Temporal interference methods for non-invasive deep brain stimulation

Joshua W. Brown, Indiana University Bloomington

IRB Study # 26941

Revised April 22, 2025

Table of Contents:

1.0 Objective

There is an urgent need for new non-invasive methods of focal deep brain stimulation. Existing methods of non-invasive transcranial neurostimulation include transcranial magnetic stimulation (TMS), direct current stimulation (tDCS), alternating current stimulation (tACS), and focused ultrasound (tFUS). All are limited in that they cannot stimulate deeper brain regions, at least

without confounding stimulation of the overlying cortex. This leaves unresolved debates about the causal roles of various deep brain structures in cognition, in particular the dorsal anterior cingulate cortex. Most of what is known about deeper regions in humans comes from fMRI studies, which show *correlation* but not *causation* between activity and cognition.

Likewise, a non-invasive deep brain stimulation method could open up new ways to treat a variety of clinical issues. Invasive stimulation of the nucleus accumbens has caused a dramatic loss of addiction in opioid users and those with alcohol dependence. Likewise cingulotomy has been used to treat OCD successfully, and Parkinson's disease can be treated with invasive stimulation of the subthalamic nucleus. The surgery required presents a barrier in all of these cases, making treatment impractical short of dire illness.

Temporal interference (TI) electrical neurostimulation is a new method of non-invasive deep brain stimulation. It works by applying several pairs of electrodes to the skull, each carrying different frequencies of alternating current. The individual currents are typically 2KHz or greater and thus are low-pass filtered by neuronal membranes so that they don't activate neurons. However where two different current frequency fields intersect, they create an interference pattern that oscillates at a beat frequency equal to the *difference* between the two applied frequencies. This beat frequency can be low enough to activate neurons, and the overlap field can be steered deep into the brain. Stil, *it is relatively unknown whether TI can increase BOLD activity in deep brain regions, nor how TI may influence deeper brain regions involved in cognitive control, decision-making, and associated clinical conditions including addiction, schizophrenia, OCD, Parkinson's disease, and others.*

The overall objective of the present application is to develop and test Temporal Interference electrical neurostimulation by validating and characterizing its effects on human BOLD signals, and by demonstrating its usefulness in resolving longstanding debates about the function of a deeper brain region, the dorsal anterior cingulate cortex.

Aim 1: Test and characterize the effects of TI on the fMRI BOLD signal. We will apply TI to human subjects with fMRI. In **Study 1.1**, we will test the ability to focally activate the nucleus accumbens without activating the overlying cortex, and also its effects on functional connectivity. In **Study 1.2,** we vary the beat frequency parameter of TI to characterize its effects on BOLD inhibition vs. activation. In **Study 1.3**, we test the ability to stimulate a different deeper brain region, the dorsal ACC, and explore the optimal beat frequency.

Aim 2: Identify causal effects of dorsal anterior cingulate cortex (dACC) on cognition. We will apply TI to the dorsal ACC of human subjects with fMRI. In **Study 2.1**, we examine whether TI can disrupt or enhance the effect of conflict-monitoring signals on subsequent behavior. In **Study 2.2**, we test the theory that dACC drives motivation to avoid risk or loss, with subjects performing a previously published difficult forced-choice task while given the option to avoid the risk by skipping more difficult trials. We will test whether TI to dACC can change the probability of choosing to avoid risky choices. As time and resources permit, **Extra Study 2.3** will run a foraging task to ascertain whether TI to dACC will influence the choice probability to forage vs. influence RT when choices are most difficult.

Based on our own pilot data and others previous findings, we *hypothesize* that TI with a 20Hz beat frequency will cause increased fMRI BOLD activation in the targeted regions, without increasing activity in the overlying cortex. Further, we predict that a lower, 5Hz beat frequency may lead to BOLD inhibition as found previously. Moreover in Aim 2, we expect that TI will disrupt cognitive control signals in dACC, disrupt motivation to avoid risk or loss, and disrupt the probability of choosing to forage. If so, these findings will lay the foundation for testing clinical interventions with TI to disrupt addiction, OCD, and schizophrenia symptoms, which are known to

involve the dACC, as well as further applications of TI outside of dACC, such as non-invasive stimulation of the STN for Parkinson's disease, and the nucleus accumbens for addiction.

2.0 Background

Need for new non-invasive deep brain stimulation. There is an urgent need for deep, focal, non-invasive, and cost-effective neurostimulation methods in humans. Both basic science and clinical needs are pressing. Much of the basic cognitive neuroscience research in humans focuses on fMRI or EEG, which can show neural *correlates* of behavior but cannot show neural *causes* of behavior, strictly speaking. Likewise clinically, many targets for intervention in addiction (nucleus accumbens) and OCD (anterior cingulate) lie deep in the brain, beyond the reach of common transcranial electrical neurostimulation methods like tDCS, tACS and standard TMS. Some deep TMS methods are now FDA-approved to target deep structures like the ACC, but these lack focality and stimulate a range of areas across the prefrontal cortex(Harmelech et al., 2021). Transcranial focused ultrasound (tFUS) can provide focal stimulation (sonication) of deep structures but requires invasive skull fixation(Webb et al., 2022). A cost-effective method that allows deep, focal, non-invasive neurostimulation in humans, without also stimulating the overlying superficial cortical regions, would allow answers to many basic questions about the causal role of particular deep brain regions in cognition, especially cognitive control. Moreover, it may open the door to a number of potential clinical treatments, especially if the equipment can be made inexpensive and portable.

