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Effects of theta-frequency binaural beats on post-exercise recovery and stress responsivity

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EFFECTS OF THETA-FREQUENCY BINAURAL BEATS ON POST-EXERCISE RECOVERY AND STRESS RESPONSIVITY

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CERTIFICATE OF APPROVAL

MASTER OF SCIENCE THESIS

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ABSTRACT

Binaural beats are an auditory illusion perceived when two or more pure tones of similar frequencies are presented dichotically through stereo headphones. This phenomenon is thought to have the potential to facilitate changes in arousal. The present study investigated the effects of 7 Hz binaural beating on post-exercise recovery and stress responsivity in college-aged students (n = 21; 18-29 years old). Psychological measures consisted of the positive and negative affect schedule and self-reported stress and relaxation. Physiological measures consisted of heart rate, heart rate variability, blood pressure and salivary α-amylase. Participants visited the laboratory twice over a two-week period at the same time of day to complete both a control (pink noise and carrier tone placebo) and an experimental (multilayered theta-frequency binaural beats with pink noise) session in a crossover controlled design. Participants exercised for 20-min at 70% of their predicted VO_{2max} with a 5-min warm-up and cool-down at 50% of their exercise workload. After exercising, participants listened to either binaural beats or placebo while relaxing alone in a quiet, low-light environment. Lastly, participants completed a short, timed 20-question mental arithmetic battery designed to elicit a stress response. Measurements were taken at four time points during each session: at baseline, post-exercise, post-relaxation and post-stress. Heart rate variability analyses comprised 2-min windows corresponding to salivary-amylase acquisition. Theta binaural beats failed to outperform placebo in altering post-exercise recovery or stress responsivity. However, after listening to binaural beats, participants reported feeling more relaxed (6.4% change) and less stressed (11.5% change). Findings from the present study suggest that listening to binaural beats may have subtle psychological effects. Further research in a sample more representative of the general population is recommended.

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CHAPTER 1

INTRODUCTION

Historical records show that for thousands of years music has served as a vehicle for human consciousness exploration, trance induction, ritual healing and cultural expression (Fachner & Rittner, 2011). Anecdotal evidence abounds touting the power of music to affect consciousness while technological advances pave the way for a clearer understanding of the role music plays in human experience. For example, advances in electroencephalography (EEG) have afforded the ability to study, in more nuanced detail, the neural mechanisms through which music is created and subsequently acts upon human nervous systems.

In the early 1960's ethnomusicologist Andrew Neher conducted a series of studies to examine the effects of music on human brainwaves, as measured by EEG (Neher, 1961, 1962). Through this work, Neher devised the 'auditory driving' hypothesis, providing empirical support for the notion that human brainwave rhythms (i.e., delta, theta, alpha, beta and gamma frequency ranges) could be experimentally induced through exposure to musical tempos that correspond to those frequency ranges (Turow, 2005). The principle underlying auditory driving is called entrainment: the process through which two or more rhythmic oscillators interact and become synchronized (Clayton, Sager, & Will, 2005).

In the mid 1970's, Robert Monroe, a disc jockey and radio station owner, patented a sound technology called the Hemi-Sync® auditory-guidance system (Atwater, 2004). At the core of this system were binaural beats, an auditory illusion experienced by the listener when pure tones of similar frequencies are presented to each ear via stereo

headphones (Oster, 1973). Monroe's nonprofit research and educational institution, the Monroe Institute (TMI), has produced a wide variety of Hemi-Sync® products which combine binaural beats, music, pink sound (sound frequencies spanning the entire audible spectrum), natural surf sounds and verbal guidance (Atwater, 2004). Taken together, the elements of the Hemi-Sync® system are designed to facilitate access into various ergotropic (increased arousal) and trophotropic (decreased arousal) altered states of consciousness (ASCs) (Atwater, 2004). Given the various elements comprising the Hemi-Sync® system, multiple forms of entrainment are possible, making empirical study of potential causal mechanisms extremely difficult. Tempo-entrainment has already received a great deal of empirical support (Clayton et al., 2005); yet, the effects of frequency-entrainment remain unclear. In other words, it is presently uncertain whether binaural beats alone can facilitate changes in physiology and behavior.

Several neuroimaging studies have demonstrated successful frequency-entrainment to binaural beats through measurement of evoked auditory-steady-state responses (ASSRs) (Draganova, Ross, Wollbrink, & Pantev, 2008; Karino et al., 2006; Pratt et al., 2009; Schwarz & Taylor, 2005). However, these studies did not examine any subjective psychological correlates or physiological indices of altered states. Two studies have examined the effects of isolated binaural beats combined with pink noise, without accompanying music or verbal guidance and both of these studies demonstrated significant results seemingly indicative of successful frequency-entrainment. The first study used theta-frequency patterns to increase theta-band power and hypnotic susceptibility (Brady & Stevens, 2000). The second study used beta-frequency patterns to increase performance vigilance and mood (Lane, Kasian, Owens, & Marsh, 1998).

While the results of these studies are intriguing and appear to support the role of binaural beats in frequency-entrainment, they rely heavily on psychological dependent variables. Future research must additionally examine the effects of binaural beats on other physiological markers of ASCs.

Stress researcher Herbert Benson spent years studying the common patterns underlying ASCs elicited through various meditative practices, ritual technologies and secular practices such as hypnosis (Benson, 1982). He found that all trophotropic practices had a common foundation: the elicitation of the relaxation response (Benson, 1982). The relaxation response is an innate physiological response characterized by diminished sympathetic nervous system activity, decreased heart rate, blood pressure and increased theta-brainwave activity (Benson, Arns, & Hoffman, 1981). Therefore, if binaural beats are effective as entraining stimuli, the theta frequencies used to increase hypnotic susceptibility (Brady & Stevens, 2000) should improve the depth and speed of the relaxation response as measured by psychological and physiological variables.

A recent study provided support for a combined exercise (ergotropic) and relaxation (trophotropic) training framework, demonstrating a reduced blood pressure response to psychological stress relative to control groups (Santaella et al., 2006). This study was limited in that blood pressure was the only dependent variable used and "shavasana" (a yogic meditative practice) was their choice of relaxation intervention. Though putatively effective, meditation involves extensive practice and often includes spiritual overtones - and thus, is commonly met with resistance by many. Therefore, it seems logical to extend these findings to more secular methods. Combining exercise training with binaural beat technology has never been studied, yet this combination

potentially offers access to a deeper relaxation response with no prerequisite experience or training of the listener.

Statement of Purpose

The present study investigated the effects of theta-frequency binaural beat patterns on post-exercise recovery and subsequent stress reactivity, as measured through multiple psycho-physiological variables.

Hypotheses

The null hypotheses for this study are:

1. Theta-frequency binaural beat patterns, in combination with pink noise, will have no effect on the post-exercise relaxation response during the 20-min relaxation protocol, compared to a pink noise and carrier tone placebo control, measured by alterations in heart rate variability measures.
2. Theta-frequency binaural beat patterns, in combination with pink noise, will have no effect on post-exercise recovery, compared to a pink noise and carrier tone placebo control, measured by alterations in positive affect, negative affect, self-reported relaxation, salivary α-amylase, blood pressure, heart rate and heart rate variability measures.
3. Theta-frequency binaural beat patterns, in combination with pink noise, will have no effect on the post-relaxation stress response, compared to a pink noise and carrier tone placebo control, measured by alterations in positive affect, negative affect, self-reported stress, salivary α-amylase, blood pressure, heart rate and heart rate variability measures.

Assumptions of the Study

For the purpose of this study, the following assumptions will be made at the start of the investigation:

1. Hormonal circadian fluctuations will not impact the results of the study as long as all participants are scheduled at the same time of day for each trial.
2. Individual differences (e.g., aerobic fitness, exercise history, personality, etc.) will be adequately controlled for with the repeated-measures design.
3. The test-retest reliability of the positive and negative affect schedule (PANAS) is sufficient to provide reliable results in a crossover design comprised of two experimental sessions.
4. The test-retest reliability of the mental arithmetic test is sufficient to provide a reliable stress response in a crossover design comprised of two experimental sessions.
5. Focus of attention during each condition will be adequately controlled for by providing a generalized verbal instruction: “Focus attentively on the music and relax as deeply as you can.”
6. The stress response can be quantitatively measured by changes in salivary α-amylase, blood pressure, heart rate and heart rate variability indices.
7. Depth of relaxation response can be adequately inferred by relative alterations in positive and negative affect, self-reported stress and relaxation, salivary α-amylase, blood pressure, heart rate and heart rate variability indices.
8. The 20-min exposure to binaural beats will be adequate to entrain the CNS to produce theta cortical rhythms.

