**Local IRB #** 202006-003 **IRBNet #** 1599521

**BUTLER HOSPITAL**

**INSTITUTIONAL REVIEW BOARD**

PROTOCOL

## 1.) Project

**Title of Project:** Effects of L-theanine on Motor Cortex Excitability in Healthy Subjects: A Paired-Pulse TMS Study

**Principal Investigator (PI):** Linda L. Carpenter, M.D.

**Other Investigator(s):** Shiwen Yuan, M.D.

## 2.) Description of Study

### A. Specific Aims

To study the effects of a single dose of L-theanine (L-γ-glutamylethylamide or $N^5$-ethyl-L-glutamine) on motor cortex excitability in healthy subjects, using paired-pulse transcranial magnetic stimulation (ppTMS) measures. The hypothesis is that L-theanine enhances motor cortex excitability in healthy subjects.

### B. Background

L-theanine is the primary psychoactive component uniquely in green tea[1]. Epidemiological studies support that green tea consumption is an independent factor associated with lower prevalence of depression[2-4]. Preclinical studies have demonstrated anti-depressant effect of L-theanine in rodents and provided evidences for its pharmacological properties of N-methyl-D-aspartate (NMDA) and gamma-aminobutyric acid (GABA) agonism[5-7]. Yet these effects have not been proven in humans. Only one open-label clinical trial[8] has studied antidepressant effect of L-theanine in major depressive disorder, and suggested its effectiveness as an augmenting agent to an existing antidepressant regimen. Furthermore, the psychotropic properties and mechanism of L-theanine are to be elucidated. Paired-pulse transcranial magnetic stimulation (ppTMS) is a well-established technique to investigate frontal cortical excitability mediated by the inter-neuron NMDA and GABA receptors[9]. Several studies[10-14] in subjects with MDD have demonstrated specific changes of ppTMS measures, including impaired short-term intracortical inhibition (SICI, mediated by GABA-A receptor and fast inhibitory postsynaptic potentials) and long-term intracortical inhibition (LICI, mediated by GABA-B receptor and slow inhibitory postsynaptic potentials) and intracortical facilitation (ICF, mediated by NMDA receptor and excitatory postsynaptic potentials), compared to healthy subjects. Treatment effects of MDD elicited by escitalopram corresponded with changes in these markers[13]. This is relevant to the study aim in that the motor cortex excitability is mediated by the glutamatergic system, and ppTMS offers an opportunity to investigate how L-theanine may manipulate the glutamatergic system in the frontal region by changing motor cortex excitability first in healthy subjects (phase 1, as described in this protocol). Then we plan to examine its potential as an augmenting agent for MDD in a placebo-controlled design (phase 2).

### C. Experimental Method

#### C1. Brief Description of Subjects

Adults (N=10 completing all 3 visits) (>18 and <65 years old) without any neuropsychiatric disorders or contraindication to application of TMS will be recruited from the local community.

#### C2. Study Design

This is a double-blinded randomized order, cross-over, placebo-controlled trial to study the acute effects of a single dose of L-theanine on the motor cortex excitability, measured by paired-pulse TMS metrics.

1) Visit 1: The subject will sign consent form to enter the study; A clinical interview (SCID 5, for 30-45min) will be conducted by a SCID interview-trained clinician (Shiwen Yuan) to ensure the inclusion and exclusion criteria are met. Releases will be signed to obtain records that confirm eligibility from a medical standpoint.

Eligible subject will then be instructed to refrain from any nutraceuticals or psychotropic medications for 7 days prior to visit 2. After medical clearance, the subject will be randomized to active or placebo pill.

2) On visit 2: the subject will receive the first session of ppTMS data collection procedures (see below for details). During this session measurement of cortical excitability happens before and after ingestion of a capsule (active L-theanine or placebo). The subject will be instructed to continue to refrain from any nutraceuticals or psychotropic medications until visit 3, which is scheduled within one week.
3) On visit 3: The subject will receive ppTMS data collection procedures that are identical to visit 2, before and 30min after drug administration. If the subject was given active L-theanine during visit 2, they will receive placebo during visit 3; if the subject was given placebo during visit 2, they will receive active L-theanine during visit 3. The PI and co-PI will still be blinded regarding the condition of the drug. Arrangements will be made for compensation and participation will conclude.