Temporal interference (TI) electrical neurostimulation has the potential to provide such a non-invasive deep brain stimulation method(Grossman et al., 2017). TI(C. C. Brown, 1975; Pope et al., 1995) works by administering two or more alternating currents of slightly different frequencies (Figure 1). These frequencies are typically too high to cause significant neural stimulation, on the order of 2KHz or higher, as they are filtered out by a neurons cell membrane, which acts as an electrical low-pass filter. Where two fields of slightly different frequencies overlap, they create an interference pattern (the beat frequency), with a frequency equal to the difference of the two carrier frequencies. This is analogous to the beat frequency created by plucking two guitar strings that are slightly out of tune with each other. This interference pattern can be aimed at deep brain regions, where the lower interference pattern frequency can cause neural stimulation without stimulating the more superficial cortical regions(Grossman et al., 2017). Such an effect requires at least four electrodes, with two pairs of alternating current electrodes. Early results in rodents showed that TI could be administered effectively and without tissue damage(Grossman et al., 2017). Subsequent studies also showed that high-frequency TI affects the sharp-wave ripples in hippocampus and suppresses epileptic biomarkers in mouse models(Acerbo et al., 2022) and may influence neural activity(Wessel et al., 2023) and memory encoding in humans(Acerbo et al., 2022; J. W. Brown, 2023; Violante et al., 2023). Theoretical results suggest how TI may activate sodium channels, essentially "jiggling" them open, but without activating hyperpolarizing currents in individual neurons(Cao et al., 2020).

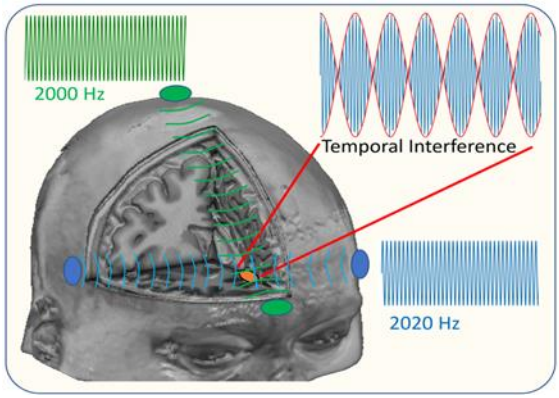

Fig. 1. Temporal interference electrical neurostimulation. Stimulation occurs where the electric fields intersect, but not from either field individually[12]. This allows deep regions

The potential implications are profound – the method may allow a number of basic science questions about the causal role of deeper brain structures to be answered, and moreover, it may allow new treatments for a number of clinical disorders that involve dysfunction of deeper brain structures (e.g. addiction(Müller et al., 2009; Zhou et al., 2011), OCD(Kim et al., 2003), and pain(Corkin et al., 1979) among others).

Developing TI neurostimulation for use in humans. Previous work by us and others has shown that TI can be safely used in humans with fMRI and may have modest value as an intervention on behavior (Modak et al., 2024; Violante et al., 2023; Wessel et al., 2023). It remains unclear exactly how TI may influence neural activity and cognitive function in healthy humans as measured by fMRI BOLD responses, and specifically how variations in the TI beat frequency may contribute to the effectiveness of TI stimulation (Aim 1), as well as how TI may influence cognitive processes in deeper brain regions (Aim 2). The present work will address these directly, showing the nature of the effects on both BOLD signals and cognition, the safety of the effects, and will provide a foundation for future work both to understand the causal role of deep brain regions in cognitive functions and develop new clinical treatments. We will first test the effects of TI on human brain activity and then demonstrate the utility of TI to adjudicate long-standing questions about the causal role of deeper brain structures in cognition, especially the dorsal anterior cingulate cortex.