Definition of Terms

1. Evoked Auditory-Steady-State Response (ASSR): neural potentials produced in response to repeated auditory stimuli which can be reliably measured via scalp electrodes (EEG) (Hofmann & Wouters, 2010).
2. Binaural Beat: an auditory illusion experienced subjectively (and measured objectively via evoked ASSRs) when two pure-tone sine waves of similar but different frequencies (under 1500 Hz and less than 40 Hz apart) are presented dichotically to each ear (Draganova et al., 2008). The consistent phase difference between the sound waves results in the experience of an amplitude-modulated standing wave which is processed by the brainstem's superior olivary nuclei and inferior colliculi (Monroe, 1993).
3. Entrainment: a process through which two autonomous rhythmic oscillators with similar but different fundamental frequencies interact (Cvetkovic, Powers, & Cosic, 2009), resulting in a resonance and subsequent synchronization of one of the oscillators' fundamental frequency on a continuum of weak to strong coupling, either in-phase or out-of-phase (Clayton et al., 2005).
4. Pink Noise: sound comprising all frequencies; similar to white noise but contains less energy at treble frequencies and is more suitable for human hearing (Winer, 2012).
5. Theta Rhythm: 4-7 Hz firing pattern seen in the human EEG when a person is in a state of deep relaxation, meditation, or hypnagogia (Schacter, 1977).
6. Affect: "The mental representation of bodily sensations that are sometimes (but not always) experienced as feelings of hedonic pleasure and displeasure with some degree of arousal" (Lindquist et al., 2012).

Delimitations

1. Participant recruitment for this study took place at Ithaca College. For practical reasons the sample was restricted to low-risk, unmedicated, non-smokers between the ages of 18-34 years with no history of chemical dependencies, epilepsy, or metabolic/psychiatric diagnoses.
2. Only theta-frequency binaural beats were utilized in this study.
3. Binaural beats were presented to the listener for 20-min at a standardized volume.
4. Psychological dependent variables were restricted to positive affect, negative affect, self-reported stress and relaxation.
5. Physiological dependent variables were restricted to salivary α-amylase, blood pressure, heart rate and heart rate variability measures.
6. Each dependent variable was measured at Time 1 (baseline), Time 2 (post-exercise), Time 3 (post-relaxation) and Time 4 (post-stressor). Self-reported stress and relaxation were only evaluated at the end of Time 4 in each session.
7. The exercise protocol was administered on a treadmill and was calculated individually based upon 70% of predicted maximum volume of oxygen consumption (VO_{2max}) (ACSM's guidelines for moderate cardiovascular exercise) (Bradshaw et al., 2005; Thompson, 2010).
8. The relaxation response was elicited while the participant is reclining in a comfortable padded recliner in a quiet low-light environment.
9. The laboratory stressor involved listening to a prerecorded audio track of 20 mental arithmetic problems comprised of both addition and subtraction components

presented over a 4-sec period with a 2-sec response interval. This protocol has been shown to be efficacious in eliciting a stress response (Sharpley & Gordon, 1999).

Limitations

1. Results of this study may not be generalizable to people outside the targeted age range, population demographic, or to those with medical conditions.
2. Results of this study may not be generalizable to other binaural beat stimuli.
3. Results of this study may not be generalizable to conditions where participants are exposed to binaural beats for greater or less than 20-min or at different volume levels.
4. Results measured by the PANAS and self-report might not be generalizable to other measures of psychological distress.
5. Physiological findings may not be generalizable to other physiological variables.
6. Results may not be generalizable to times outside of the experimental time-frame.
7. Results may not be generalizable to other exercise modalities or training intensities.
8. Results of this study might only apply to the post-exercise relaxation response and not to a relaxation response elicited without exercise. Also, results might not be generalizable to other environments.
9. Effects of relaxation on the laboratory stressor may not be generalizable to 'real life stressors' or to populations outside of the delimited sample. Any improved stress resiliency effects demonstrated may not be generalizable to times outside of the experimental time-frame.

CHAPTER 2

REVIEW OF LITERATURE

The purpose of this study was to examine the effectiveness of theta-frequency (4-7 Hz) binaural beats in the facilitation of the post-exercise relaxation response and attenuation of a subsequent stress response. This chapter is devoted to providing relevant background literature to inform this study. In this review of literature, the following topics are discussed: (a) relevant neuroanatomy and neurophysiology, (b) overview of clinical binaural beat research, (c) autonomic arousal: the stress and relaxation responses, (d) consciousness, attention and trance, (e) exercise and (f) summary.

Relevant Neuroanatomy and Neurophysiology

Brainstem anatomy. The human brainstem lays inferior to the thalamus, the brain's sensory integration center and is comprised of the medulla oblongata, pons, midbrain, reticular formation, superior olivary complex and the superior and inferior colliculi (Purves et al., 2008b). The structures of the brainstem are involved in basic life-maintaining functions such as reflexes, respiration, cardiovascular and vasomotor control and regulating levels of consciousness (Purves et al., 2008b). Most relevant to the present study are the nuclei that comprise the superior olivary complex, the inferior colliculi and the 'net-like' reticular formation. The superior olivary complex marks the location where auditory information is integrated from each ear, allowing for initial sound localization in space (Purves et al., 2008c). The inferior collicular nuclei are responsible for generating the orienting reflex through which the body responds to novel sights, sounds and other sensations (Seeley, Stephens, & Tate, 2006). The reticular

formation's nuclei are scattered throughout the brainstem and are responsible for regulating attention and the sleep-wake cycle (Purves et al., 2008b).

The human auditory system and music. Sound stimuli consist of mechanical pressure waves that are tranduced by the auditory system into neural signals and subsequently into auditory perceptions (Purves et al., 2008c). For sound perception to occur, the pressure variations of a sound wave must fall within a frequency range of 20 Hz to 20,000 Hz (Purves et al., 2008c). Basic perceptual qualities of sound include loudness (sound intensity), pitch (perception of sound wave frequency on a tonal continuum) and timbre (subjective quality of sound derived from the shape and contours of the sound source) (Purves et al., 2008c). Music consists of complex periodic sound stimuli (beats) characterized by all basic perceptual qualities of sound and additionally by rhythm (referring to the accentuated beats in music), tempo (rate of beat presentation per minute) and meter (number of beats per measure) (Purves et al., 2008c).

Central neural processing of sound stimuli begins in the cochlear nucleus and continues via axonal projections to the superior olivary complex, the inferior colliculus and the nuclei of the lateral lemniscus (Purves et al., 2008c). From this point, all auditory information is routed to the thalamus, the brain's sensory integration center and then sent out to the cerebrum's primary auditory cortices (Purves et al., 2008c).

Overview of the electroencephalogram. The electroencephalogram (EEG) serves as a non-invasive, relatively inexpensive tool for measuring neurological electrical activity through the scalp (Cvetkovic et al., 2009). The EEG signal is comprised of "several time-series components with different dominant frequency ranges generated from different sites in the brain" (Jin et al., 2002). These dominant frequency ranges are

categorized into beta (13-30 Hz), alpha (8-13 Hz), theta (4-7 Hz) and delta (1-4 Hz) brain-waves (Jin et al., 2002) and are thought to represent the collective firing of large numbers of neurons (Cvetkovic et al., 2009). It is currently believed that rhythmic EEG signals are derivative of summated postsynaptic potentials in the cortex and evidence suggests regulation by the extended reticular thalamic activating system (ERTAS) (Atwater, 2001; Cvetkovic, 2005; Purves et al., 2008a). Specifically, cholinergic, noradrenergic and serotonergic neurons in the ERTAS modulate levels of consciousness along a continuum from deep sleep to peak performance (Pop-Jordanova, 2011; Purves et al., 2008a).

Overview of entrainment. Broadly, entrainment is a process through which two autonomous rhythmic oscillators with similar but different fundamental frequencies interact (Cvetkovic et al., 2009). This interaction results in a resonance and subsequent synchronization of one of the oscillators' fundamental frequencies on a continuum of weak to strong coupling, either in-phase or out-of-phase (Clayton et al., 2005). Classic examples of entrainment include the synchronizing of human sleep-wake cycles to the 24-hour cycle of light and dark (Clayton et al., 2005), the synchronization of a heartbeat to a cardiac pacemaker (Cvetkovic et al., 2009) and the use of rhythmic auditory stimulation in the rehabilitation of motor functions (Thaut & Abiru, 2010).