C**3. Specific Procedures or Treatments**

At both visits 2 and 3, all subjects will undergo a standard ppTMS protocol once before and again after administration of a single dose of active L-theanine or placebo. This protocol involves delivery of programed ppTMS stimuli (magnetic pulses) to the head over the motor cortex while measuring the resulting evoked motor potentials through surface electromyography (EMG) on the subject's hand.

MOTOR THRESHOLD DETERMINATION: EMG will be recorded from the right abductor pollicus brevis (APB; thumb) muscle in order to measure the amplitude of motor evoked potentials (EMG) recorded by LabChart software throughout the procedure. The subject will be seated comfortably, given earplugs, and instructed to maintain relaxed throughout the experiment. Sticky electrodes will be attached to the skin around the thumb and wrist of the subject's right hand. Single pulses of TMS will be applied to the area of the left motor cortex corresponding with right thumb muscle ABP (using with a Magstim figure-of-eight magnetic coil and Magstim stimulators) to determine the resting motor threshold (RMT), defined as the intensity that produces a MEP in relaxed APB muscle of > 50 uV during 5 of 10 trials. This value is used to calibrate the intensity of pulses for the ppTMS measures of cortical excitability.

PAIRED-PULSE TMS PROTOCOL: Paired pulses of TMS will subsequently be used to assess various measures of cortical excitability. For example, for intercortical facilitation (ICF) measurement, a subthreshold conditioning stimulus (CS) at 80% RMT precedes a suprathreshold testing stimulus (TS) at 120%RMT. Inter-stimulus intervals (ISIs) of different durations, e.g., 10 ms and 20 ms will be used. ICI will be tested with a suprathreshold CS at 120% of RMT followed by test stimulus at 120% of RMT. Again, trials will be repeated with at ISIs of different durations, e.g. 2, 5, 50, and 100ms.

Each ppTMS protocol will consist of 25 trials for each of the six conditioned stimuli in random order. MEP amplitudes will be recorded from each trial. SICI, ICF and LICI metrics are estimated from averages of the conditioned MEP amplitudes.

DRUG ADMINISTRATION: The subject will be dispensed a single dose of either drug or placebo as prepared by an unblinded pharmacist. The subject will take placebo or L-theanine 400mg of with water and remain sitting in a quiet room for relaxation for 30min, after which the ppTMS protocol will be repeated. The PI/co-PI and the subject will be blinded to the result of randomization.

SAFETY ASSESSMENTS: Prior to starting TMS procedures, the subject will be screened for TMS risk with a standard TMS Safety Screen questionnaire. Visual analog scales (e.g. sense of calm, anxiety level, sedation, irritability, level of fatigue), will be completed prior to and following each ppTMS protocol to evaluate effects of the stimulation as well as possible effects of L-theanine on mood. Verbal query will be used to elicit any other adverse effects. Subjects will be discharged from the research facility after each visit when it is clear they

are comfortable and suitable to drive home. The total estimated time for each visit is estimated to be 2.5 hours to 3 hours.

C**4. Data Analysis**

No previous studies have been done on the effects of L-theanine on intracortical facilitation or inhibition. However, many similar studies have been done with ppTMS to evaluate pharmacological effects of a drug on these same cortical excitability measures. Previous studies on the effects of NMDA receptor antagonists (dextromethorphan[15], memantine[16]) on ICF and of GABA receptor agonist (benzodiazepine[17]) on ICI. Those studies had sample sizes that ranged from 8-10 healthy individuals. The estimated clinically meaningful effect size is 10% of change in the ICI and/or ICF. Thus, after consultation with Rich Jones, PhD on sample size estimation, we plan to collect a full set of data from N=10 healthy subjects. The Pharmacy Department will participate in the process of L-theanine/placebo packing, storage, randomization and distribution.

