EXPLORATIONS OF AUDITORY THETA RHYTHMS TO IMPROVE HUMAN COGNITION

by

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Recent research on improving human cognition has emphasized stimulation of the brain with sensory, electrical, or magnetic stimulation. This is done to improve disorders or enhance normal cognition. Auditory or electrical stimulation in the theta range (4-8 Hz) has been found in some studies to enhance memory and attention in typically developing young adults. The mechanism of this effect is unknown but may include changes in mood or motivation or more specific changes in underlying neural functions such as synaptic plasticity. My thesis tries to replicate these findings by comparing brief auditory stimulation with a control group of pink noise and examining the effects of theta on mood, attention, and memory. While my findings did not successfully replicate previous studies using auditory theta stimulation, I found significant correlations between mood and attention and mood and memory, supporting the body of evidence that mood impacts both working memory and attention. Further studies determining the effects of brief auditory theta entrainment on cognition are necessary to continue building the body of research about improving attention networks exogenously.

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Introduction

Recent studies have found that neuromodulation improves several aspects of human cognition. Neuromodulation is generally defined as brain stimulation that results in the manipulation of brain activity in one or more target regions in the brain. Sensory, electrical, and magnetic stimulation are the most highly researched and most clinically performed forms of neuromodulation. Electrical stimulation uses alternating or direct current to entrain brain activity in brain areas, often at particular frequencies designed to enhance brain plasticity.

Deep Brain Stimulation (DBS), a more invasive form of electrical stimulation, involves surgically implanted electrodes connected to a pulse generator. The most popular clinical use of electrical stimulation is to treat Parkinson's disease or migraine headaches by either stimulating the vagus nerve on the neck or the trigeminal nerve on the forehead (Benabid, 2003; Little et al., 2013; Schoenen et al., 2013; Volkmann, 2004; Yarnitsky, et al., 2017).

Non-invasive methods can be used with volunteer participants. Research with normal volunteers has found that electrical stimulation of areas such as the anterior cingulate, medial prefrontal cortex and/or ventro-lateral prefrontal cortex may improve attentional control and memory (Piscopo et al., 2018; Reinhart, 2017; Reinhart & Nguyen, 2019; Weible, Rothbart, Posner & Niell, 2017)

Magnetic neuromodulation involves a pulsed magnetic field that causes a current flow in the brain. Placed above the scalp is a magnetic coil that produces a brief, high-current pulse, thus producing a magnetic field (Hallet, 2000). Unlike electrical stimulation, magnetic stimulation does not require direct contact with the scalp.

Researchers have used magnetic stimulation as an investigatory tool to research motor function, language, vision, and brain disorders (Hallet, 2000). Researchers have also found that magnetic neuromodulation can potentially be used therapeutically to alleviate depression and migraine headaches (Conforto et al., 2014; Connolly et al., 2012).

Sensory neuromodulation has been found to be an effective tool for modulating behavior. Forms of sensory neuromodulation that have been examined include visual, somatosensory, and auditory stimulation. Clinicians and researchers performing visual stimulation sometimes modulate visual perception by using an optokinetic drum (a rotating cylinder with alternating black and white stripes) or creating images on a computer monitor of linearly moving white dots or stripes presented on a black background (Karnath, 1996; Vicario et al., 2007). Visual stimulation is often used to treat individuals with nystagmus, disturbed perception of body orientation, and unilateral spatial neglect (Iacarino et al., 2016; Karnath, 1996; Vallar et al., 1993). Somatosensory stimulation involves placing contact electrodes on the tongue and has been found to help those with balance disorders (Wildenberg et al., 2010, 2011).

Auditory stimulation involves the presentation of a sound with a particular amplitude that reflects the brain oscillation the researcher or clinician wishes to manipulate in the brain. Intensity, frequency, and entrainment are key parameters to ascertain that the acoustic stimulus frequency will interact with internal brain oscillator. A researcher or clinician chooses the frequency of their auditory stimulus based on the desired behavioral result of the subject. For example, researchers may use acoustic stimulation between 0.5-3Hz to enhance slow-wave sleep (Bellesi et al., 2014). This frequency mirrors the natural delta brainwave which is generated during non-REM sleep. Recent studies have also found that auditory stimulation using theta frequencies of 4-8Hz improves attentional control and memory (Roberts, Clarke, Addante, and Ranganath, 2018).

Brain oscillations are defined as the rhythmic firing of a local neuronal population. This neuronal coordination allows for network communication within a region or separates regions in which the oscillations are correlated. Different oscillation frequencies are associated with different cognitive states. For example, the gamma frequency (32-100 Hz) is associated with heightened perception and cognitive processing while the delta frequency (0.5-4 Hz) is associated with deep sleep. The theta frequency (4-8 Hz) has been implicated with states of restful awareness and is frequently found during cognitive processing (Cavanaugh, et al., 2014).

Theta oscillations are associated with several aspects of cognition including successful learning and retention, memory encoding and retrieval, attentional and self-control, and improvement in overall mood. A study in the rat hippocampus found that intrinsic theta oscillations function by temporally structuring neural ensemble activity during both memory retrieval and spatial navigation (Hasselmo et al., 2002). Studies looking at human EEG recordings in the 4-8 Hz frequency range found theta oscillations present in the hippocampus, medial prefrontal cortex, parietal lobes, and anterior cingulate cortex. Depending on the brain region stimulated, stimulating theta may impact different aspects of cognition. For example, Reinhart (2017) found that in-phase electrical theta stimulation (6 Hz) of the medial prefrontal cortex and ventral-lateral prefrontal cortex improved attentional control of behavioral timing. These brain regions are important nodes of the executive attention network and therefore theta stimulation of this region improved attention-related tasks.

