shock and should not cause pain.

This study will include four sessions scheduled at your convenience, each lasting approximately 1-3 hours. You will be compensated for your time after each session, with total compensation amounting to $150. We will ask that you maintain a regular sleep schedule during the study, and adhere to restrictions on alcohol consumption the day prior to each session. Are you still interested in participating?

(If 'No', thank them for their time)
(If 'Yes', proceed)

Great! In order to make sure you're eligible for the study, I need to ask you a few questions. Please answer yes, no, I do not know, or I prefer not to answer. If you are not sure what the question is asking please ask for clarification. You do not need to provide any details in your answer.

Age:_____ Sex:_____

---

1. Have you ever had a serious illness/accident that required hospitalization? Are you currently receiving any medical treatment?	❒ No	❒ Yes
2. Have you ever had suffered from a brain disease (e.g. epilepsy) or concussion?	❒ No	❒ Yes
3. Have you ever been diagnosed with a learning disability or ADD/ADHD?	❒ No	❒ Yes
4. Do you suffer from a mental/psychiatric disease, such as depression or schizophrenia?	❒ No	❒ Yes
5. Is there any family history of mental/psychiatric diseases?	❒ No	❒ Yes
6. Do you consume any elicit drugs, such as cannabis?	❒ No	❒ Yes
7. Is there any chance that you are pregnant or could become pregnant during the course of the study? (If female) Please note that we will also administer a pregnancy test at the start of every stimulation session if you answer yes or are unsure.	❒ No	❒ Yes
8. Do you have any implanted electronic devices?	❒ No	❒ Yes
Study obligations		
1. Do you think you can comply with all the study duties, which include maintaining a regular sleep schedule, and no caffeine, alcohol, or excessive exercise the day before or of a session?	❒ No	❒ Yes

Keeping in mind sessions last about 3 hours, what days and times work best for you?