Preliminary results of combined TI and fMRI

In a set of pilot subjects (Modak et al., 2024), we applied 2mA per each channel of TI stimulation using the Soterix Interferential Neuromodulation System (Soterix Medical, Inc.), via carbon fiber electrodes while subjects were scanned with whole-brain fMRI (Fig. 2). Frequencies were 2000 Hz and 2020 Hz, for a 20Hz beat frequency. We aimed to target the left nucleus accumbens, and our finite element simulations predicted an optimal electrode placement with one channel at {F9, F10} in the EEG 10-10 locations and the other channel at {Fp1, CPz} (Fig. 2 top). Each subject received alternating 2 minutes on / 2 minutes off stimulation, with 30-second current ramp up and down. Sham stimulation consisted of current ramp up followed immediately by current ramp down, to control for initial scalp tingling(Gandiga et al., 2006). The sham stimulation will be controlled by the experiment script on a computer and is designed so that subjects will not be able to discern the difference between active and sham stimulation; the subject's experience will be the same, involving mild tingling in some subjects. Subjects received four sets of on/off stimulation for each of stimulation and sham conditions, with the order of on vs. off first counterbalanced. The results (Fig. 2 bottom) show increased BOLD signal in the region predicted by the finite element simulation. Moreover, when we changed the frequencies to be the same (i.e. 2000 Hz in both channels, the high frequencies remained, but the TI beat frequency was abolished. In 8 pilot subjects, the BOLD activation seen in Fig. 2 (bottom) was likewise abolished, which suggests that TI and not the high frequencies *per se* are essential for the BOLD activation observed. Our pilot subjects tolerated the stimulation well and revealed no substantial safety concerns. These pilot results will be tested more fully in **Aim 1.**

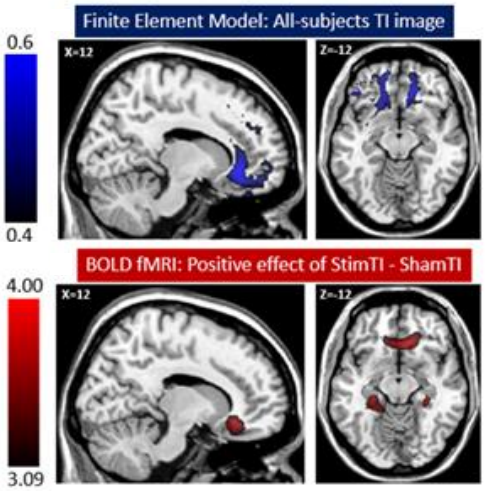

Fig. 2. Top: Finite element simulation prediction of TI effect of 2000Hz vs. 2020Hz at {F9, F10} and {Fp1, CPz}. Bottom: Increased fMRI BOLD effect due to TI in our pilot subjects (TI stimulation minus sham, Z statistic across n=16 subjects).

Characterizing the effect of TI on fMRI BOLD and cognition

The most fundamental open question here is how TI may influence activity as measured by the fMRI BOLD signal. New work suggests TI at 5 Hz beat frequency may suppress BOLD activity (Violante et al., 2023), but our pilot data with a 20 Hz beat frequency suggests TI may increase BOLD activity. **Study 1.1** will test whether a 20Hz beat frequency TI may increase BOLD activity. Second, it remains to be established whether the BOLD effects are due to the beat frequency rather than an overlap of two high frequency signals of the same frequency. **Study 1.1** also includes a comparison between 20Hz beat frequency TI vs. a control condition of zero Hz beat frequency, i.e. when the two electrode pairs have the same frequency (2000Hz). Third, because theoretical work suggests that higher beat frequencies may cause more neural excitation(Cao et al., 2020), we will directly compare 5 Hz TI vs. 20 Hz TI in **Study 1.2**. Fourth, it remains to be established how specifically TI can target various deep brain regions. In preparation for Aim 2, we will test how well TI at 20 Hz can focally target the dorsal anterior cingulate cortex (around MNI 0, 30, 30) (**Study 1.3)**.

Resolving long-standing debates about the role of dorsal anterior cingulate cortex

The causal role of several deep brain regions in cognition remains unresolved. This is because we have lacked a method to temporarily and non-invasively disrupt deep brain regions without also disrupting the overlying cortex, which leads to confounds. This limitation applies to transcranial direct and alternating current stimulation, and transcranial magnetic stimulation, even with deep TMS coils(Harmelech et al., 2021). Transcranial focused ultrasound holds some promise(Kubanek, 2018) but can only minimize the superficial stimulation, not eliminate it.

The function of the dorsal anterior cingulate cortex (dACC) in particular, including the adjacent medial prefrontal cortex, remains contested and has been the focus of vigorous and unresolved debate for nearly three decades(Clairis & Lopez-Persem, 2023). Almost all functional arguments are based on measurements of activity rather than manipulations, and so the evidence shows functional correlates rather than the causal roles. TI stimulation provides a fresh path forward to resolve some of these longstanding debates. An early theory proposed that dACC signaled conflict and thus increased control, as evidenced by the Gratton effect, in which response conflict leads to slower responses on subsequent trials(Botvinick et al., 1999). However neuropsychological studies have yielded mixed results(Fellows & Farah, 2005; Sheth et al., 2012). We will test this in **Study 2.1.** Other findings suggest a different function, i.e. that dACC drives motivation(Parvizi et al., 2013), especially to avoid risk or loss(J. W. Brown & Braver, 2007; Magno et al., 2006), which we will test in **Study 2.2.** Still others have argued that dACC drives foraging behavior(Kolling et al., 2012), with ongoing debate on whether apparent foraging value represents difficulty instead(Kolling et al., 2016; Shenhav et al., 2014), or perhaps prediction errors in service of learning(J. W. Brown & Alexander, 2017). We may test this in **Extra Study 2.3.** The unresolved debates in turn hinder our ability to understand dysfunctions of the dACC and its roles in clinical disorders such as addiction(Fishbein et al., 2005; Forster et al., 2016), OCD(Fitzgerald et al., 2005), and schizophrenia(Adams & David, 2007). In Aim 2, we apply TI to test for causal roles of dACC on behavior, with the goal of adjudicating the ongoing debates.