Piscopo et al. (2018) found that targeted electrical stimulation of the anterior cingulate (ACC) (4-8) in mice led to white matter increase in the region, demonstrating that theta stimulation of the ACC can cause changes in white matter in mice. They also found that frontal theta is implicated in the activation of previously dormant oligodendrocytes responsible for myelination, therefore increasing white matter in the region following theta stimulation.

Other studies have found that auditory theta stimulation improves cognition related to memory in humans (Reinhart & Nguyen, 2019; Roberts, Clarke, Addante, & Ranganath, 2018). In one memory study (Roberts, Clarke, Addante & Ranganath, 2018), participants were presented with two sets of words respectively classified as “alive” or “manmade” and then asked to identify whether the word is classified as animate versus inanimate or manufactured versus natural. Following this task, the experimental group was presented with auditory theta stimulation (5.5 Hz) (or sham stimulation) for 36 minutes. They were then presented with a memory task in which they were asked to recognize words exposed to them previously and the context in which the words were presented (“alive” vs “manmade”). The group exposed to theta stimulation performed better at determining the classification of each word but mirrored the control group in recognition of whether the word was from the previous task. One potential hypothesis to explain this improvement in memory is that the increase in intrinsic theta following auditory stimulation may enhance synchronization of the information recalled prior to the enhanced theta activity and would thus improve participants’ retrieval scores.

Research has also found that theta oscillations following meditation training amplified theta around the ACC and prefrontal cortex (PFC) (i.e., frontal theta), leading to improved attention, self-regulation, stress-levels, and mood (Tang et al., 2007; Lutz et al., 2008; Tang & Posner, 2009; Holzel et al., 2011; Tang, 2011; Tang et al., 2012). For example, Tang et al (2010) found that one month of integrative body-mind training (IBMT) led to an increase in white matter around the ACC and PFC. Although these results were found using meditation practice alone in comparison with an active control group given relaxation training, they demonstrate that amplified frontal theta improves participants’ mood and other cognitions in addition to memory.

Because these theta studies with human participants used stimulation too brief to produce significant white matter change as a mechanism to improve cognitive functions (unlike with rodents, as demonstrated by Piscopo et al, 2018), other synaptic mechanisms are likely responsible for these cognitive improvements. For example, Larson & Mukascy (2015) demonstrated that a fast burst of theta stimulation more successfully induced hippocampal long-term potentiation when synchronized with intrinsic theta rhythms. Long term potentiation is the process of strengthening synapses so that long-term signal transmission increases between neurons. This finding indicates that brief theta stimulation, while not necessarily inducing white matter change, may improve cognition by enhancing endogenous theta and increasing synaptic plasticity.

A recent study compared the ability of auditory and electrical theta stimulation to enhance endogenous theta within the ACC (Voelker et al., 2020). It was found that combining electrical or auditory stimulation and an attention task that activated the ACC produced a significant increase in intrinsic theta following stimulation in comparison to either stimulation or the task alone. Because the Covid-19 pandemic required us to operate remotely, we decided to examine the ability of auditory theta to improve memory, attention, and mood. By examining both mood and performance we might be able to determine whether any enhancement of memory and attention correlates with mood change.

In this study we aim to determine whether brief auditory theta stimulation enhances memory, attention, and mood in comparison to pink noise, which serves as our control condition. To determine improvements in memory, we used a visual working memory test (Luck & Vogel, 2013) where participants were presented with 5 colored squares distributed across the screen. After a brief delay the participants were then asked to report the colors presented by selecting from a set of options at each location. To determine improvements in attention we used the Attention Network Test (ANT), which measures the efficiency of the alerting, orienting, and executive network. Finally, we measured mood using the Profile of Mood States (POMS) questionnaire where the participant responds to a list of 40 moods using a 5-point Likert scale.

Each participant will undergo two hour-long sessions; in one session they will be presented with theta stimulation, and in another they will be presented with pink noise (a random noise having equal energy per octave) a week apart. Before and after stimulation, participants underwent the working memory task followed by the POMS questionnaire. During stimulation each participant performed the Attention Network Task (ANT). I hypothesize that positive mood will be enhanced, and negative mood reduced in the theta condition compared to the control condition. Theta stimulation should also improve working memory performance, orienting attention, and mood in comparison to pink noise stimulation. I will also examine possible correlations between mood, memory, and attention performance to determine if mood is a likely reason for improved performance.

Methods

Participants

96 volunteer participants between the ages of 18-30 were recruited by personal contact, flyers, or through the pool maintained by the psychology department at the University of Oregon. The research was approved through the University of Oregon Institute Review Board. Participants were randomly assigned to one of two groups, varying only in the order in which they received theta and control stimulation.

Two 1-hour sessions were scheduled, consisting of one experimental treatment and one control treatment. 71 of the recruited participants successfully completed both sessions, which were separated by a one-week interval. All sessions operated remotely (using the website Gorilla: https://app.gorilla.sc) and consisted of the following: a pre- and post-stimulation visual working memory test and POMS questionnaire, and an experiment or control auditory stimulation while performing the ANT.