SICI, ICF and LICI values were calculated by the ratio of conditioned MEP / unconditioned MEP. We used Wilcoxon signed-rank tests to compare the baseline-to-post-drug changes ($\Delta$=post-drug value minus pre-drug value) of SICI, ICF and LICI between placebo and L-theanine conditions. Two-sided P value < 0.05 is considered statistically significant.

**D. Material Inducements**

Subjects will be compensated in the form of gift cards for their participation:
$20 for participating in informed consent and initial SCID-5 interview;
$50 for participating in the first session of placebo/L-theanine administration and ppTMS procedures;
$80 for participating in the second/last session of placebo/L-theanine administration and ppTMS procedures.
Therefore, Subjects will be compensated in the form of gift cards for their participation as follows: $20 for completion of visit 1; $50 for completion of visit 2; and $80 for completion of visit 3. They will be compensated $150 in total if they completed the whole study.

**E. Training of Research Personnel**

The PI and co-principal investigator (Shiwen Yuan) have received SCID-5 training for clinical interview and will have TMS research credentials completed/documented by the Butler Neuromodulation Research Facility, per policy. The PIs have received CITI training program for conflict of interest in human subject research and good clinical practice, REDCap programming training for data management. A special ppTMS workshop with Pete Fried, PhD from BIDMC is planned for technical training on the ppTMS protocol.

## 3) Human Subjects

**A. Subject Population**

We plan to enroll enough subjects to ensure N=10 subjects who meet inclusion criteria complete all 3 visits:

1) Adult, aged between 18 and 65 years old;
2) Able to read/speak English and give informed consent
3) No current or history of Axis I psychiatric disorders by DSM-5.
4) Free of psychotropic medication use

Exclusion Criteria:

1) History of significant acute or chronic neurological or medical disorder or condition that increases risk for seizure with TMS;
2) History of alcohol use disorder, nicotine dependence, adjustment disorder;
3) History of allergic reactions to L-theanine or green tea;
4) Pregnancy;
5) Unable/unwilling to abstain from nutraceutical supplements and psychotropic agents during participation in the study

6) Unable/ unwillingness to refrain from recreational substance use (e.g. alcohol or marijuana) during participation in the study;
7) Meet criteria for exclusion from TMS or MRI procedures, including intracranial metal implants or nonremovable ferromagnetic items in the head/neck.

**B. Recruitment and Consent Procedures**

Subjects will be recruited through media advertisement, including social media, flyers. Potential sources of subjects include the community, hospitals, schools, etc. Telephone screening will be used to explain the study and establish preliminary eligibility for those who express interest. Written informed consent will be obtained at the first visit, after describing the study in detail and answering all questions; if COVID-19 pandemic precautions preclude in-person evaluation, virtual screening and consent procedures will take place.

Given continued COVID-19 pandemic situation, we will implement video and phone session for the first visit. A "VERBAL CONSENT FOR USE OF E-MAIL AND VIDEOCONFERENCING IN A RESEARCH PROJECT" will be used. In this consent process, prospective subjects will be contacted by phone and informed of the consent proposal. They will be asked if they understand and agree to videoconferencing for evaluation and email communication for scheduling purposes. They will need to confirm if they have access to a computer, tablet, or smartphone with a built-in camera. The investigators will then follow the scripts described in the consent form word by word to the subjects. Risks (including confidentiality) and benefits (including reduced risk of exposure to COVID-19) will be reviewed.
At the end of the consent form, the investigator will sign and date to indicate that all the instructions have been followed and the subject gave informed consent.