3.0 Design

Overview. This is a study examining the effects of TI on brain activity as measured by fMRI BOLD signals (Aim 1), and the effects of TI-related changes in deeper brain region activity on cognitive functions. Both aims build on our preliminary results showing focal changes in BOLD signals resulting from TI. In Aim 1, we will first establish the effects of TI on BOLD signals, then show that

these are not confounded with the high frequency carrier signal effects alone, and then show how TI can manipulate activity can propagate across brain networks to influence functionally connected brain regions. In Aim 2, we apply TI to stimulate a deeper brain region, the dorsal anterior cingulate cortex, to resolve longstanding questions about its causal role in cognition. We anticipate recruiting 200 subjects, allowing for attrition to achieve our goal of 30 subjects for each individual study below.

Subjects will spend 2-3 hours for the session and be paid $35/hour.

Aim 1: Test and characterize the effects of TI on the fMRI BOLD signal
In all studies, we will administer the Stimulation experience scale before starting the stimulation and after the stimulation session is complete. Subjects will also be asked periodically to rate any discomfort on a scale of 1-5 (5=highest discomfort), and indicate the location and qualities of any discomfort.

Study 1.1. Test the effect of TI in NAcc, controlling for the effects of high frequency stimulation. Healthy subjects (n=30) will be placed in the fMRI scanner and administered the same protocol used to generate our pilot results in Figure 2. Specifically, subjects will have two pairs of carbon fiber electrodes attached with conductive gel, at {F9, F10} and {Fp1, CPz}. They will receive one 8-minute block of stimulation at 2mA per electrode pair. The stimulation sequence will be 2 minutes on, 2 minutes off, 2 minutes on, and 2 minutes off with 30 second ramp up and ramp down beginning at the start of each 2-minute period. The first block will apply active TI stimulation with 2000Hz in one channel and 2020Hz in the other channel. The second block will be a sham TI stimulation, identical to the first block but with the “on” condition immediately ramping down as soon as it reaches 2mA after ramp up. The third and fourth blocks will be identical to the first and second blocks, except that both electrode pairs will stimulate at 2000Hz, resulting in a “NO-TI active” and “NO-TI sham” conditions. The order of blocks, and whether the “on” or “off” condition occurs first within a block, will be counterbalanced across subjects.

Study 1.2. Test effect of varying TI beat frequencies on BOLD inhibition vs. activation. In the same left NAcc region, also with n=30 subjects, we will repeat Study 1.1, but instead of the NO-TI condition with a zero Hz beat frequency, we will replace it with a 5Hz beat frequency (2000Hz and 2005Hz in the two channels). Likewise the 20 Hz beat frequency condition will be replaced with 30 Hz (2000Hz and 2030Hz stimulation). We will then compare the degree of BOLD activation for 5 vs. 30 Hz beat frequencies as in Study 1.1.

Study 1.3. Confirm effect of TI on the dorsal ACC, around MNI 0, 30, 30. We will repeat Study 1.1, but targeting the dorsal ACC (n=30 subjects), to confirm we can activate it with 20Hz TI. Instead of zero Hz as in Study 1.1, we will use the frequency from Study 1.2 that is found to produce the strongest BOLD activation other than 20 Hz, which will be 5Hz, 30Hz, or 40Hz. We may find that certain beat frequencies are futile, we may select an alternative beat frequency in the range of 1 to 100 Hz.

Aim 2: Identify causal effects of dorsal anterior cingulate cortex (dACC) on cognition
Study 2.1. Test whether 20Hz TI in the dACC enhances or disrupts cognitive control. For this, n=30 human subjects will be placed in the fMRI scanner to perform a flanker task(Eriksen & Eriksen, 1974), in which subjects must indicate whether the central arrow points left or right. The

trials will be 50% congruent (“<<<<<” and “>>>>>”) and 50% incongruent (“<<>><<” and “>><>>”) trials. Previous work shows not only elevated reaction time and error rates for incongruent trials, but also a “Gratton effect” in which RT and error rate (as well as dACC activity) are even more elevated for incongruent trials that follow a previous congruent trial (Botvinick et al., 1999). This is arguably because cognitive control-related activity in the dACC prior to the start of the trial was lower, thus requiring more cognitive control-related activation as a reaction to the incongruent task stimulus(Botvinick et al., 1999). We will test whether the Gratton effect is reduced by TI to the dACC in the scanner, and whether this is accompanied by increased baseline dACC activity due to TI. Subjects will be divided into two groups. One will receive 2 consecutive eight-minute blocks of TI stimulation (2000 and 2020 Hz in the two electrode pairs, or alternatively the most effective beat frequency identified in Aim 1) to dorsal ACC, followed by 2 eight-minute blocks with no-TI stimulation, as subjects perform the flanker task in the fMRI scanner. The second group of subjects will undergo an identical procedure, except that the first two blocks will be no-TI stimulation, followed by 2 eight-minute blocks of TI stimulation. In all subjects, blocks 1 and 3 will include the Flanker task, and blocks 2 and 4 will include the visual search task of Magno et al., (2006). The visual search task will provide the data needed for Study 2.2.