Materials

The working memory test presented to participants is a change detection task presented on the computer. This task is based on the Visual Working Memory (VWM) capacity test developed by Luck & Vogel (2013), which is a reliable measure of working memory. Four blocks of 40 trials are presented, where the participant is shown 2-6 randomly chosen colored squares at 2-6 different locations on the screen and, after a brief delay, is required to report the color at each location. We measured the capacity of visual working memory performance using the K value; the formula is written below (Pashler, 1988):

$$K = setsize * (P(hit) - P(FA))(1 - P(FA))$$

Where P(hit)=hits/(hits + misses) and P(FA)=false alarms/(false alarms + correct rejections)

In order to induce ACC activity through an executive control-requiring task, all subjects performed the Attention Network Task (ANT) (Fan et al., 2002) during theta or control stimulation. Using computer graphics, the ANT presents five arrows in a horizontal row that appear above or below the fixation point. Subjects press a key indicating the direction the central arrow is pointing, which may require them to ignore the flanker arrows. Two sets of 64 trials were presented, with an equal but random assortment of the cueing and congruence conditions. Completion of the task allows calculation of three scores related to the efficiency of attentional networks, measuring how response times are influenced by alerting cues, spatial cues, and flankers. The alerting scores (median RTs for trials where the cue does not appear minus median RT in trials where the double cue appeared) provide a measure of the benefit in performance provided by a warning signal. The orienting measure (median RTs for trials with a central cue minus median RT in trials with orienting valid-cues) includes both the benefit obtained from a valid cue and the cost of the spatially uninformative cue. The executive attention score (median RTs for incongruent minus median RT in congruent) indicates the amount of interference experienced in performing the task when stimulation conflicting with the target was presented in the display. Larger interference scores indicated less efficiency in resolving conflict.

We also used an abbreviated, revised POMS (McNair, Lorr, & Droppleman, 1971) to measure mood state before and after theta or sham stimulation. The participant responded to a list of 40 moods using a 5-point Likert scale.

Participants will either receive pulses of auditory theta stimulation at two tones that are separated by a 6 Hz that converge every 6 Hz to produce an audible pulse, or pink noise, which is a random noise that has equal energy per octave (thus having more low-frequency components than white noise). All participants were requested to use over-the-ear headphones or earbuds for auditory stimulation. The participant was asked to indicate their available listening device before the onset of auditory stimulation.

Experimental Procedure

The entire experimental procedure took place on the online experiment builder, *Gorilla.* Participants had access to the website through a secure link sent to their email or SONA account. Firstly, the participants completed a virtual consent form online and provided an electronic signature. This consent form was approved by the University of Oregon IRB and listed all risks of participation, a general outline of what will take place during the study, and a reminder that they may decide to not take part in the study at any time for any reason. During this phase, participants were also asked to silence and set aside cellphones and to avoid noise distractions during the experiment. Participants were then required to fill out a demographic questionnaire detailing sex, age, ethnicity, years of post-secondary education, and smoking habits. Following completion of the demographic questionnaire, participants filled out the POMS questionnaire.

Participants were then introduced to the memory task. They began with a set of practice trials where they are notified whether their answers were correct. Upon completion of the memory task, participants were asked to listen to the theta frequency, (experimental group) or pink noise (control group) while completing a session of the ANT. The ANT was the only session performed with sound, while the mood and memory measures were taken both before and after the sound. I attempted to have the participant adjust the sound level to approximately 55 decibels, the volume of sound typically experienced during a verbal conversation. The theta stimulation was provided as two tones (100 and 106 Hz) producing sound with an amplitude varying at 6 Hz. The pink noise consisted of all audible tone frequencies, with each random noise having an equal energy per octave. Both auditory stimuli were provided continuously as the participant performed the ANT. Upon completion of the task, participants took the visual working memory test and completed the POMS an additional time.

There was an error in which some participants (6 theta, 28 control) only received 50% to 99% of sound during the ANT trials rather than the full 100% sound duration (41 theta, 10 control) during their first session. To address this issue, we ran 20 participants who we were certain received 100% of the appropriate sound during all ANT trials. We then utilized a within subject comparison to determine whether there was a significant difference in ANT, K, or POMS performance between participants who experienced 50%-99% sound versus participants who experienced 100% sound. We found no significant difference and therefore included participants who received 50%-100% sound. Thus, we used all participants who had more than 50% of the ANT trials with the appropriate sound.

Analysis

We looked at Within Subject comparison for the tasks repeated in a session, namely the POMS and K. We also looked at between subject comparisons to investigate differences in ANT performance. We are interested in determining whether the condition type (theta or control) influenced performance. Additionally, we looked for learning effects over repeated tasks. Based on previous work, we expected most of the effects of theta on K would be seen in the first session.

Results

ANT

Mean values for total reaction times and percentage error are shown in *Table 1*, in addition to alerting, orienting, and conflict for sessions 1 and 2. Among participants in session 1 who received sound during the ANT for at least 50% of the task (theta stimulation n=43; control n=35), there was a marginal difference in total reaction time between treatment groups (t (82) =-1.92, p=.059) where the control group performed a little faster. Between groups, alerting, orienting, and conflict were not different (alert: t (82) =0.77, p=.443, orienting: t (82) =-1.12, p=.267, conflict: t (82) =0.66, p=.514). The incongruent reaction time was marginally different between groups (t (82) =-1.96, p=.054), with the control group performing faster.

Among participants in session 2 who received sound during the ANT for at least 50% of the task (theta stimulation n=43; control n=35) there was no significant difference in total reaction time (t (71) =.65, p=.518). Between groups alerting, orienting, and conflict were similarly not different (alert: t (71) =0.64, p=.518, orienting: t (71) =0.70, p=.487, conflict: t (71) =0.51, p=.61). Therefore, there was no effect of theta on ANT performance in session 2. Alerting, orienting, and conflict scores were all positive as was obtained in previous work (Fan et al., 2002).