Measurement of auditory entrainment via the EEG has been demonstrated by exposing individuals to monaural (acoustic, one ear), diotic (acoustic, two ears, same frequency) and dichotically (binaural, two ears, different frequencies) presented pure tones (Draganova et al., 2008; Karino et al., 2006; Kennerly, 2004; Pratt et al., 2009; Schwarz & Taylor, 2005). Entrainment of the EEG is measured objectively through a

phenomenon called an evoked-auditory-steady-state response (ASSR) (Schwarz & Taylor, 2005). Research in cats has shown binaural beat induced ASSRs in the reticular formation, a finding which lends support to the claim that binaural beats provide information to the ERTAS (Faingold & Caspary, 1979).

While there is a large body of literature that discusses auditory entrainment, little effort is made to differentiate between tempo-entrainment, frequency-entrainment and entrainment resulting from other musical properties. Music is characterized by a variety of elements including rhythm, pitch, timbre, loudness, tempo and melody, all which theoretically might be capable of providing for entrainment effects (Cvetkovic, 2011).

For example, shamanic drumming is usually performed at a tempo of 3-5 beats per second (180 to 300 beats per minute), which roughly corresponds to the delta brainwave frequency band (Jovanov, 2011). This type of music is commonly utilized to facilitate entry into altered states of consciousness, although it is still unclear what the causal mechanism might be. Moreover, there is no literature that discusses the potential differential entrainment elicited through low-frequency (low pitch) 180 bpm drumming (e.g., a bass drum) and high-frequency (high pitch) drumming (e.g., an alto drum). Theoretically, an ASC could be evoked by tempo alone or by combinations of rhythm, pitch, timbre, loudness, tempo and/or melody.

Binaural beat entrainment. Binaural beats are an auditory illusion experienced subjectively (as an illusory beat) and measured objectively (via evoked ASSRs), when two or more pure-tone sine waves of similar but different frequencies (under 1500 Hz and less than 40 Hz apart) are presented dichotically (Draganova et al., 2008). For example, if a 510 Hz pure tone was presented to a listener's right ear while a 500 Hz pure tone was

presented to the listener's left ear, via stereo headphones, the listener would perceive an illusory binaural beat with a frequency (perceived tempo) of 10 Hz. An interaural frequency difference, under normal circumstances, would provide directional localizational information to the listener (Atwater, 2004). However, when variant tones are presented through stereo headphones, neural spikes in the cochlea become phase-locked to the interaural frequency difference and this information is transmitted from the auditory nerve fibers to the medial superior olivary nuclei, the reticular formation, the inferior colliculus neurons and ultimately to the primary auditory cortices (Karino et al., 2006).

Binaural beat perception originates in the brainstem's superior olivary nuclei, where sound signals from each ear are integrated and continues as the neural impulses travel through the net-like reticular formation up the midbrain to the thalamus, auditory cortices and other cortical regions (Atwater, 2001). In theory, once entrainment has occurred at the level of the brainstem, the evoked ASSRs provide information to the thalamus which then acts to regulate cortical arousal accordingly by altering the activity of cholinergic thalamocortical neurons (Atwater, 2001). Since the ERTAS interprets and reacts to a variety of external and internal stimuli such as light levels, ambient noise, attentional focus, emotions, cognitions and present state of psychophysiological arousal, each of these factors must be conducive to the desired state of consciousness one is attempting to entrain (Atwater, 2004). Mental set and physical setting are both essential to achieving a desired performance (Leary & Metzner, 1995).

Binaural beat ASSRs differ qualitatively from acoustic beat ASSRs and their perception and temporal encoding are influenced by the listener's focus of attention

(Schwarz & Taylor, 2005). Neuromagnetic and EEG recordings have demonstrated that binaural beat ASSRs are characterized by smaller amplitudes and a much fainter perception than acoustic ASSRs (Karino et al., 2006). Other research examining the effects of theta-frequency binaural beats on evoked neuromagnetic fields demonstrated an entrainment of human cerebral cortex function that "could reflect both a cognitive process, which is influenced by attention and an unconscious transporting process of interaural phase difference information, which is not affected by attention" (Karino et al., 2006). More recent research recorded low-frequency binaural beat ASSRs from the scalp, providing further evidence that the locations of ASSRs and thus the cortical structures involved, are a function of the beat frequency presented (Pratt et al., 2009). This supports the claim that presenting different beat frequency ranges might elicit differential states of consciousness.

Given that human cortical EEG oscillations range from zero to around 40 Hz and humans have an auditory range of 20 to 20,000 Hz, some claim that entrainment of human brain-waves to acoustic beats cannot occur below 20 Hz (Cvetkovic, 2005). This conflicts with evidence of tempo-entrainment to indigenous music (Neher, 1961, 1962). Clearly, it is important to differentiate between tempo-entrainment and frequency-entrainment which may or may not share similar neurological mechanisms. Frequency-entrainment to acoustic beats cannot occur below 20 Hz since this is below the auditory threshold; however through the utilization of binaural beats, entrainment of alpha, theta and delta brain-wave frequencies may be possible. Since binaural beat perception results from the interaural frequency difference between two or more pure tones presented above the audible threshold, the temporal encoding at the level of the brainstem provides a

mechanism through which slow cortical rhythms (i.e., alpha, theta and delta) may be entrained. Quantitative electroencephalography (QEEG; high-resolution topographic brain-mapping of EEG data) research has shown that theta- and delta-frequency binaural beats can cause entrainment of cortical rhythms in as little as five minutes (Kennerly, 2004).

Overview of Clinical Binaural Beat Research

A recent literature review of brain entrainment research revealed several studies which have examined the effectiveness of binaural beat entrainment on heart rate, blood pressure, electrodermal response, finger temperature (Kennerly, 2004), performance vigilance and mood (Lane et al., 1998), hypnotic susceptibility (Brady & Stevens, 2000), mental and physical relaxation (Foster, 1990), attention and memory recall (Kennerly, 1994), depression and mood regulation (Cantor & Stevens, 2009), anxiety and burnout (Le Scouarnec et al., 2001), as well as on pre-operative anxiety and intra-operative anaesthesia requirements (Dabu-Bondoc et al., 2003; Dabu-Bondoc et al., 2010; Kliempt et al., 1999; Lewis, Osborn, & Roth, 2004; Padmanabhan, Hildreth, & Lewis, 2005).

These studies utilized five distinct categories of binaural beat variables: Hemi-Sync® Surgical Support Series (combination of verbal guidance, instrumental music and complex-multilayered binaural beat patterns), Holosync Solution® (verbal guidance, instrumental music and delta-frequency binaural beats), instrumental music combined with complex-multilayered binaural beat patterns, simple pure-tone binaural beats combined with ‘pink-noise’ (sound equalized for human hearing that covers the entire audible frequency range) and complex-multilayered binaural beats with pink noise.

Only one of these studies failed to demonstrate a significant effect of binaural beats on measured variables (Dabu-Bondoc et al., 2003) and the conclusions of this study were reversed after the completion of a follow-up study several years later (Dabu-Bondoc et al., 2010). Delta-frequency beats with music (Le Scouarnec et al., 2001) and verbal guidance (Padmanabhan et al., 2005), were shown to have potent anxiolytic effects, as measured through the STAI-S and self-reports. Of the anaesthesia studies, the general finding was that Hemi-Sync® could decrease pre-operative anxiety and reduce intra-operative fentanyl requirements, a powerful anaesthetic, by as much as 78% (Lewis et al., 2004).

Most relevant to the present study were the designs that utilized complex-multilayered binaural beats with pink noise. To truly ascertain the clinical effectiveness of binaural beats they must be experimentally isolated from possible confounding variables such as verbal guidance and instrumental music. Both studies which did this found significant effects. A well-controlled, double-blinded study with a fairly large sample size (n = 29) demonstrated that complex-multilayered beta-frequency binaural beats combined with pink noise significantly ($p < .001$) improved performance on a continuous 30-min vigilance performance task (Lane et al., 1998). This finding is consistent with the ERTAS arousal-modulating hypothesis. Unfortunately, the effect size was not reported. The second study (n = 6) tested the effects of complex-multilayered theta-frequency binaural beats combined with pink noise on hypnotic susceptibility (Brady & Stevens, 2000). They found statistically significant improvements ($p < .05$) in hypnotic susceptibility and significant ($p < .01$) increases in EEG theta power for 83% (5 out of 6) of participants (Brady & Stevens, 2000). This provides some evidence that the

binaural beat audio tapes utilized in this study are efficacious in increasing EEG theta power. For this reason, these tapes will also be used in the present study to facilitate the post-exercise relaxation response.