Study 2.2. Test the causal role of dorsal anterior cingulate in avoidance. We will apply TI stimulation as in Study 2.1 to the dorsal ACC with subjects in the fMRI scanner, and for efficiency, we will run this task on the same subjects as Study 2.1 concurrently and without leaving the scanner. Subjects will perform the task of Magno et al. (2006), during blocks 2 and 4 of the sessions described in Study 2.1. Specifically, subjects will perform a visual search task, identifying “target present” or “target absent” within a short deadline. There are two trial types, easy (few non-targets) and hard (many non-targets). They gain +1 reward for correct responses (converted to cash after the session), -1 reward for errors, and they can also skip the trial for 0 reward to avoid risk. Previous work shows dorsal ACC activity only when subjects skip the trial to avoid the risk of an error, but anterior insula activity occurs instead when subjects choose to respond and then make an error (Magno et al., 2006).

Study 2.3. Test the causal role of the dorsal ACC in foraging decisions. Using the TI stimulation of Study 2.1 with humans subjects in the scanner (n=30), subjects will perform a foraging and difficulty task(Shenhav et al., 2014) to adjudicate between competing theories of dorsal ACC, whether it drives foraging or reflects task difficulty. Subjects will be divided into two groups as in Study 2.1 and will perform the task of Shenhav et al. (2014)(Shenhav et al., 2014) during 4 eight-minute blocks, with TI and no-TI conditions in the scanner as in Study 2.1 for each of the two groups. In the task, subjects must decide whether to accept playing a gamble with known probabilities or forage to find a better gamble with more favorable probabilities. The relative value of foraging (RVF), i.e. the difference in expected value for playing the gamble vs. foraging, varies between an easy decision (large positive or negative RVF) or a difficult decision (near-zero RVF).

Risks of participation and mitigation of risks:
Subjects may feel uncomfortable completing the interview. For instance, there are questions about whether they may be pregnant, surgical history, and about the existence of tattoos and piercings on the body. They may decline to answer specific questions or decline to continue participating.

This MRI scan is not a medical test. It is designed to address research questions and it is not a

complete scan for any clinical purpose. If there is an abnormality, the scan, the MRI technician, or the researcher may not detect it. If the technician or researcher suspects a possible abnormality, the scan will be sent without any participant identifiers to a neuroradiologist for further review. If the neuroradiologist recommends further action, the subject will be notified.

MR imager: MRI has not been shown to produce health problems in normal, healthy individuals. The imager DOES NOT produce ionizing radiation, which is radiation associated with conventional radioactive sources, such as x-rays, radioactive iodine, uranium, or other substances. No medication, needle stick, or injections of drug or contrast agents are involved. There are hundreds of imagers of this type used in the U.S. and abroad, both to assist doctors in clinical diagnoses and for research. To view a copy of the Food and Drug Administration safety guidelines for MR imagers, subjects may simply ask the MRI operator.

Because of the strong magnetic fields used for MR imagers, persons who have magnetic life-support devices (e.g., pacemakers and aneurysm clips), metal prostheses or other metallic objects (e.g. cochlear implants, steel pins implanted to help repair and strengthen broken bones, metal fragments from previous injuries) cannot participate in this research. Subjects will be screened for MR contraindications

The radio frequency energy used in this exam has produced burns (most of them minor) in about one in a million cases. If subjects feel any burning sensation they will immediately inform the staff, so that the scan can be stopped.

MRI may be harmful to an unborn child. If subjects are of childbearing potential (that is, if subject is a woman with sexual partner(s) and do not use an adequate birth control method), they must be excluded. Reliable birth control (i.e. oral, implanted, or barrier methods) should be used by all participants and/or their sexual partner to prevent pregnancy while participating in MR imaging. If subjects find that they were pregnant while participating and undergoing MRI, you should notify their physician immediately. If subjects use an IUD for birth control they will be excluded unless they can document the model of the IUD and we can verify its safety for the MRI environment. Pregnancy should be self-reported, and a pregnancy test will not be administered.

While there is no evidence of increased risk with multiple scans, the risks associated with multiple scans are not known. The IUB imaging center is adopting an arbitrary maximum of 40 hours of scanning time per individual per year and the time involved in the present study is well below that limit.

Though uncommon, there is also the risk that the imaging procedure may result in nausea, dizziness, sweating, or headaches. Individuals who suffer from migraines may be more susceptible to these side effects as a result of the noise level inside the scanner. These symptoms are generally temporary, and the scan can be stopped at any time if you begin experiencing discomfort.