65 participants successfully performed both sessions with more than 50% sound in each. Using a paired samples t test to determine a change in performance between sessions 1 and 2 we found that there was a significant improvement in incongruent reaction time (t (64) =2.61, p=0.011) and conflict (t (64) =3.34, p=.001). Total reaction

time also trends in this direction (t (64) =1.806 p=0.076). Upon performing a repeated measure within subject analysis, we found that the total reaction time was slower overall in the theta group (F (1,63) =6.45, p=0.014), which was not influenced by condition order (F (1,63) =2.55, p=.115).

Session 1	**ToRT**	**Alert**	**Orient**	**Conflict**	**%error**
Theta N=47					
Mean	588.02	18.55	45.99	71.76	0.1
SD	68.71	45.76	55.2	49.99	0.13
Control N=38					
Mean	576.13	17.92	34.54	70.1	0.09
SD	45.2	35.28	39.43	37.48	0.18
Session 2	**ToRT**	**Alert**	**Orient**	**Conflict**	
Theta N=37					
Mean	551.26	17.35	43.88	46.47	0.07
SD	69.67	32.53	34.85	43.79	0.06
Control N=36					
Mean	561	26.63	39.39	51.88	0.06
SD	69.67	73.69	29.62	47.51	0.06

Table 1: Mean, standard deviation, and % error for reaction time (RT) and ANT scores for sessions 1 and 2

POMS

Mean values for average positive and negative mood scales before and after stimulation for session 1 and session 2 are listed in *Table 3.* Between subjects, there was no significant difference in mood between the theta and control groups for session 1. Using a repeated measure analysis, we did identify a within-subject change in positive mood during the first session. Positive and negative moods are listed in *Table 2.* Both components of the positive mood, esteem-related affect (ERA) and vigor significantly declined from the before after treatment (ERA: F (1,83) =19.39, p<.001, vigor: F (1,83) =33.77, p<.001). The vigor measure showed a marginal difference between conditions (F (1,83) =3.60, p=.061), with the control group showing a sharper decline with testing. There was a significant within-subject decline in depression with stimulation (F (1,83) =13.96, p<.001). The other mood measures did not differ significantly.

Vigor and esteem-related effect significantly declined between test 1 and test 2 in the second session (VIG: t (72) =2.93, p=0.005, ERA: t (72) =2.71. p=0.008) and TMD (a negative measure subtracting negative measures of mood from positive measures of mood) marginally increased (t (72) =-1.91, p=0.60). There is no evidence that theta influenced any other significant difference in POMS performance.

The control-first group had a significantly larger decline in VIG during the control session, and a relatively smaller decline during the theta session. The decline in VIG is consistent between sessions among the theta-first group. Therefore, VIG is significantly different by condition (F (1,63=4.07, p=0.048) and by order of condition (F (1,63) =4.33, p=0.042).

Positive Mood	**ERA** (esteem related affect): Proud Competent Confident Satisfied Embarrassed (reversed) Ashamed (reversed)	**VIG** (vigor): Lively Active Energetic Full of pep Vigorous			
Negative Mood	**TEN** (tension): Tense On-edge Uneasy Restless Nervous	**DEP** (depression): Unhappy Sad Hopeless Discouraged Miserable Helpless Worthless	**ANG** (anger): Angry Grouchy Annoyed Resentful Bitter Furious	**FAT** (fatigue): Worn out Fatigued Exhausted Weary Bushed	**CON** (confusion): Confused Can't concentrate Bewildered Forgetful Uncertain

Table 2: List of subscales of positive and negative moods used in POMS questionnaire

Session 1	**Neg1a**	**Neg1b**	**Pos1a**	**Pos1b**
Theta N=44				
Mean	23.66	21.11	20.07	17.82
SD	15.97	13.59	7.89	7.19
Control N=36				
Mean	21.19	20.22	22.11	18.75
SD	18.72	18	8.31	7.5
Session 2	**Neg2a**	**Neg2b**	**Pos2a**	**Pos2b**
Theta N= 37				

Mean	16.76	17.84	21	19.14
SD	11.43	13.87	8.25	9.18
Control N= 36				
Mean	18	17.5	21.78	20
SD	15.41	13.03	6.99	7.29

Table 3: Mean and standard deviation for negative and positive moods before and after stimulation for sessions 1 and 2

K

There was no significant difference in performance in K by treatment group (F (1,83) =0.72, p=.399) or within-subject between before and after measurements (F (1,83) =1.34, p=.290) in session 1. Additionally, there was no apparent change in K performance over the four sessions (F (2,192) =1.51, p=.213,). However, when comparing the amount of improvement within a session (after-before) between sessions 1 and 2 is a practice effect. Performance was significantly improved in the first session (F (1,64) =4.90, p=0.030, *See Figure 1*)

Session 1	**KbeforeS1**	**KafterS1**	**K(after-before)**
Theta N=44			**N=65**
Mean	2.81	2.82	.1
SD	.60	.67	.29
Control N=36			
Mean	2.93	3.02	
SD	.55	.59	

Session 2	**KbeforeS2**	**KafterS2**	**K(after-before)**
Theta N= 37			**N=65**
Mean	2.86	2.86	-0.32
SD	0.66	0.66	0.33
Control N= 36			
Mean	2.92	2.96	
SD	0.65	0.67	

Table 4: Mean and standard deviation for K scores before and after stimulations for sessions 1 and 2

Correlations

Change in K Performance and Orienting in ANT

In the first session, there was a significant correlation between change in K performance and orienting in the ANT (r (84) =0.22, p=0.048, r=Pearson's correlation (degrees of freedom)). Among those with greater improvement in the K task over time, these participants also demonstrated a greater orienting score (or larger difference in reaction time when not receiving an orienting cue than when receiving it). However, change in K does not correlate with orienting in session 2 (r (72) =0.06, p=6.48, r=Pearson's correlation (degrees of freedom)). As demonstrated in *Figure 1,* the change in K is significant from session 1 to 2 and does not vary by condition.