Autonomic Arousal: Stress and Relaxation Responses

To a large extent, the cardiovascular system is controlled dynamically by the sympathetic (SNS) and parasympathetic (PNS) branches of the autonomic nervous system (Smith & Fernhall, 2010). Simply put, the stress response and the relaxation response are polar autonomic nervous system responses (Benson, 1983). However, unlike the stress response, the relaxation response can be voluntarily elicited (Dusek & Benson, 2009). Each will now be discussed in turn along with related topics (e.g., heart rate variability, salivary α-amylase and hypnosis).

The stress response. The stress response is primarily mediated by the sympatho-adreno-medullary (SAM) and the hypothalamic-pituitary-adrenal (HPA) axes (Dusek & Benson, 2009). Corticotrophin-releasing hormone, secreted by the hypothalamus, stimulates the pituitary gland to secrete adrenocorticotropic hormone (ACTH) which activates both the SAM and HPA axes. The SAM axis responds quickly to circulating ACTH, resulting in the secretion of catecholamines (epinephrine and norepinephrine) from the adrenal medulla (Dusek & Benson, 2009). The HPA axis, on the other hand, responds more slowly, ultimately resulting in the secretion of cortisol from the adrenal cortex (Dusek & Benson, 2009). Circulating catecholamines significantly decrease heart rate variability (HRV) and have a direct effect of elevating heart rate, respiration, oxygen consumption and blood pressure (Motzer & Hertig, 2004). Therefore, to evaluate the stress response, SAM axis activity will be a focus of this study.

Heart rate variability. HRV measures serve as indices of the beat-to-beat variability of the cardiac R-R interval, resulting from the interaction of SNS and PNS innervation of the sinoatrial (SA) node, which controls heart rhythm (Motzer & Hertig, 2004). HRV data can be measured over short-term (e.g., 2-5 min) and long-term recordings (e.g., > 24 hours) (Camm, A.J., Malik, M., Bigger, J.T., Breithardt, G., Cerutti, S., Cohen, R.J., et al., 1996) and data can be analyzed by both time-domain and frequency-domain methods (Terathongkum & Pickler, 2004). However, it is inappropriate to compare HRV recordings of different durations considering that recording length influences HRV results (Camm et al., 1996). For short-term recordings, 5-min has been suggested as standardized recording length and at least 2-min are needed to fully address low-frequency components of the signal (Camm et al., 1996).

Time-domain measures. Time-domain data are more easily measured than frequency-domain data and are determined statistically based upon the mean R-R interval and variations of interval standard deviations over time (Terathongkum & Pickler, 2004). These data are particularly suited to long-term recordings (Task-Force, 1996). Common time-domain variables are described below and in Table 2.1. These variables all describe high-frequency oscillations of the heart rate signal (Task-Force, 1996). Along these lines, it is important to note that R-R interval length is a function of PNS (vagal) activity and therefore decreases with stress and increases with relaxation (Motzer & Hertig, 2004). Evidence for this claim has been provided in recent meditation and relaxation studies (Sakakibara, Takeuchi, & Hayano, 1994; Tang et al., 2009; Terathongkum & Pickler, 2004). Further, in other studies, more HRV at baseline was associated with

lower blood pressure and decreased risk of sudden death or cardiac arrest (Task-Force, 1996).

Table 2.1

Common Heart Rate Variability Time-Domain Variables (Task-Force, 1996).

Variable	Units	Description
SDNN	ms	Standard deviation of all NN (normalized R-R) intervals.
RMSSD	ms	The square root of the mean of the sum of the squares of differences between adjacent NN intervals.
NN50	----	Number of pairs of adjacent NN intervals differing by more than 50 ms in the entire recording.

Note. SDNN represents the standard deviation of R-R intervals "normalized for the standard deviation of the mean ventricular cycle length" (John Camm et al., 1999).

Frequency-domain measures. Frequency-domain HRV data are assessed through spectral analysis of the electrocardiogram's QRS complex (typically through using a Fast-Fourier Transformation algorithm or auto-regressive techniques) (Murai, Stone, & Hayashi, 2008). The HRV frequency spectrum is comprised of very-low-frequency (VLF), low-frequency (LF) and high-frequency (HF) components (Murai et al., 2008). VLF signal components are difficult to interpret from short-term recordings and are typically removed from the sample during data analysis (Tarvainen & Niskanen, 2008; Task-Force, 1996). Absolute power values (ms^2) of VLF, LF and HF components are conventionally reported in normalized units (n.u.) which "represent the relative value of each power component in proportion to the total power minus the VLF component" (Task-Force, 1996).

Both LF and HF power are measured from the R-R interval, with the LF value being calculated from the 0.04 to 0.15 Hz range, reflecting SNS and PNS activity at the

SA node and the HF value being calculated from the 0.15 to 0.40 Hz range reflecting PNS activity only (Murai et al., 2008). This frequency range (0.15 to 0.40 Hz) is also called the respiratory sinus arrhythmia (RSA) and is heavily influenced by breathing rate and PNS activity (Tarvainen & Niskanen, 2008). A high LF/HF ratio is indicative of elevated SNS activity and a low LF/HF ratio indicates PNS dominance (Murai et al., 2008). Thus, LF/HF power ratio provides a reliable non-invasive estimate of sympathovagal balance (Smith & Fernhall, 2010).

Artifact removal and pre-processing of the HRV signal. HRV data can be significantly impacted by the inclusion of artifacts generated by physiological events such as ectopic beats or arrhythmias and also by noise resulting from movement, electromagnetic fields, or other sources of interference (Tarvainen & Niskanen, 2008). The general recommendations for signal pre-processing are to manually check the signal visually for artifacts and to only select artifact-free segments for analysis (Tarvainen & Niskanen, 2008). If a recorded signal contains over 2% artifacts, it should be completely omitted from analysis (Bricout, DeChenaud, & Favre-Juvin, 2010; Task-Force, 1996). Lastly, HRV signals are typically detrended via a smoothness priors based approach in order to control for slow linear trends and other nonstationarities that can make analysis problematic (Tarvainen & Niskanen, 2008). Simply stated, detrending the HRV signal removes excess 'noise' (i.e., low-frequency spectral components and nonstationarities) that improve the quality of the HRV signal (signal-noise ratio).

Salivary α-amylase. Salivary α-amylase (sAA) is produced by oral salivary glands and functions to digest carbohydrates and to inhibit the adherence and growth of bacteria (Rohleder & Nater, 2009). Salivary α-amylase accounts for approximately 50%

of salivary protein and most of (80%) the enzyme is synthesized in the parotid salivary gland (Nater & Rohleder, 2009). Research has shown positive correlations between sAA and plasma norepinephrine (r = 0.64) (Rohleder & Nater, 2009), STAI-S scores in response to mental arithmetic stress (r =0.59) (Noto et al., 2005) and the anaerobic threshold (r = 0.93) (Nater & Rohleder, 2009). Stress-induced sAA concentrations are independent of saliva flow rate (Rohleder, Wolf, Maldonado, & Kirschbaum, 2006).

Exercise research demonstrated that aerobic exercise can induce at least a 3-fold increase in sAA levels and this increase is intensity dependent (Chatterton et al., 1996). Further, post-exercise sAA levels can return to baseline levels within 30-45 minutes but can also remain elevated for as long as two hours post-exercise depending upon exercise intensity (Chatterton et al., 1996). There do not appear to be significant gender differences in basal sAA activity, however known confounding variables include cigarette smoking, excessive alcohol consumption, acute caffeine consumption, high carbohydrate consumption, somatic and psychiatric diseases and medical drugs such as anti-hypertensives, asthma medication and adrenergic agonists/antagonists (Rohleder & Nater, 2009). There are no data available on basal sAA differences between trained and untrained individuals, but utilizing a cross-over experimental design should control for any possible variance (Rohleder & Nater, 2009).

Recently a hand-held monitor has been developed which measures sAA using a dry-chemistry system (Yamaguchi et al., 2006). This device, commercially labeled the COCORO Meter™, can reliably measure sAA activity in less than two minutes with less than 5μl of saliva (Yamaguchi et al., 2006). Although wide variability in sAA levels exists between subjects, the calibration curve for the monitor has a high coefficient of

determination (R^2 = 0.97), meaning that it reliably assesses moment-to-moment fluctuations in an individual's sAA levels (Yamaguchi et al., 2006). Unfortunately, it does not appear that the COCORO Meter™ has been empirically validated against standard laboratory 'bench-top' measurement techniques for sAA.