**C. Potential Risks**

1. We acknowledge the risk of loss of confidentiality in the course of the study.

2. Potential risks from L-theanine administration
L-theanine is a generally safe nutraceutical agent. There are two clinical trials that studied patients with MDD[8] (N=20) or schizophrenia/schizoaffective disorders[18](N=52), and reported minimal side effects from 250mg-400mg daily dosing for 8 weeks. Hidese et al[8] reported "1 patient reported slight sleepiness; 2 patients reported increased duration of sleep of up to 2 hours than usual; 2 patients reported increased dream activity. All tolerated the medication without drop-out". Ritsner et al[18] reported "L-theanine did not produce any side effects". Studies[29, 30] that administered a single dose of 250mg L-theanine before evaluating task electroencephalography (EEG) in healthy human subjects (N=13-15) reported no side effects. L-theanine was still tolerated well after two weeks of supplementing with doses nearly 60 times the usual amount used in animal studies.[19,20] Therefore, we anticipate minimal if any side effects from 400mg one-time dosing of L-theanine, as proposed in this study. Potential risks are limited to allergic reaction (extremely rare) and sleepiness/increased length of sleep or increased dream activity (rare).

3. Potential risks from ppTMS procedure
Headache (5%-10%)[21], Scalp discomfort (5%-30%)[22], dizziness or fainting episode, seizure, with an estimated risk of 0.003%[23] are the potential risks of high-frequency repetitive TMS delivered to the dorsolateral prefrontal cortex as a treatment modality. ppTMS (2 pulses each time) delivers a significantly lower amount of energy, which reduces the potential risks, but the ppTMS protocol pulses are delivered over the motor cortex, which could increase risk of seizure induction if given in high frequency trains. The ppTMS procedure does not delivery high frequency pulses and thus carries the same risks associated with a standard motor threshold procedure, which include scalp discomfort, hearing acuity damage, seizure. Single or paired-pulse TMS has an estimated risk of 2/100,000 of inducing seizure in low-risk healthy subjects (Rossi 2020, review manuscript).

## D. Protection of the Subject

### D1. Measures to Minimize Potential Risks

1. Minimize risks of loss of confidentiality: see D2.
2. Minimize potential risks from L-theanine:
    1) Clinical interview on visit 1 to ensure inclusion and exclusion criteria. Prevent subjects from exposure to L-theanine if there is a history of known allergic reaction.
    2) Observation for 1 hours after the administration of L-theanine (during ppTMS procedure and safety assessment after the procedure), to ensure that the subject has no sedation or other side effects.
3. Minimize potential risks from ppTMS procedure:
    1) Clinical interview at visit 1 to ensure inclusion and exclusion criteria.
    2) Medical clearance procedure and screening with TMS safety screen tools to prevent subjects from exposure to TMS if there is any increased risk of seizure.
    3) Comfortable positioning, ongoing direct monitoring of subject for adverse effects
    4) All TMS procedures will be conducted by PI/co-PI who are medical doctors and TMS-trained
    5) Ear plugs will be provided to subjects to prevent hearing damage.
    6) All procedures to be conducted in the BNRF, a specialized neuromodulation research facility at Butler with all-day coverage by TMS-trained physicians, emergency response buttons/protocols, etc. Subjects will also be monitored closely for dizziness, fainting or syncopal episode, and differentiate from seizure. Staff receive training in detection and management of seizure and syncope during neuromodulation procedures.

### D2. Measures to Ensure Confidentiality

1. The clinical interview and medical clearance documents will be stored in a locked cabinet within the Butler Neuromodulation Research Facility (BNRF) facility, which is locked after hours and has limited keyed access for a small number of BNRF research staff.
2. All subjects will be assigned a study ID number upon enrollment and ID will be used for research data capture.
3. This study will use Care New England's instance of REDCap for the collection and storage of data. REDCap is a secure, web-based application developed by Vanderbilt University for building and managing surveys and databases. It is primarily designed to support online or offline data capture for research studies, quality improvement, and operations. Care New England's instance of REDCap is hosted within the Care New England data center in Warwick, RI. This REDCap instance is role-based and is fully integrated with CNE's Active Directory structure. It enjoys 24/7/365 enterprise-level support and security inherit to CNE's HIPAA-compliant data center. Network transmissions (data entry, survey submission, and web browsing) to and from REDCap are protected via TLS 1.2 encryption. REDCap's data is stored on encrypted servers within CNE's data center.