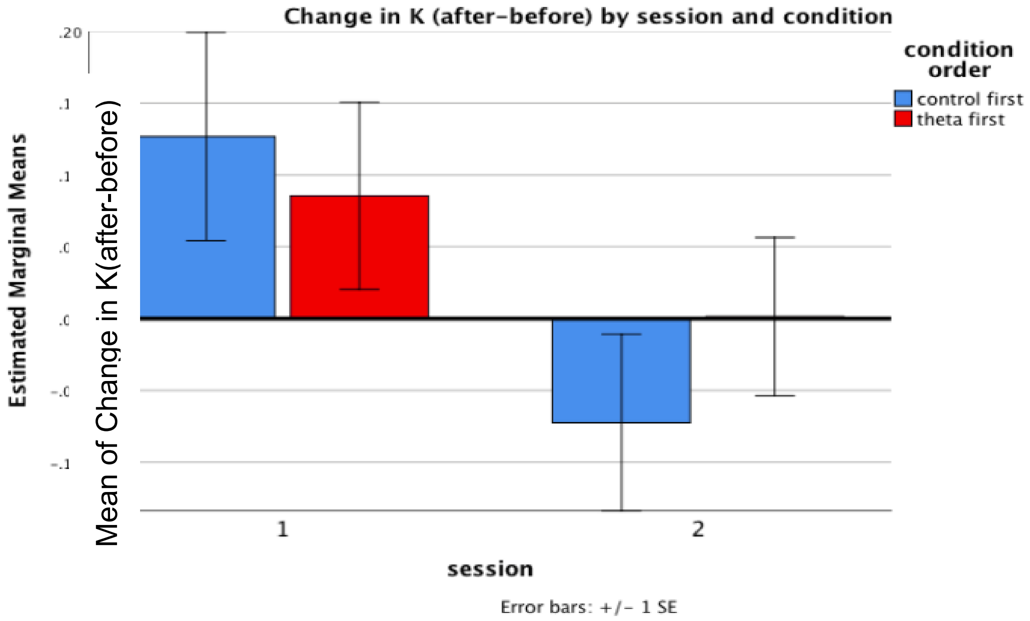

Figure 1: Change in K before and after stimulation by session and condition

POMS and K

There was a significant correlation between POMS scores of anger (ANG) and depression (DEP) and K after stimulation in session 1 and a significant correlation between DEP and K before stimulation in session 2 (see *Table 5*).

Session 1 N=85	**K (after)**	**ANG (after)**	**DEP (after)**
Pearson Correlation	1	-0.3	-0.24
Sig. (2-tailed)	.	.006	.025
Session 2 N=73	**K (before)**	**ANG (before)**	**DEP (before)**
Pearson Correlation	1	-0.28	-.31
Sig. (2-tailed)		0.016	.009

Table 5: Correlation between POMS and K before and after stimulation by session

Discussion

I hypothesized that theta stimulation would improve working memory performance, orienting attention, and mood in comparison to pink noise stimulation. In addition, I hypothesized that positive mood would be enhanced, and negative mood reduced in the theta condition compared to the control condition. I also aimed to determine whether mood, memory, and attention performance were correlated. Overall, there was little difference between the theta and control groups in mood, memory, and attention. While depression was reduced in both groups, positive mood also declined across both groups. Although there were some significant correlations between mood and attention and mood and memory, we did examine many correlations, making it likely that these could be due to chance. If further research shows reliable correlations of this type, it will be necessary for researchers to consider more carefully the possible role that mood might play in changes observed in memory or attention.

Mood

While there wasn't a significant difference in mood between theta and control groups, we identified a within-subject decline in positive mood (ERA and VIG) before and after auditory stimulation in both the first and second session. The control-first group demonstrated a sharper decline in vigor than the theta group and demonstrated a relatively smaller decline during their theta session. This potentially demonstrates a marginal effect of theta on positive mood decline. Despite the decrease in positive mood, individuals demonstrated a significant decline in depression following stimulation. However, the evidence is not compelling enough to determine whether theta influenced any difference in mood changes.

Attention

While the control group performed marginally faster in total reaction time than the theta group in session 1, there was no significant difference in total reaction time in session 2. Additionally, there was no difference between groups in alerting, orienting, and conflict, demonstrating that there was likely no effect of theta on ANT performance in either session. However, in all cases the three derived scores were positive as has been reported in previous studies. There was significant improvement in incongruent reaction time and conflict and marginal improvement in total reaction time between sessions for both groups. These improvements are likely a result of practice and has been seen in other studies with repeated sessions of the ANT

The total reaction time was marginally longer overall in the theta group, which was not influenced by condition order. These results mirror previous findings that participants undergoing brief auditory theta stimulation trended toward slower total reaction times than those undergoing brief electrical stimulation or sham stimulation during the ANT (Voelker et al., 2020). Due to the briefness of the theta entrainment brain connectivity was likely unaffected. Because frontal theta is amplified during meditative states (Xue, Tang, Tang, & Posner, 2014), the slower reaction time could be attributed to a more relaxing treatment in comparison to pink noise. Also, while meditation appears to improve theta activity and support cognitive function, external auditory theta stimulation (as delivered in this experiment) could interfere with endogenous theta rhythms and result in less efficient processing.