Consciousness, Attention and Trance

Many modern theorists view consciousness as comprised of two broad components: awareness/attention (of self and/or environment) and arousal (alertness, vigilance, or wakefulness) (Cvetkovic, 2011; Purves et al., 2008a). In philosopher David Chalmer's view, states of consciousness are characterized by the distinct activity patterns of underlying neural correlates of consciousness (NCC), the subjective sensations associated with the state (e.g., alertness, fatigue, pleasantness/unpleasantness, *etc.*) and the present contents of consciousness (perceptions) (Pop-Jordanova, 2011). Brain rate, an EEG spectrum parameter that expresses the mean frequency of brainwave rhythms, serves as a valid global indicator of the arousal dimension of consciousness and provides a gross measure of NCC activity (level of consciousness) (Pop-Jordanova, 2011).

Attention, on the other hand, can be defined as "a set of mechanisms whereby the brain selects a subset of the incoming sensory information for higher level processing, while the non-attended portions of the input are analyzed at a lower bandwidth" (Tononi & Koch, 2008). Alternatively, attention can simply be defined as anything that alters the firing rate of a neuron (Barrett & Bliss-Moreau, 2009). Research has shown that as the arousal component of consciousness increases, attention moves from a diffuse state to a narrowly focused state and vice versa during trophotropic practices (Landers & Arent, 2010).

Other researchers argue that mammalian consciousness is foundationally affective, including a third component, hedonic valence, in the model of consciousness (Barrett & Bliss-Moreau, 2009; Panksepp & Watt, 2011). Lisa Feldman Barrett and Bliss-Moreau go so far as to hypothesize that affect is a human psychological primitive and that all human conscious experience is colored by affective responses (Barrett & Bliss-Moreau, 2009). According to this theory, core affect can be conceptualized along an *affective circumplex*, ranging along dimensions of arousal and hedonic valence (Figure 2.1) (Barrett & Bliss-Moreau, 2009).

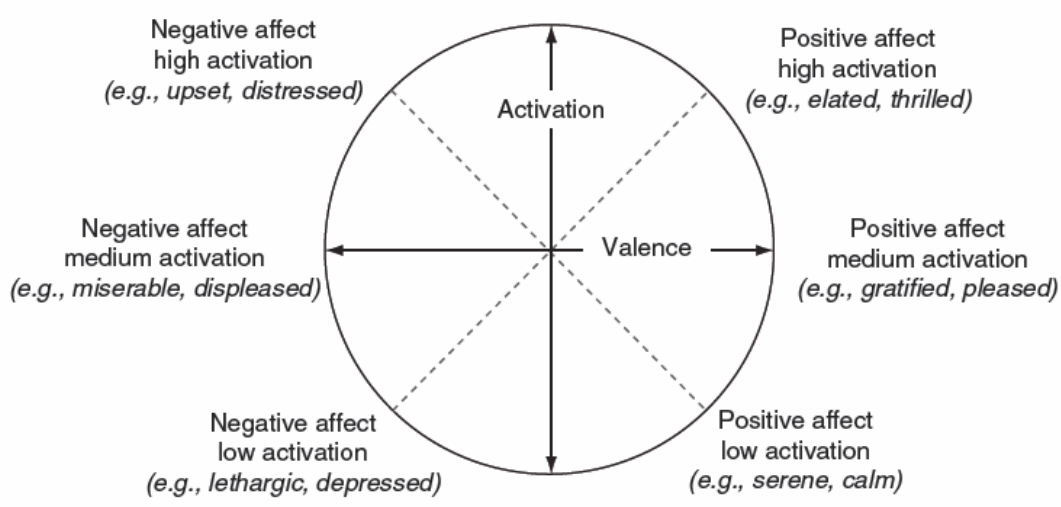

Figure 2.1. The affective circumplex (Barrett & Bliss-Moreau, 2009).

Trance theorist G. Rouget stressed the role of attention in trance-induction, pointing out that the individual must be willing to become absorbed in the trance state and must also have a specific aim as well as adequate intellectual preparation (Fachner & Rittner, 2011). For the purposes of this paper, trance can be used as a synonym for an altered state of consciousness (Fachner & Rittner, 2011). During trophotropic practices such as relaxation response training, meditation, or hypnosis, consciousness is altered

through shifting the awareness component internally, towards some mental device (e.g., breathing, counting, sound) while the arousal component is decreased through the deepening and regularization of breathing patterns, the relaxation of musculature and alteration of sympathovagal tone (Benson, 1982).

The potential for discrete trance states appears unlimited, due to indefinite possible combinations of arousal, awareness and environmental setting. However, EEG frequency bands have been shown to be highly correlated with some well-defined states of consciousness (Cvetkovic, 2005). For example, beta frequencies are associated with concentration, focused thinking, alertness and in the high range, anxiety (Huang & Charyton, 2008). Alpha waves are seen during light relaxation, or when the eyes are closed and when dominant have been described as a "state of electrical readiness which exists when the subject is awake and conscious but inattentive" (Foster, 1990). Early stages of sleep are marked by high-theta and low-alpha frequency 'sleep spindles' (Steriade, McCormick, & Sejnowski, 1993), theta waves are seen in deep relaxation, meditation and hypnagogia (Schacter, 1977) and delta waves are indicative of deep, dreamless sleep states (Salansky, Fedotchev, & Bondar, 1998).

The relaxation response. Herbert Benson's investigations into the stress response led him to identify common patterns underlying the altered states of consciousness elicited through various meditative practices, ritual technologies and secular practices such as hypnosis (Benson, 1982). He found that all trophotropic (arousal decreasing) practices had a common foundation: the elicitation of the relaxation response (Benson, 1982). The relaxation response is an innate physiological response characterized by diminished sympathetic nervous system activity, decreased heart rate,

blood pressure and increased theta brainwave activity (Benson et al., 1981). For the relaxation response to be elicited, the practitioner must sit quietly in a comfortable position with eyes closed, deeply relax the musculature, breathe deeply and regularly, maintain a passive attitude and focus their attention on a mental device (e.g., a mantra, an image, breathing, music)(Benson, 1982).

The relaxation response is believed to be mediated by the hypothalamus and is characterized by decreased oxygen consumption (with no change in respiratory quotient), arterial blood lactate, muscle tone, heart rate, as well as increased alpha and theta wave EEG activity (Benson, 1982). Benson calls the relaxation response "a self-induced altered state of consciousness" (Benson, 1983). Research indicates that regular elicitation of the relaxation response can mitigate the stress response and effectively treat a variety of musculoskeletal disorders, gastrointestinal disorders, cardiovascular disorders, insomnia and can also reduce hostility and anxiety (Jacobs, 2001).

Hypnosis. The relaxation response and hypnosis are functionally distinct phenomena but both are associated with alterations in autonomic cardiac tone (Aubert et al., 2009). Hypnosis is defined as "a natural state of aroused, attentive focal concentration coupled with a relative suspension of peripheral awareness" (Astin, Shapiro, Eisenberg, & Forys, 2003). The altered state of hypnosis is characterized by a deep absorption in an object of concentration, dissociation from external awareness and a heightened level of suggestibility absent cognitive censor or criticism (Astin et al., 2003). It has been suggested that while the relaxation response and hypnosis are distinct, variability in clinical relaxation response effectiveness may be a function of trait hypnotic susceptibility (Benson et al., 1981).

A correlation between hypnotic ability and therapeutic outcome in the treatment of psychosomatic disorders has been reported (Brady & Stevens, 2000). Hypnotizability is usually divided into three levels: high, medium and low, with the medium level being most amenable to modification of ability through training (Brady & Stevens, 2000). Training methodologies have employed strategies such as sensory deprivation, biofeedback, binaural beats and psychotomimetic drugs to improve hypnotizability (Brady & Stevens, 2000).

Theta-frequency binaural beats appear to be a non-invasive, inexpensive and efficacious tool to increase hypnotizability and thus the depth of the relaxation response. This will increase the likelihood that an individual can reap maximum therapeutic benefit of the relaxation response, such as decreased blood pressure, reduction of the frequency of premature ventricular contractions, decreased baseline sympathetic tone and increased measures of health and well-being (Benson, 1982).