Brain stimulation:

1. Mild physical discomforts: Approximately 66% of participants who receive this kind of stimulation experience mild physical discomforts that are short-lived, such as tingling, itching, redness, or mild burning sensations on the skin under the pads. Some participants have also reported occasional mild headaches and fatigue. There is also a risk of seeing flashing lights.

These effects are mild, short lived, and benign. The electric current is very mild and is approximately 1000 times smaller than the average static electric shock one might receive touching a door knob or light switch after walking on a carpet with socks. While it is rare, some participants may experience a stronger unpleasant sensation. If subjects

experience any discomfort or wish to stop the experiment, subjects will let the experimenter know and the procedure will be stopped immediately.

2. Long term effects: Long term effects of exposure are largely unknown, though no long term adverse effects have been reported from any previous study. As a precaution, the stimulation will not last longer than 20 minutes continuously at one time.

3. Medical precautions: Though a significant adverse event has never been reported with similar brain stimulation procedures, there theoretically may be a risk to persons with conductive metal (i.e. implants) in their head, and to persons who suffer from migraines, epilepsy, or other neurological syndromes. If subjects have any of these conditions they may be ineligible to participate in this study. Also, subjects may be ineligible to participate in the study if they have a history of seizure disorder, history of cognitive impairment, symptoms of psychosis or if you are taking medications for cancer, attention deficit hyperactivity disorder (ADHD), autoimmune deficiency syndrome (AIDS), or other mental illness.

These risks will be managed by trained researchers; at least one will always be present when the machine is operating. The researcher will stop the experiment if they experience discomfort that you do not wish to tolerate. Furthermore, the stimulator does not transfer current greater than 3 mA per electrode pair, which is still, relatively, a very small current.

Other risks
There is a small risk of boredom during the session. Subjects can withdraw themselves if they do not wish to continue.

There is also a small risk of loss of confidentiality. We will minimize that risk by storing the records in secure (locked or password-protected) locations, Electronic data will be identified only by the subject's study ID number, with the master list linking the subject ID to identifiable information stored on paper. Electronic data will be stored on the PI's servers or on Indiana University servers. All systems will have at least password protection. Paper data will be stored in locked cabinets inside a locked room.

Study procedure summary

Days before the session:
- Recruitment (via paper flyers and digital ads)
- Pre-screening (via phone)

Day of session:
- Consent
- Fill out questionnaires
    - MR safety screening
    - Demographic form
    - Edinburgh handedness inventory
    - WHODAS 2.0
    - PHQ-9
    - GAD-7
    - Stimulation experience scale (SES)
- Subjects randomized to receive active vs. sham stimulation first, using an alternating list, in which supccessive subjects are assigned to the conditions of active first, then sham first, then active first etc.
- Brief cognitive task explanation and practice (Aim 2 only)
- Attach electrodes (day of session, in MR facility)
- Scanning while undergoing TI (and if Aim 2, performing cognitive tasks) (day of session)
    - Brief stimulation scale

    - Multiple scanning and stimulation runs, where each stimulation run is followed by administering the brief stimulation scale
- Subject removed from scanner, electrodes removed
- SES repeated
- Subject paid and dismissed

4.0 Eligibility Criteria
Inclusion:

- between the ages of 18 and 50, must have at least a 6th grade education, ability to speak and read English for all phases.

Exclusion:

- **Medical -** if they are on psychotropic medications for ADHD, other mental illness or medication for cancer, epilepsy (i.e. individuals with any history of seizure disorder), migraines, or other neurological syndromes, or AIDS (which can cause cognitive deficits (Watkins & Treisman, 2015), history of head trauma, history of cognitive impairments, metal implants in the head or under the scalp, personal experiences consistent with symptoms of psychosis (i.e. hearing or seeing things that aren't there, being secretly controlled, or having special powers, seeing or hearing things that aren't really there), history of holes bored into skull or known fissures in cranial bones
- **MRI Safety** - Subjects will be excluded if they do not meet fMRI safety screening criteria (i.e. metal implants in their body, tattoo on head or neck, permanent jewelry, etc.) or if a participants uses an IUD for birth control they will be excluded unless the subject can document the model of the IUD and we can verify its safety for the MRI environment. Pregnancy should be self-reported, and a pregnant test will not be administered. Participants must weigh less than 440 lbs. Presence of pacemakers

5.0 Outcome Characteristics and Endpoint Definitions:

For Study 2.1, Behaviorally, we predict that the Gratton effect on both RT and error rate will be reduced in active TI vs. sham stimulation conditions. We will perform an ANOVA test with factors of on vs. off TI stimulation, previous trial congruent/incongruent, and current trial congruent/incongruent. We expect to find a significant interaction, with post-hoc tests showing reduced effects of previous trial congruency, and reduced interactions of previous trail congruency with current trial congruency. Neurally, we expect to find that the BOLD signal shows a main effect of increase during the TI stimulation vs. sham. Also, we expect to find that the contrast of dACC responses to incongruent minus congruent trials is attenuated in the active TI stimulation condition vs. no-TI. If so, we will conclude that TI disrupts cognitive control by attenuating the monitoring function of dorsal ACC. Potential problems and alternate approaches It is possible that TI stimulation here may enhance rather than disrupt neural function, in which case we may find larger rather than smaller Gratton effects with TI. In that case, we will ask whether the contrast of BOLD activity for incongruent minus congruent trials in dorsal ACC is correspondingly increased during active TI vs. sham stimulation.