Memory

There was no significant difference in performance in K between the control and theta group and before and after stimulation for either sessions 1 and 2. There were significant improvements by participants in the first session, however, indicating that short term practice improved memory scores. There was no evidence of improved performance in session 2, suggesting that practice effects on K occur only at the start of training.

However, we did identify a significant correlation between change in K performance and orienting solely in session 1. Those who improved more in K over time also demonstrated a greater orienting score. In other words, those who performed better in K with practice were better able to use the positioning cue to undergo the ANT more quickly. This demonstrates a relationship between orienting in attention and memory improvement. Because K assesses spatial working memory, it is likely those who show better orienting efficiency would also show better improvements in spatial memory. This finding supports the existing literature that neural systems for visual orienting relate to spatial working memory (Corbetta, Kincade & Schulman, 2002).

Study Limitations

Use of Pink Noise Control

We chose pink noise as the control because it is a random noise that has equal energy per octave, which contrasts auditory theta at two tones 6Hz apart. However, pink noise (in relation to silence or white noise) has steady low-frequency components which may have replicated the effects of theta on the listener. Additionally, studies have shown that pink noise may reduce the brain wave complexity resulting in a restful state (appropriate for stable sleep) (Zhou et al., 2012), thus inducing a similar state as theta. A benefit of using pink noise is that it offers a steady sound across a spectrum of frequencies, as opposed to no noise, which may lead the listener to become distracted by uncontrolled noises in the environment (including their own breathing). However, it is possible we saw little difference in mood, attention, and memory between the control and theta group because the low-frequency nature of pink noise mirrors auditory theta.

Brief Stimulation

Additionally, it is possible that the theta entrainment session was too brief to affect any significant changes to mood, memory, or attention. Although brief theta stimulation has been shown to successfully induce hippocampal long-term potentiation when synchronized with intrinsic theta rhythms (Larson & Mukascy, 2015), it is possible that auditory theta stimulation may take longer to take effect than electrical theta-burst stimulation. In Voelker et al.'s (2020) study, they found that combining electrical stimulation and ANT produced a larger increase in intrinsic theta than auditory stimulation and ANT. While there was a significant increase in intrinsic theta with combined auditory stimulation and ANT compared to ANT or stimulation alone, the brief auditory theta entertainment in this experiment may have not increased intrinsic theta enough to improve mood, attention, or memory in comparison to control participants.

Remote Experiment Structure

Because this experiment was operated entirely remotely, it was impossible to fully monitor participant performance. There was no way to enforce an optimally controlled environment for participants or to enforce compliance and attention. Thus, it was difficult to determine the level of discipline and compliance of each participant or whether they understood the instructions for each task. Additionally, we encountered an error in which some participants only received 50%-99% of the sound during ANT trials rather than the 100% sound duration during their first session. Furthermore, the participant was responsible for their own computer, internet, and headphones. Therefore, each participant underwent the tasks with their own personal devices. Overall, the remote format of this experiment allowed for variation in participants' environments, level of compliance, and allowed for technological errors. It is strongly recommended that this study be replicated post-COVID-19 so that researchers may enforce a controlled environment for participants.

Future Issues

Theta Stimulation to Enhance Cognition and Treat Disorders

Although I found no evidence of cognitive improvement in the theta group in relation to the control group, there have been successful reports of improvement in memory (Reinhart & Nguyen, 2019; Roberts et al., 2018) using brief auditory theta stimulation. There is no evidence that such brevity of auditory stimulation could lead to white matter increase; therefore the mechanism leading to improved cognition following brief theta must involve increasing the amplitude of intrinsic theta (Larson &

Mukascy, 2015). One possible mechanism for improved cognition may involve phase locking of high frequency spiking within the theta rhythm, thus potentially enhancing the likelihood and speed of Long-Term Potentiation (LTP)-induced synaptic change (Albensi, Oliver, Toupin & Odero, 2007; Lynch, 1998). When intrinsic theta is amplified, LTP may be enhanced and thus improve memory storage. Brief auditory theta stimulation repeated over time may enhance synaptic plasticity and eventually increase brain connectivity as seen in mice in the Piscopo et. al. (2018) study.

Auditory theta entrainment to enhance synaptic plasticity may mitigate symptoms of individuals with substance abuse, a condition which involves a disordered executive attention system. Tang, Tang, & Posner (2013) found that two weeks of mindfulness meditation training (which has been found to increase midline theta) improved connectivity in the ACC and led to a 60% reduction in smoking in participants who had no intention or knowledge of their smoking cessation. Although the research is preliminary, it provides compelling evidence that amplification of theta frequencies led to increased connectivity that mitigated symptoms of substance abuse in individuals with no intention to quit or reduce their smoking. A future study using auditory theta entrainment on chronic smokers could strengthen this preliminary evidence and show the potential effects of synaptic plasticity enhancement by amplifying intrinsic theta.

Auditory theta stimulation could also be used to treat mental illnesses potentially associated with the executive attention network including anxiety and attention deficit hyperactivity disorder (ADHD) (Ghassemzadeh, Posner & Rothbart, 2019; Heeren, Hoebeke & Coussement, 2019; Posner, Rothbart & Ghassemzadeh, 2019). For many mental health disorders, the underlying physiology remains unknown. Therefore, further studies are necessary to determine whether theta stimulation (via auditory or electrical entrainment) may mitigate symptoms of the disorder.