Exercise

Exercise, albeit a physiological stressor, has well-established acute anxiolytic effects (Raglin & Morgan, 1987), as well as the potential for improving long-term stress-resiliency (Salmon, 2001). In healthy populations, cardiovascular exercise has fairly predictable, intensity-dependent effects. For example, exercise induces decreases in blood pressure (Raglin & Morgan, 1987), decreased post-exercise blood pressure responses to laboratory stressors (Santaella et al., 2006), heart rate (McGowan, 1985), electromyography (EMG) activity (McGowan, 1985), electrodermal skin conductance (Boettger et al., 2010) and acute decreases in HRV (indicative of very-low PNS vagal control) (Boettger et al., 2010), followed by training-induced increases in basal HRV

(Sandercock, Bromley, & Brodie, 2005). Both relaxation response training and cardiovascular exercise training appear to improve stress resiliency and result in long-term improvements in basal PNS cardiac control (Boettger et al., 2010).

One study examined pre- and post-exercise EEG alpha power and found significant increases in alpha power for exercisers, which correlated with reductions in self-reported anxiety (Boutcher & Landers, 1988). Another study demonstrated similar findings, showing exercise induced increases in both theta and alpha activity and corresponding reductions in beta power (Kubitz & Pothakos, 1997). The exercise induced changes reported in these studies persisted for 20-min post-exercise but no additional follow-up recordings were provided (Boutcher & Landers, 1988; Kubitz & Pothakos, 1997). Additionally, another study found a correlation between pre-exercise EEG asymmetry and trait-anxiety, as well as a correlation between post-exercise anxiety reductions and EEG hemispheric symmetry (Petruzzello & Landers, 1994a).

Recent research has examined the effects of exercise and shavasana, a yogic technique, on blood pressure and post-exercise stress response to a laboratory stressor (Santaella et al., 2006). Their findings showed that exercise and shavasana combined had the greatest effect of reducing blood pressure post-exercise and blunting the blood pressure response to a laboratory stressor. It seems that exercise training followed by secular relaxation training may provide for a deeper relaxation response than either intervention alone. This combination may provide for increased alpha and theta brain-wave activity, as well as increased therapeutic benefit.

Summary

Binaural beats are an auditory illusion experienced by the listener as a result of the structural and functional organization of the human auditory system and central nervous system. Research has demonstrated evidence supporting the 'auditory-driving' hypothesis, the idea that music and sound can alter neuronal firing rates through the process of tempo-entrainment. Since human consciousness can be considered as having *at least* two broad dimensions, awareness/attention and arousal, it follows that any process of entrainment (e.g., tempo-entrainment, frequency-entrainment, circadian-entrainment, rapport, dance, etc.) will likely affect consciousness in unique ways ranging from very subtle to very dramatic (e.g., experience of 'exotic' states of consciousness such as jet lag, hypnotic trance, lucid dreaming, deep meditation, peak performance, etc.). Frequency-entrainment and tempo-entrainment are commonly confused in the literature and very few existing studies have attempted to disentangle potential confounders in an effort to uncover casual mechanisms and psychophysiological correlates.

If frequency-entrainment works, then theta-frequency binaural beat based auditory-driving should serve to facilitate a trophotropic response which would be easily measured through HRV indices, blood pressure, sAA and state affect. By first inducing an ergotropic response via moderate cardiovascular exercise, it seems plausible to assume that any subsequent trophotropic shifts resulting from auditory-driving will be more apparent and potentially more robust.

CHAPTER 3

METHODS

The following chapter outlines the experimental design for this study. Sections on participants, laboratory equipment, psychometric measures, experimental procedures, data processing and data analysis are presented. All procedures were approved in advance by Ithaca College's Human Subjects Review Board.

Participants

Twenty-two volunteers were recruited by announcement from Ithaca College classes wherein the researcher read aloud a recruitment script (Appendix A) and provided a sign-up sheet. Interested participants were emailed instructions for the study along with an informed consent document, Pre-Participation Screening Questionnaire (PPSQ), Physical Activity Readiness (PA-R) and Perceived Functional Ability forms (PFA) (Appendices B – E respectively). Participants completed the informed consent document prior to their participation but remained naïve to the true nature of the experiment. They were told only that we were studying the effects of music on the post-exercise relaxation response. A full debriefing followed completion of the final experimental session (Appendix F).

One subject failed to complete both sessions due to medical reasons so his data were excluded from all analyses. The remaining sample (n = 21) included 14 males and seven females, with 76.2% (16 out of 21) "White/Caucasian", 9.5% (2 out of 21) "Black/African American" and 4.8% (1 out of 21) "Asian" and "Hispanic/Latino(a)". One participant chose not to report ethnicity. Age ranged from 18 to 29 years (M = 20.33, SD = 2.69). Baseline descriptive statistics for the sample are found in Table 3.1.

Table 3.1

Descriptive Statistics for Participants' Baseline Data.

	Mean (*M*)	Standard Deviation (*SD*)
Height (cm)	171.12	10.24
Weight (kg)	77.41	15.31
Body Mass Index	26.10	4.10
VO_2MAX (mL/kg/min)	48.30	6.50
ExRx-MPH	5.80	0.80

Note. VO_2MAX = maximal *predicted* volume of oxygen consumption; ExRx-MPH = Exercise prescription in miles per hour.

Participants were risk stratified through completion of a pre-participation screening questionnaire (Appendix C). Only low-risk participants, as defined by ACSM, were considered for this study (Thompson, 2010). Anyone with a history of diagnosed mental or physical illness which may have confounded the study (e.g., hearing loss, auditory disorders, diabetes, cardiovascular disease, hypertension, substance dependency, depression, ADHD, bipolar disorder, epilepsy) were excluded. Further, all participants were screened for habitual smoking, chronic alcohol usage, prescription medication usage and caffeine dependency, as these conditions can unpredictably influence basal and stress-induced sAA (Rohleder & Nater, 2009).

Equipment and Measures

Cardiovascular measures and analyses. A Polar RS800CX heart rate monitor (Polar Electro Oy, Kempele, Finland) was chosen because it provides a reliable non-invasive measure of both heart rate and HRV (Gamelin, Berthoin, & Bosquet, 2006; Nunan et al., 2009; Wallén et al., 2011). These measures are both important indicators of relaxation (Peng et al., 2004; Sakakibara et al., 1994) and sympathovagal tone (Sharpley

et al., 2000). The Polar HR monitoring system included a wireless chest-strap transmitter and a wrist-watch receiver.

Participants' heart rate signal was recorded at a one-second recording frequency and R-R interval data was sampled at a 1000 Hz sampling frequency, allowing for a one-millisecond temporal resolution of HRV (Polar RS800CX User Manual, 2011). HR data for each participant was then uploaded from the Polar wrist-watch to the Polar ProTrainer 5 software (Version 5.40.172) which was used to store data, check for artifacts and export R-R interval data.

A mercury sphygmomanometer was used to measure blood pressure. A Precor 956 treadmill (Woodinville, WA) was used to exercise the participants because treadmill exercise elicits a strong sympathetic nervous system response without requiring a metronome to keep a steady pacing.

Enzymatic analysis. A COCORO meter™ (NIPRO Corporation, Osaka, Japan) was used in this study because this device has been shown to be highly sensitive to sympathetic nervous system activity with a minimal (5μl) amount of saliva (Yamaguchi et al., 2004) independent of salivary flow rate (Rohleder et al., 2006). Subsequent research has demonstrated a calibration curve with an excellent coefficient of determination (R^2 = 0.97) for a wide range of sAA activity (Yamaguchi et al., 2006). Additionally, further research has demonstrated rapid declines in sAA activity with relaxation (Murai et al., 2008).

Acoustic equipment. A sound level meter (model 33-2050) (Radio Shack, Tandy Corporation, Fort Worth, Texas) was used with a C-weighted slow-response setting to calibrate a pair of Koss HQ1 collapsible full size headphones (Milwaukee, WI)

and to standardize volume levels. This setting was indicated by the instruction manual as the appropriate setting for sound presented uniformly over a range of 32-10,000 Hz for an extended period of time (Tandy, 1993). Since no coupler was available, volume level was measured by placing the recording end of the sound level meter directly against the center of the headphone earpiece. Volume levels for both left and right earpieces and for both control and experimental audio tracks, ranged from 61-63 decibels (dB).