### D3. Data Safety Monitoring Plan

We plan to establish a data monitoring panel with the PI (Carpenter) and co-PI (Yuan), and several unaffiliated clinical researchers with relevant expertise. The group will review the study and its progress prior to initiation. The PI/co-PI will present all adverse effect data to the panel and report any other issues related to safety. Weekly meetings between Dr. Carpenter, Dr. Yuan (and any BNRF staff involved in the project) will be held to review progress and adverse events.

We plan to do an interim analysis as mentioned above to re-estimate sample size, when N=5 subjects have been enrolled. In cases of any adverse effects from TMS or L-theanine, a comprehensive evaluation of the severity of the effect (and reported to the Butler IRB if determined a reportable event per IRB guidelines) and if

the subjects wish to stay in the study they will be aware they have the opportunity to withdraw at any time of the study. When statistically significant differences are observed for the ppTMS measures (SICI, LICI, ICF) in a dose-dependent correlation with L-theanine, or there are no differences observed whatsoever, the study will be concluded.

**E. Potential Benefits**

The study will contribute valuable data to elucidate the effects of L-theanine on motor cortex excitability. If the study yields positive results, it could support a phase 2 study on the effects of L-theanine on motor cortex excitability in patients with MDD and establish the foundation of L-theanine as a potential augmenting agent for pharmacotherapy of MDD.

**F. Risk-Benefit Ratio**

The risk-benefit ratio is estimated to be minimal considering the significant benefit that could be potentially generated from the study, versus the very low risks of the two procedures as mentioned above, as well as the measures that will be taken to further minimize those risks.

## 4) REFERENCES


*1. Vuong QV, Bowyer MC, Roach PD. L-Theanine: properties, synthesis and isolation from tea. J Sci Food Agric. 2011;91(11):1931-9.*

*2. Kim J, Kim J. Green Tea, Coffee, and Caffeine Consumption Are Inversely Associated with Self-Report Lifetime Depression in the Korean Population. Nutrients. 2018;10(9).*

*3. Hintikka J, Tolmunen T, Honkalampi K, et al. Daily tea drinking is associated with a low level of depressive symptoms in the Finnish general population. Eur J Epidemiol. 2005;20:359-363.*

*4. Hozawa A, Kuriyama S, Nakaya N, et al. Green tea consumption is associated with lower psychological distress in a general population: The Ohsaki Cohort 2006 Study. Am J Clin Nutr. 2009;90:1390-1396.*

*5. Wakabayashi C, Numakawa T, Ninomiya M, et al. Behavioral and molecular evidence for psychotropic effects in L-theanine. Psychopharmacology 2012;219:1099–1109.*

*6. Egashira N, Ishigami N, Pu F, et al. Theanine prevents memory impairment induced by repeated cerebral ischemia in rats. Phytother Res. 2008;22(1):65-8.*

*7. Lardner AL. Neurobiological effects of the green tea constituent theanine and its potential role in the treatment of psychiatric and neurodegenerative disorders. Nutr Neurosci. 2014;17(4):145-55.*

*8. Hidese S, Ota M, Wakabayashi C, et al. Effects of chronic L-theanine administration in patients with major depressive disorder: an open-label study. Acta Neuropsychiatr. 2017;29(2):72-79.*

*9. Paulus W, Classen J, Cohen LG, et al. State of the art: Pharmacologic effects on cortical excitability measures tested by transcranial magnetic stimulation. Brain Stimul. 2008;1(3):151-63.*

*10. Radhu N, de Jesus DR, Ravindran LN, et al. A meta-analysis of cortical inhibition and excitability using transcranial magnetic stimulation in psychiatric disorders. Clin Neurophysiol. 2013;124(7):1309-20.*