Conclusions

My hypothesis, based on prior human studies, was that brief auditory theta entrainment while performing a task known to amplify intrinsic theta would enhance cognition. Specifically, I hypothesized that working memory performance, attention, and mood in comparison to pink noise stimulation would improve following auditory theta stimulation. I also examined possible correlations between mood, memory, and attention performance. My effort to test this hypothesis showed significant correlations between mood and memory and mood and attention but did not show improved mood, memory, and attention relative to condition/theta.

I cited several studies showing cognitive improvements in memory and attention following brief auditory and electrical theta entrainment. I also described the connection between brief theta stimulation, amplification in intrinsic theta, increased LTP, and enhanced synaptic plasticity. Further studies should be conducted to determine whether brief auditory theta stimulation over time leads to structural changes in the brain. In addition to pursuing further studies with healthy participants, clinical trials to determine whether auditory theta entrainment mitigates symptoms in patients with disordered mental states could lead to breakthroughs in noninvasive treatments for individuals with conditions such as ADHD and substance abuse disorders.

Bibliography

Albensi, B.C., Oliver, D.R., Toupin, J., & Odero, G. (2007) Electrical stimulation protocols for hippocampal synaptic plasticity and neuronal hyper-excitability: Are they effective or relevant? *Experimental Neurology* 204, 1–13

Bellesi, M., Riedner, B. A., Garcia-Molina, G. N., Cirelli, C., and Tononi, G. (2014). Enhancement of sleep slow waves: underlying mechanisms and practical consequences. *Front. Syst. Neurosci*. 8:208. doi: 10.3389/fnsys.2014.00208

Benabid, A.L. (2003). Deep Brain Stimulation for Parkinson's Disease. *Current Opinion in Neurobiology, 13(6), 696-706.*

Cavanagh, J. F., & Frank, M. J. (2014). Frontal theta as a mechanism for cognitive control. *Trends in cognitive sciences*, 18(8), 414-421.

Conforto, A. B., Amaro, E. Jr., Gonçalves, A. L., Mercante, J. P., Guendler, V. Z., Ferreira, J. R., et al. (2014). Randomized, proof-of-principle clinical trial of active transcranial magnetic stimulation in chronic migraine. *Cephalalgia* 34, 464–472. doi: 10.1177/0333102413515340

Connolly, K. R., Helmer, A., Cristancho, M. A., Cristancho, P., and O'Reardon, J. P. (2012). Effectiveness of transcranial magnetic stimulation in clinical practice post-FDA approval in the United States: results observed with the first 100 consecutive cases of depression at an academic medical center. *J. Clin. Psychiatry* 73, e567–e573. doi: 10.4088/jcp.11m07413

Corbetta, M., Kincade, J. M., & Shulman, G. L. (2002). Neural systems for visual orienting and their relationships to spatial working memory. *Journal of cognitive neuroscience*, 14(3), 508-523.

Fan, J., McCandliss, B.D., Sommer, T., Raz, M. & Posner, M.I. (2002). Testing the efficiency and independence of attentional networks. *Journal of Cognitive Neuroscience*, 3(14):340-347.

Ghassemzadeh, H. Rothbart, M.K. & Posner, M.I. (2019) Anxiety and brain networks of attentional control. *Cognitive and Behavioral Neurology* 32/1, 54-62

Hallett, M. Transcranial magnetic stimulation and the human brain. *Nature*, 406, 147–150 (2000). https://doi.org/10.1038/35018000.

Hasselmo, M. E., Bodelón, C., & Wyble, B. P. (2002). A proposed function for hippocampal theta rhythm: separate phases of encoding and retrieval enhance reversal of prior learning. *Neural computation*, 14(4), 793-817.

Heeren, A., Hoebeke, Y., & Coussement, C. (2019). Unfolding the complex dynamic interplay between attentional processes and anxiety: A commentary on Ghassemzadeh, Rothbart, and Posner. *Cognitive and behavioral neurology,* 32(1), 63-66.

Holzel, B.K, et al. (2011) How does mindfulness meditation work? Proposing mechanisms of action from a conceptual and neural perspective. *Perspect Psychol Sci 6*:537–559.

Iaccarino, H. F., Singer, A. C., Martorell, A. J., Rudenko, A., Gao, F., Gillingham, T. Z., et al. (2016). γ frequency entrainment attenuates amyloid load and modifies microglia. *Nature* 540, 230–235. doi: 10.1038/nature20587

Karnath, Hans-Otto. "Optokinetic stimulation influences the disturbed perception of body orientation in spatial neglect." *Journal of Neurology, Neurosurgery & Psychiatry* 60, no. 2 (1996): 217-220.

Larson, J., & Munkácsy, E. (2015). Theta-burst LTP. *Brain research*, 1621, 38-50.

Little, S., Pogosoyan, A., Neal, S., Zavala, B., Zrinzo, L., Haritz, M., … & Brown, P. (2013). Adaptive Deep Brain Stimulation in Advanced Parkinson Disease. *Annals of neurology,* 74(3), 449-457.

Luck, SJ & Vogel, EK (2013) Visual working memory capacity: from psychophysics and neurobiology to individual differences. *Trends in Cognitive Science* 17/8 391-400

Lutz, A.; Slagter H.A.; Dunne, J.D.; & Davidson, R.J. (2008) *Attention regulation and monitoring in meditation*. Trends Cogn Sci 12(4):163–169.