For Study 2.2, behaviorally, we predict that TI will disrupt the dACC, leading to a smaller likelihood of skipping difficult trials. We will perform a logistic regression to determine how TI stimulation (on vs. off) and trial difficulty (easy vs. hard), and their interaction, predict the probability of skipping a trial. Neurally, we expect to find increased BOLD signal both for TI stimulation and for skipping a trial, in overlapping regions. Potential problems and alternate approaches As with Study 2.1, it is possible that TI stimulation here may enhance rather than

disrupt neural function, in which case we may find an increase rather than decrease in the probability of skipping with TI stimulation. We will use two-tailed statistical tests to detect this possibility.

For Study 2.3, we will test whether TI to dorsal ACC increases or decreases foraging probability, or alternatively, whether TI reduces the reaction time differences of easy vs difficult decisions.

6.0 Statistical Analysis and Sample Size:

Power Analysis: From the pilot data shown in Fig. 2, we used the effect size of the stim-sham contrast to calculate the number of subjects required to detect a significant cluster of activity in the region shown. We assumed $p<0.001$ as the cluster defining threshold and required 90% power. The required number of subjects is 25, and we budget for 30 subjects per study to allow for attrition. We do not have effect size estimates for the behavioral effects of Aim 2.

7.0 Data Management

Electronic data will at all times be stored and transmitted securely, using encryption such as TLS or SSH/SFTP protocols. We will use subject ID numbers to key the data. The data will be stored and processed on PI Brown's internal lab server cluster, which gives secure nightly incremental backups to a separate location. We will use standard software tools for analysis, including PI Brown's existing fMRI processing software for the neuroimaging and temporal interference studies. The final archival data will be preserved in the IU Scholarly Data Archive.

There will also be a DSMB from IUSM (chaired by Sean O'Conner) to review the data collection and the subject safety. The DSMB will monitor the study quarterly or biannually, as the DSMB sees fit, and will consist of two medical doctors/engineers and one physician that works with TMS, a similar form of brain stimulation. In the case of an event, with regard to participant safety, occurs before 3 months after any DSMB meeting and the DSMB would otherwise wait 6 months until the next review, the PI will alert the DSMB and have that event reviewed within 3 months/quarterly.

The PI will monitor the quality of data, the number of subjects recruited, any adverse events, and any new information that comes to light that may impact the assessment of risk to the subjects. The PI will monitor the research as it is carried out, on a day-to-day basis. Formal monitoring evaluation will take place annually prior to continuing review or close-out, and quarterly or semi-annually by the DSMB, as the DSMB sees fit.

There are no statistical criteria that would lead to a premature end to the study, because a minimum sample size is necessary given the unknown statistical power. The study may be ended early if adverse events were to occur.

8.0 Principal Investigator and Study Site Qualifications and Resources

The PI Joshua Brown has published previous research on combined fMRI and temporal interference methods (Modak et al., 2024), as well as a number of papers on computational neural models and fMRI. Current research in his lab focuses on understanding real-time neural mechanisms of drug use decisions, in which heavy smokers gamble for drugs and are allowed to inhale from an e-cigarette briefly when they win a gamble -- all while in the fMRI scanner. He also has a degree in engineering with the expertise necessary to implement and use neurostimulation technology.

References

Bullen, C., H. McRobbie, et al. (2010). "Effect of an electronic nicotine delivery device (e cigarette) on desire to smoke and withdrawal, user preferences and nicotine delivery: randomised cross-over trial." Tob Control 19(2): 98-103.

Chander Bankim S., Witkowski Matthias, Braun Christoph, Robinson Stephen E., Born Jan, Cohen Leonardo G., Birbaumer Niels, Soekadar Surjo R. (2016) tACS Phase Locking of Frontal Midline Theta Oscillations Disrupts Working Memory Performance. *Frontiers in Cellular Neuroscience* 10:120.

Cohen Kadosh R, Soskic S, Iuculano T, Kanai R, Walsh V (2010) Modulating Neuronal Activity Produces Specific and Long-Lasting Changes in Numerical Competence. *Current Biol.* 20(22):2016-2020.

CentersforDiseaseControlandPrevention (2005). "Annual Smoking-Attributable Mortality, Years of Potential Life Lost, and Productivity Losses–United States, 1997–2001." Morbidity and Mortality Weekly Report 54(25): 625-628.

Devinsky, O., Morrell, M. J., & Vogt, B. A. (1995). Contributions of anterior cingulate cortex to behaviour. Brain, 118(1), 279-306.