*11. Bajbouj M, Lisanby SH, Lang UE, et al. Evidence for impaired cortical inhibition in patients with unipolar major depression. Biol Psychiatry. 2006;59(5):395-400.*

*12. Lefaucheur JP, Lucas B, Andraud F, et al. Inter-hemispheric asymmetry of motor corticospinal excitability in major depression studied by transcranial magnetic stimulation. J Psychiatr Res. 2008;42(5):389-98.*

*13. Levinson AJ, Fitzgerald PB, Favalli G, et al. Evidence of Cortical Inhibitory Deficits in Major Depressive Disorder. Biol Psychiatry. 2010;67(5):458-64.*

*14. Jeng JS, Li CT, Lin HC, et al. Antidepressant-resistant depression is characterized by reduced short- and long- interval cortical inhibition. Psychol Med. 2019;3:1-7.*

*15. Ziemann U, Chen R, Cohen LG, Hallett M. Dextromethorphan decreases the excitability of the human motor cortex. Neurology 1998;51:1320-1324.*

*16. Schwenkreis P, Witscher K, Janssen F, et al. Influence of the N-methyl-D-aspartate antagonist memantine on human motor cortex excitability. Neurosci Lett 1999;270:137-140.*

*17. Di Lazzaro V, Oliviero A, Saturno E, et al. Effects of lorazepam on short latency afferent inhibition and short latency intracortical inhibition in humans. J Physiol 2005;564(2):661-668.*

*18. Ritsner MS, Miodownik C, Ratner Y, et al. L-theanine relieves positive, activation, and anxiety symptoms in patients with schizophrenia and schizoaffective disorder: an 8-week, randomized, double-blind, placebo-controlled, 2-center study. J Clin Psychiatry. 2011 Jan;72(1):34-42.*

*19. Rao T, Ozeki M, Juneja LR. Suntheanine: A pure and safe L-theanine dietary supplement for relaxation and stress relief. Nutracos 2007; 26-30.*
*20. Borzelleca JF, Peters D, Hall W. A 13-week dietary toxicity and toxicokinetic study with l-theanine in rats. Food Chem Toxicol. 2006;44(7):1158-66.*
*21. Taylor JJ, Borckardt JJ, George MS. Endogenous opioids mediate left dorsolateral prefrontal cortex rTMS-induced analgesia. Pain. 2012;153(6):1219-25.*
*22. Health Quality O. Repetitive Transcranial Magnetic Stimulation for Treatment-Resistant Depression: A Systematic Review and Meta-Analysis of Randomized Controlled Trials. Ont Health Technol Assess Ser. 2016;16(5):1-66.*
*23. Carpenter LL, Janicak PG, Aaronson ST, Boyadjis T, Brock DG, Cook IA, et al. Transcranial magnetic stimulation (TMS) for major depression: a multisite, naturalistic, observational study of acute treatment outcomes in clinical practice. Depress Anxiety. 2012;29(7):587-96.*
*24. Levkovitz Y, Isserles M, Padberg F, Lisanby SH, Bystritsky A, Xia G, et al. Efficacy and safety of deep transcranial magnetic stimulation for major depression: a prospective multicenter randomized controlled trial. World Psychiatry. 2015;14(1):64-73.*
*25. Rossi S, Hallett M, Rossini PM, Pascual-Leone A. Safety, ethical considerations, and application guidelines for the use of transcranial magnetic stimulation in clinical practice and research. Clinical neurophysiology: official journal of the International Federation of Clinical Neurophysiology. 2009;120(12):2008-39.*
*26. AH C. Guidance for Industry and Food and Drug Administrative Staff: Class II Special Controls Guidance Document: Repetitive Transcranial Magnetic Stimulation (rTMS) Systems. US Department of Health and Human Services, Food and Drug Administration. 2011.*
*27. McClintock SM, Reti IM, Carpenter LL, McDonald WM, Dubin M, Taylor SF, et al. Consensus Recommendations for the Clinical Application of Repetitive Transcranial Magnetic Stimulation (rTMS) in the Treatment of Depression. J Clin Psychiatry. 2018;79(1).*
*28. Wassermann EM. Risk and safety of repetitive transcranial magnetic stimulation: report and suggested guidelines from the International Workshop on the Safety of Repetitive Transcranial Magnetic Stimulation, June 5-7, 1996. Electroencephalogr Clin Neurophysiol. 1998;108(1):1-16.*
*29. Gomez-Ramirez M, Kelly SP, Montesi JL, Foxe JJ. The Effects of L-theanine on Alpha-Band Oscillatory Brain Activity During a Visuo-Spatial Attention Task. Brain Topogr (2009) 22:44–51.*
*30. Gomez-Ramirez M, Higgins BA, Rycroft JA, et al. The Deployment of Intersensory Selective Attention: A High-density Electrical Mapping Study of the Effects of Theanine. Clin Neuropharmacol 2007;30:25-38.*