Lynch, G. (1998) Memory and the brain unexpected chemistry and new pharmacology *Neurobiology of Learning and Memory* 70, 82-100

McNair, D. M., Lorr, M., & Droppleman, L. F. (1971). *Manual for the profile of mood states (POMS*). San Diego: Educational and Industrial Testing Service.

Pashler, H. (1988). Familiarity and visual change detection*. Perception & psychophysics,* 44(4), 369-378.

Piscopo, D., Weible, A., Rothbart, M.K., Posner, M.I. & Niell,C.M. (2018) Changes in white matter in mice resulting from low frequency brain stimulation. *Proceedings of the National Academy of Sciences*, 115/27 6639-6646. https://doi.org/10.1073/pnas.1802160115

Posner, M.I., Rothbart, M.K. & Ghassemzadeh, H (2019) Restoring Attention Networks. *Yale Journal of Biology and Medicine* 92/1, 139-143.

Reinhart R.M.G and Nguyen J.A (2019). Working memory revived in older adults by synchronizing rhythmic brain circuits. *Nature Neuroscience,* 22(5):820-827

Reinhart R.M.G. (2017). Disruption and rescue of interareal theta phase coupling and adaptive behavior. *Proceedings of the National Academy of Sciences,* 114(43):11542-11547.

Roberts, B.M., Clarke, A., Addante, R.J., & Ranganath, C. (2018). Entrainment enhances theta oscillations and improves episodic memory. *Cognitive Neuroscience*, 9(3-4), 181-193.

Schoenen, J., Vandersmissen, B., Jeangette, S., Herroelen, L., Vandenheede, M., Gerard, P., et al. (2013). Migraine prevention with a supraorbital transcutaneous stimulator: a randomized controlled trial. *Neurology* 80, 697–704. doi: 10.1212/WNL.0b013e3182825055

Tang, Y.Y. (2011) Mechanism of integrative body-mind training. *Neurosci Bull* 27(6):383–388.

Tang, Y.Y, Lu, Q., Geng, X., Stein, E.A., Yang, Y., & Posner, M.I. (2010) Short term mental training induces white-matter changes in the anterior cingulate *PNAS* 107 16649-16652

Tang, Y.Y., Lu, Q., Fan, M., Yang, Y., & Posner,M.I. (2012) Mechanisms of White Matter Changes Induced by *Meditation Proceedings of the National Academy of Sciences USA* 109 (26) 10570-10574 doi10/.1073pnas.1207817109

Tang, Y. Y., Ma, Y., Wang, J., Fan, Y., Feng, S., Lu, Q., ... & Posner, M. I. (2007). Short-term meditation training improves attention and self-regulation. *Proceedings of the National Academy of Sciences,* 104(43), 17152-17156.

Tang, Y.Y. & Posner, MI. (2009) Attention training and attention state training. *Trends Cogn Sci* 13(5):222–227.

Tang, Y.Y., Tang, R., & Posner, M. I. (2013). Brief meditation training induces smoking reduction. *Proceedings of the National Academy of Sciences*, 110(34), 13971-13975.

Vallar, Giuseppe, Gabriella Antonucci, Cecilia Guariglia, and Luigi Pizzamiglio. "Deficits of position sense, unilateral neglect and optokinetic stimulation." *Neuropsychologia* 31, no. 11 (1993): 1191-1200.

Vicario, Carmelo Mario, Carlo Caltagirone, and Massimiliano Oliveri. "Optokinetic stimulation affects temporal estimation in healthy humans." *Brain and cognition 64,* no. 1 (2007): 68-73.

Voelker, P., Parker, A. N., Luu, P., Davey, C., Rothbart, M. K., & Posner, M. I. (2020). Increasing the amplitude of intrinsic theta in the human brain. *AIMS neuroscience,* 7(4), 418.

Volkmann, J. (2004). Deep brain stimulation for the treatment of Parkinson's disease. *Journal of Clinical Neurophysiology*, 21(1), 6-17.

Weible, A.P., Piscopo, D.M., Rothbart, M.K., Posner, M.I.,& Niell, C.M. (2017) Rhythmic Brain Stimulation Reduces Anxiety-Relate Behavior in a Mouse Model Based on Meditation Training. *Proceedings of the US National Academy of Sciences,*114/no10 2532-2537.

Wildenberg, J. C., Tyler, M. E., Danilov, Y. P., Kaczmarek, K. A., and Meyerand, M. E. (2011). High-resolution fMRI detects neuromodulation of individual brainstem nuclei by electrical tongue stimulation in balance-impaired individuals. *NeuroImage* 56, 2129–2137. doi: 10.1016/j.neuroimage.2011.03.074

Wildenberg, J. C., Tyler, M. E., Danilov, Y. P., Kaczmarek, K. A., and Meyerand, M. E. (2010). Sustained cortical and subcortical neuromodulation induced by electrical tongue stimulation. *Brain Imaging Behav*. 4, 199–211. doi: 10.1007/s11682-010-9099-7

Xue, S., Tang, Y., Tang, R., & Posner, M. (2014). Short-term meditation induces changes in brain resting EEG theta networks. *Brain And Cognition*, 87, 1-6. doi: 10.1016/j.bandc.2014.02.008

Yarnitsky, D., Volokh, L., Ironi, A., Weller, B., Shor, M., Shifrin, A., & Granovsky, Y. (2017). Nonpainful remote electrical stimulation alleviates episodic migraine pain. *Neurology*, 88(13), 1250-1255.

Zhou, J., Liu, D., Li, X., Ma, J., Zhang, J., & Fang, J. (2012). Pink noise: effect on complexity synchronization of brain activity and sleep consolidation. *Journal of theoretical biology*, 306, 68-72.