Participants listened to 20-min of audio at a constant volume (61 – 63 dB) on a Sansa c250 2 GB MP3 player (SanDisk Corporation, Milpitas, CA) during each experimental session. During the control session participants listened to pink noise (sound equalized for human hearing that covers the entire audible frequency range (Monroe, 1993)) with carrier tones. The pink noise used in this study sounds similar to ocean surf sounds. During the experimental session participants listened to wide-band complex-multilayered theta-frequency binaural beat patterns overlaying pink noise **(**The Monroe Institute, Faber, VA). The binaural beat tracks used in the present study were created for the David Brian Brady research project (Brady & Stevens, 2000) with the intention of increasing theta-band EEG power and hypnotic susceptibility. The audio tracks were designed to be perceptually indistinguishable from one another in naïve listeners. Both audio tracks (i.e., pink noise and binaural beats overlaying pink noise) were provided by The Monroe Institute, Faber Virginia (Atwater, 1996) (Appendix G).

Pre- and post- screening questionnaires. Participants were screened for eligibility to participate and to prescribe exercise based on calculated maximal volume of oxygen consumption (VO_{2max}).

Pre-participation screening questionnaire (PPSQ). The PPSQ stratifies cardiovascular risk for participating in physical activity (Appendix C). The questionnaire assesses both personal and family health history. After completion, the participant is classified at high, moderate, or low risk (Thompson, 2010). The PPSQ used in this study was a modified version also used to assess usage of drugs, alcohol and medication (Bradshaw, 2003).

Physical activity rating (PA-R). The PA-R asks the participant to select a number, on a scale of 0-10, that best describes their overall level of physical activity for the previous six months (Bradshaw, 2003) (Appendix D). The scale ranges from avoidance of exertion (zero) to vigorous activity (running over 25 miles per week or eight hours doing comparable physical activity). The PA-R score is included in the non-exercise regression model used to predict VO_2max scores (Bradshaw et al., 2005).

Perceived functional ability questionnaire (PFA). First, the PFA asks participants to select the appropriate number, on a scale of 1-13, that corresponds to the speed they could comfortably cover a distance of one mile without becoming short of breath (Bradshaw, 2003)(Appendix E). Second, participants are asked to circle another number, on a scale which corresponds to their ability to cover a distance of three miles without becoming short of breath (Bradshaw, 2003).

Post-participation screening questionnaire (Post-PSQ). The Post-PSQ was used to gauge participants' familiarity with binaural beats, their meditative experience and their ability to discern differences between the experimental and control conditions (Appendix K). This was administered at the end of the study after completing both experimental conditions.

Psychometric instruments. Participants' levels of positive and negative affect were screened at each time point of the study. Rating of perceived exertion (RPE) was measured during exercise. At the end of each session, a stress response was induced using an empirically validated mental arithmetic protocol.

***Mental arithmetic battery*.** Participants were given 20 mental arithmetic (MA) problems via a pre-recorded audio track over a 2-min period and were required to write down their answers onto an answer sheet provided (Appendices H and I). Each problem comprised both addition and subtraction components and was presented over a 4-sec period with a 2-sec response interval. This MA protocol has been shown to be efficacious in eliciting the stress response (Sharpley & Gordon, 1999).

The positive and negative affect schedule (PANAS). The PANAS has both a trait (PANAS-T) and a state (PANAS-S) form, each comprising 20 questions that assess positive and negative affect on a five-point scale (Watson, Clark, & Tellegen, 1988) (Appendix J). The PANAS-T queries participants about how they have felt over the past week, while the PANAS-S asks how they are feeling at present. The Chronbach's alpha reliabilities for both the positive and negative affect scales are high, ranging from 0.84 to 0.90 (Watson et al., 1988). According to validation studies, the PANAS scales are valid, efficient and "provide reliable, precise and largely independent measures of positive and negative affect, regardless of the subject population studied or the time frame and response format used" (Watson et al., 1988).

Rate of perceived exertion scale (RPE). The RPE scale ranges from 6-20 (light exertion to vigorous exertion), which loosely corresponds to HR and is used to determine subjective exertion during exercise (Borg, 1982).

Procedures

Prior to each experimental session participants were provided with the following instructions via email:

1. Avoid strenuous exercise and do not consume any medication or alcohol 24 hours prior to the session.
2. Do not consume any caffeine or brush your teeth with toothpaste within three hours prior to the session.
3. Avoid ingesting any food or drink (besides water) for at least an hour before the study.
4. Eat a standard breakfast on the morning of the experiment and drink at least a cup of water 90 minutes before your session time.

To control for diurnal fluctuations of sAA, each participant was scheduled at the same time of day for each experimental session. Participants were alternatively assigned to A-B and B-A conditions as to control for order effects. The researcher conducting the experiment was blinded to condition (i.e., binaural beats or pink noise) during data collection and analysis, the MP3 tracks were labeled 'A' and 'B' by a third party.

On the day of their scheduled session, participants arrived individually at the Ithaca College Exercise Physiology Research Laboratory. The researcher synchronized the Polar HR monitor watch with a second wrist-watch and checked all equipment. A 24-hour history questionnaire (Appendix L) was administered to each participant to determine their eligibility to participate. Those who were unable to answer 'no' to all questions were excluded from participation (one subject due to medical reasons).

On participants' first visit, height and weight was measured. Each participant was given a six ounce glass of distilled water and was instructed to thoroughly rinse out their mouths, spit into the sink and then to finish drinking the remaining liquid. After applying

electrode gel, the researcher demonstrated how to attach the Polar HR monitor chest strap by “fitting it snugly around the chest, beneath the sternum, clear of any chest muscle tissue”. Participants were then given leave to use the bathroom, remove jewelry, get dressed and fit the Polar HR monitor chest strap.

Upon their return, participants were seated and instructed to complete the PA-R and PFA. After completion, they were told to sit quietly and relax for 5-min. During this 5-min period, the researcher calculated the workload for the day using an empirically validated non-exercise regression formula (Bradshaw et al., 2005). An exercise protocol was individually determined as per ACSM guidelines for moderate cardiovascular exercise (70% of predicted VO_{2max}) (Thompson, 2010). All measurements of sAA, blood pressure and heart rate were taken while subjects were seated quietly in an upright position.

At baseline (Time 1; T_1), the researcher started recording continuously with the Polar HR monitor and had participants complete the PANAS-T, provide a saliva sample for sAA measurement and recorded their blood pressure. Afterwards, participants began the exercise protocol on the treadmill. The exercise protocol began with a 5-min warm-up at 50% of the prescribed workload and then involved running for 20-min at the targeted heart rate intensity. During the exercise, at 5-min intervals, participants were asked to rate their RPE on a scale of 6-20 (Borg, 1982). Afterwards, participants cooled-down for another 5-min at 50% of the prescribed workload.

After completion of the exercise protocol (Time 2; T_2), participants completed the PANAS-S, provided another saliva sample for sAA measurement and had their blood pressure recorded. After completion of T_2 data collection, participants were given

another six ounce glass of distilled water to drink.

Next, participants were instructed to sit in the padded recliner (a comfortable padded egg-shaped conical chair with a foot rest) and to make themselves comfortable, sitting however they liked, but making sure to sit in the same way on the following visit. The lights were dimmed, a curtain was drawn around the recliner and the following short script was read:

> *"This relaxation period will last 20 minutes. I will close this curtain and leave the room so you can relax and have the space to yourself. I will return after 20 minutes and open the curtain and we will move back over to the other chair. Once you put on the headphones and begin listening to the music, I need you to close your eyes, focus attentively on the music and relax as deeply as you can. Do you have any questions?"*

Participants were then given a set of stereo headphones connected to the MP3 player and the appropriate track was started at a standardized volume level. The researcher then left the room, returning 20-min later to rouse the participant. After the relaxation session (Time 3; T_3), participants again took the PANAS-S, provided another saliva sample for sAA measurement and had their blood pressure recorded.

Next, participants were given standardized instructions to listen to the MA audio track and provide written answers to the 20 problems within a 2-sec time interval. After the test (Time 4; T_4), participants took a final PANAS-S, provided another saliva sample for sAA measurement and had their blood pressure recorded. Lastly, at the end of the session, participants were asked the following two questions:

1. *"On a scale of 1-10, with 10 being the most relaxed and one being the least*

relaxed, how relaxed did you feel during the relaxation session today?"

2. *"On a scale of 1-10, with 10 being the most stressed and one being the least stressed, how stressed did you feel during the math test today?"*

During the second scheduled session participants returned and completed the exact same protocol outlined above, the only differences being that they did not have their height and weight measured again and they received the alternate MP3 track. Participants were given a post-participation questionnaire (Post-PSQ) to assess their familiarity with binaural beats, experience with meditation and their ability to discern differences between the experimental and control conditions. After completion of the second experimental session, a full debriefing followed (Appendix F).