**5) CRITERIA FOR WAIVER OF AUTHORIZATION FOR USE OF PROTECTED HEALTH INFORMATION (PHI)**

**5A. Does the requested use of PHI involve more than minimal risk to privacy?**

☐ **YES** ***[if " YES," project is not eligible for PHI Waiver]***
☒ **NO** ***[if "NO," address 1-3 below]***

1. **Plan to Protect Patient Identifiers from Improper Use and Disclosure:** The clinical interview and medical clearance documents will be stored in a locked cabinet within the BNRF facility, which is locked after hours and has limited keyed access for a small number of BNRF research staff. All subjects will be assigned a study ID number upon enrollment and ID will be used for research data capture. All data will be de-identified during analysis.

2. **Plan to Destroy Identifiers or Justification for Retaining Identifiers:** The investigators plan to destroy identifiers once the data that need to be collected using the identifiers (including obtaining medical clearance documents from PCP) are obtained, by assigning study ID upon enrollment, shredding identifiable paper documents, etc.

3. **Assurances that the PHI will not be Re-used or Disclosed:** The investigators assure that the PHI will not be reused or disclosed.

**5B. Could the research be practicably conducted without a waiver?** ☒ **YES** ☐ **NO**
**5C. Could the research be practicably conducted without access to and use of the PHI?** ☐ **YES** ☒ **NO**
**5D. PHI is only needed for activities preparatory to research** ☐ **YES** ☒ **NO**

**6) DESCRIPTION OF PHI TO BE COLLECTED UNDER WAIVER**

Name, age, sex, handedness, contact information, medical history, substance and nutraceutical use history. (Medical clearance documents to be requested from PCP will be obtained following signature on a separate CNE Authorization form)

**7) ADVERTISEMENTS**

The advertisement will be submitted as amendment if the protocol is approved.

**8) RESRULTS INFORMATION**

Primary results:

Compared to matching placebo, 400 mg single dose L-theanine elicited significantly higher post-pre drug change ($\Delta$) of ICF (Mean±SE $\Delta ICF_{L\text{-}theanine}$=0.073±0.073 vs. $\Delta ICF_{Placebo}$=-0.341±0.176, p=0.016, Cohen's d = 1.02) and LICI (Mean±SE $\Delta LICI_{L\text{-}theanine}$=0.145±0.100 vs. $\Delta LICI_{Placebo}$=-0.068±0.053, p=0.037, Cohen's d = 0.91) within each individual. No significant difference was found for SICI ($\Delta SICI_{Placebo}$= 0.005±0.060 vs. $\Delta SICI_{L\text{-}theanine}$=0.059±0.028, p=0.450, Cohen's d = 0.35). L-theanine did not change unconditioned MEPs (or baseline rMT) differently than placebo (Table 1). No adverse effects from L-theanine were observed. The subjects were unable to guess the condition of drug (placebo vs L-theanine) after each session of experiment more accurately than random. (SE = standard error of the mean)