Eissenberg, T. (2010). "Electronic nicotine delivery devices: ineffective nicotine delivery and craving suppression after acute administration." Tob Control 19(1): 87-8.

Enoch, M. A. (2008). "The role of GABA(A) receptors in the development of alcoholism." Pharmacol Biochem Behav 90(1): 95-104.

Fishbein, D. H., D. L. Eldreth, et al. (2005). "Risky decision making and the anterior cingulate cortex in abstinent drug abusers and nonusers." Brain Res Cogn Brain Res 23(1): 119-36.

Fregni, F., Liguori, F., et al. (2008). "Cortical stimulation of the prefrontal cortex with transcranial direct stimulation reduces cue-provoked smoking craving: A randomized, sham controlled study." J Clin Psychiatry 69(1): 32-40.

Goldstein, R. Z., D. Tomasi, et al. (2007). "Subjective sensitivity to monetary gradients is associated with frontolimbic activation to reward in cocaine abusers." Drug Alcohol Depend 87(2-3): 233-40.

Hester, R. and H. Garavan (2004). "Executive dysfunction in cocaine addiction: evidence for discordant frontal, cingulate, and cerebellar activity." J Neurosci 24(49): 11017-22.

Huff, W., Lenartz, D., Schormann, M., Lee, S. H., Kuhn, J., Koulousakis, A., ... & Sturm, V. (2010). Unilateral deep brain stimulation of the nucleus accumbens in patients with treatment-resistant obsessive-compulsive disorder: Outcomes after one year. Clinical neurology and neurosurgery, 112(2), 137-143.

Koob, G. F. and F. E. Bloom (1988). "Cellular and molecular mechanisms of drug dependence." Science 242(4879): 715-23.

Koob, G. F. and M. Le Moal (2001). "Drug addiction, dysregulation of reward, and allostasis." Neuropsychopharmacology 24(2): 97-129.

Kufahl, P. R., Z. Li, et al. (2005). "Neural responses to acute cocaine administration in the human brain detected by fMRI." Neuroimage 28(4): 904-14.

Kuhn, J., Lenartz, D., Huff, W., Lee, S. H., Koulousakis, A., Klosterkoetter, J., & Sturm, V. (2009). Remission of alcohol dependency following deep brain stimulation of the nucleus accumbens: valuable therapeutic implications?. Case Reports, 2009, bcr0720080539.

Matsumoto, H., & Ugawa, Y. (2017). Adverse events of tDCS and tACS: a review. Clinical neurophysiology practice, 2, 19-25.

Nachev, P., Lopez-Sosa, F., Gonzalez-Rosa, J. J., Galarza, A., Avecillas, J., Pineda-

Pardo, J. A., ... & Strange, B. (2015). Dynamic risk control by human nucleus accumbens. Brain, 138(12), 3496-3502.

Onoda, K., Kawagoe, T., Zheng, H., & Yamaguchi, S. (2017). Theta band transcranial alternating current stimulations modulates network behavior of dorsal anterior cingulate cortex. Scientific reports, 7(1), 3607.

Redish, A. D. (2004). "Addiction as a computational process gone awry." Science 306(5703): 1944-7.

Risinger, R. C., B. J. Salmeron, et al. (2005). "Neural correlates of high and craving during cocaine self-administration using BOLD fMRI." Neuroimage 26(4): 1097-108.

Starr, C. J., Sawaki, L., Wittenberg, G. F., Burdette, J. H., Oshiro, Y., Quevedo, A. S., & Coghill, R. C. (2009). Roles of the insular cortex in the modulation of pain: insights from brain lesions. Journal of Neuroscience, 29(9), 2684-2694.

Vanneste S, Walsh V, Van de Heyning P, De Ridder D (2013) Comparing immediate transient tinnitus suppression using tACS and tDCS: a placebo-controlled study. *Exp Brain Res* 226(1):25-31.

Vansickel, A. R., C. O. Cobb, et al. (2010). "A clinical laboratory model for evaluating the acute effects of electronic "cigarettes": nicotine delivery profile and cardiovascular and subjective effects." Cancer Epidemiol Biomarkers Prev 19(8): 1945-53.

Varjačić, A., Mantini, D., Levenstein, J., Slavkova, E. D., Demeyere, N., & Gillebert, C. R. (2018). The role of left insula in executive set-switching: Lesion evidence from an acute stroke cohort. cortex, 107, 92-101.

Venancio, R. C., Pelegrini, S., Gomes, D. Q., Nakano, E. Y. & Liebano, R. E. (2013) Effects of Carrier Frequency of Interferential Current on Pressure Pain Threshold and Sensory Comfort in Humans. *Arch. Phys. Med. Rehabil.* **94**, 95–102.

Watkins C, Treisman G (2015) Cognitive impairment in patients with AIDS – prevalence and severity. *HIV AIDS (Auckl)* 7:35-47.

Xu, J., A. Mendrek, et al. (2007). "Effect of cigarette smoking on prefrontal cortical function in nondeprived smokers performing the Stroop Task." Neuropsychopharmacology 32(6): 1421-8.