HRV Data Processing

All heart rate signal time-series were inspected for artifacts using Polar ProTrainer to ensure that no signal contained more than 2% artifacts (Bricout et al., 2010; Task-Force, 1996). Heart rate R-R interval data (time-series data from the peak of one QRS complex to the next) was then exported from the ProTrainer software into an ASCII text file and subsequently imported into Kubios HRV (Version 2.0) software for detailed analysis of HRV indices. As per the Kubios user's manual (Tarvainen & Niskanen, 2008), the R-R time series for all participants were detrended using a smoothness priors based detrending approach with $\lambda = 500$ (smoothing parameter with cut-off of 0.010 Hz times the sampling frequency), $f_c = 0.035$ Hz (estimated cut off frequency of the filter). Detrending removes slow-trend (Schmidt et al., 2010) and non-linear trend (Manimmanakorn et al., 2011) components which can cause distortion in the signal. Next, a conservative interpolation artifact correction algorithm was employed, using a

'strong' level of correction which excluded all obvious artifacts from analysis (Tarvainen & Niskanen, 2008).

Frequency-domain HRV indices were calculated using a Fast-Fourier Transformation (FFT) based Welch's Periodogram method (Tarvainen & Niskanen, 2008) with a 256 second window width and a 50% window overlap. A standard setting with a 4 Hz interpolation rate was used with the following frequency bands: very-low frequency (VLF, 0-0.04 Hz), low-frequency (LF, 0.04-0.15 Hz) and high-frequency (HF, 0.15-0.4) (Tarvainen & Niskanen, 2008).

After detrending and applying artifact correction, each signal was cut into four 2-min samples corresponding to sAA measurements, providing adequate duration to assess short-term spectral components (Task-Force, 1996). An additional 20-min sample was defined to correspond with the relaxation protocol.

Statistical Analyses

All statistical analyses were performed using PASW (Version 18; SPSS Inc., Chicago, IL, USA). Repeated measures analyses of variance (2x4 RM ANOVA; Condition x Time) were run for all dependent variables to assess Condition x Time interaction effects, main effects of Condition and main effects of Time. Mauchly's test was performed for all RM ANOVA to check for violations of the sphericity assumption. When Mauchly's test was found to be significant, Greenhouse-Geisser corrected degrees of freedom were reported (Field, 2009). When appropriate, the Sidak method for multiple comparisons was used for *post hoc* analyses (Field, 2009).

Further paired-samples *t*-tests were run to assess differences between conditions on each of the heart rate variability measures during the 20-min relaxation protocol and

when 2x4 ANOVAs approached significance. Effect sizes (Pearson's correlation coefficient, *r*) were calculated for all paired samples *t*-tests using the following formula: $r = \sqrt{((t^2)/(t^2 + df))}$ (Field, 2009). Effect sizes are useful to report because they provide an indication of magnitude of difference between two experimental conditions (Field, 2009). An alpha level of .05 was used for all statistical tests.

CHAPTER 4

RESULTS

In the present study the effects of wideband theta-frequency binaural beats on the post-exercise relaxation response and subsequent stress response, were assessed in a double-blinded, repeated measures, crossover-controlled experimental design. Dependent variables were assessed at four time points: baseline (T_1), post-exercise (T_2), post-relaxation (T_3) and post-stress (T_4) in both binaural beat and control conditions. A detailed summary of statistical results follows with data organized into subsections by dependent variable: state positive affect (PA), state negative affect (NA), self-reported relaxation (RELAX), self-reported stress (STRESS), salivary alpha-amylase (sAA), systolic blood pressure (SYS-BP), diastolic blood pressure (DIA-BP), heart rate (HR), average R-R interval (avRR), standard deviation of R-R intervals (SDNN), root mean square standard deviation of R-R intervals (RMSSD), frequency of N-N intervals separated by more than 50 ms (NN50), low-frequency power (LF), high-frequency power (HF) and low-frequency to high-frequency power ratio (LF/HF). Additionally, all of the HRV measures (HR, avRR, SDNN, RMSSD, NN50, LF, HF and LF/HF) during the 20-min relaxation protocol were compared between conditions and data for these variables are presented at the end of the chapter.

Psychological Measures

State positive affect (PA). Descriptive statistics for PA are found in Table 4.1. Repeated measures ANOVA (2x4) revealed no significant Condition x Time interaction effect or significant main effect of Condition for PA (Table 4.2). The 2x4 ANOVA revealed a significant main effect of Time ($p < .001$) (Table 4.2).

Table 4.1

Descriptive Data for Subjective Psychological Measures over Four Time Periods

	Control	Binaural Beat		
	M (SD)	*M (SD)*	*MM (SD)*	*n*
Positive Affect				
T_1 Baseline	32.62 (5.84)	32.62 (6.23)	32.62 (5.50)	21
T_2 Post-Exercise	33.38 (4.72)	33.57 (5.64)	33.48 (4.72)	21
T_3 Post-Relaxation	23.29 (6.69)	23.10 (6.01)	23.19 (5.87)$^{\#}$	21
T_4 Post-Stress	22.14 (5.92)	24.62 (6.74)	23.38 (5.87)$^{\#}$	21
MM (SD)	27.85 (3.85)	28.48 (4.31)		
Negative Affect				
T_1 Baseline	17.43 (3.65)	17.67 (3.79)	17.55 (3.39)	21
T_2 Post-Exercise	12.62 (2.78)	12.29 (2.35)	12.45 (2.29)$^{\#\#}$	21
T_3 Post-Relaxation	11.95 (2.27)	11.52 (1.72)	11.74 (1.79)$^{\#\#}$	21
T_4 Post-Stress	16.19 (4.61)	14.71 (3.68)	15.45 (3.67)$^{\#\#}$	21
MM (SD)	14.55 (2.57)	14.05 (2.00)		
RELAX	8.29 (1.15)	8.86 (1.01)		21
STRESS	6.19 (2.18)	5.48 (2.14)		21

Note. RELAX = Self-reported relaxation during relaxation protocol (1-10 scale); STRESS = Self-reported stress during math test (1-10 scale); *MM* = marginal mean; *SD* = standard deviation. $^{\#}$ $p < .001$; $T_3 < T_2$, $T_4 < T_2$ $^{\#\#}p < .001$; $T_4 > T_3$, $T_4 > T_2$

Table 4.2

Repeated Measures ANOVA for Positive Affect

Source	SS	*df*	MS	*F*	η^2
Condition	16.10	1.00	16.10	.70	.03
Error	457.16	20.00	22.86		
Time	4018.57	2.00	1339.52	30.74**	.61
Error	2614.68	60.00	43.58		
Condition x Time	49.05	3.00	16.35	2.01	.12
Error	488.70	60.00	8.15		

** $p < .001$.

Mauchly's Test of Sphericity was non-significant. Mean PA decreased significantly from T_2 (post-exercise) to T_3 (post-relaxation) ($p < .001$) and was significantly higher at T_2 than at T_4 ($p < .001$), but did not differ significantly between T_1 and T_2, or between T_3 and T_4 (post-stress) ($p > .05$) (Figure 4.1). Both control and binaural beat relaxation protocols had the effect of reducing PA, by 30.2% and 31.2%, respectively (Figure 4.1).

State negative affect (NA). Descriptive statistics for NA are found in Table 4.1. Repeated measures ANOVA (2x4) revealed no significant Condition x Time interaction effect or significant main effect of Condition (Table 4.3). However, 2x4 repeated measures ANOVA revealed a significant main effect of Time ($p < .001$) (Table 4.4). Mauchly's Test of Sphericity was significant for both the effect of Time ($W = .505$, $\chi^2(2) = 12.990$, $p < .05$) and the Condition x Time interaction ($W = .664$, $\chi^2(2) = 7.773$, $p < .05$), so the Greenhouse-Geisser correction was employed using $\varepsilon = .669$ and $\varepsilon = .749$ respectively. Mean NA decreased significantly from T_1 to T_2 (post-exercise) ($p < .001$), increased significantly from T_3 (post-relaxation) to T_4 (post-stress) ($p < .001$) but did not differ significantly between T_2 and T_3 ($p > .05$) (Figure 4.2). NA scores at T_4 were not significantly different than they were at baseline ($p > .05$).

Exercise significantly decreased NA from trait levels in both control and binaural beat conditions, by 27.6% and 30.4%, respectively (Figure 4.2). These changes were highly significant and represented large effect sizes, for both the control, $t(20) = 5.623$, $p < .001$, $r = .78$ and the binaural beat, $t(20) = 6.294$, $p < .001$, $r = .82$, conditions. The MA stressor significantly increased NA in both conditions ($p < .001$).

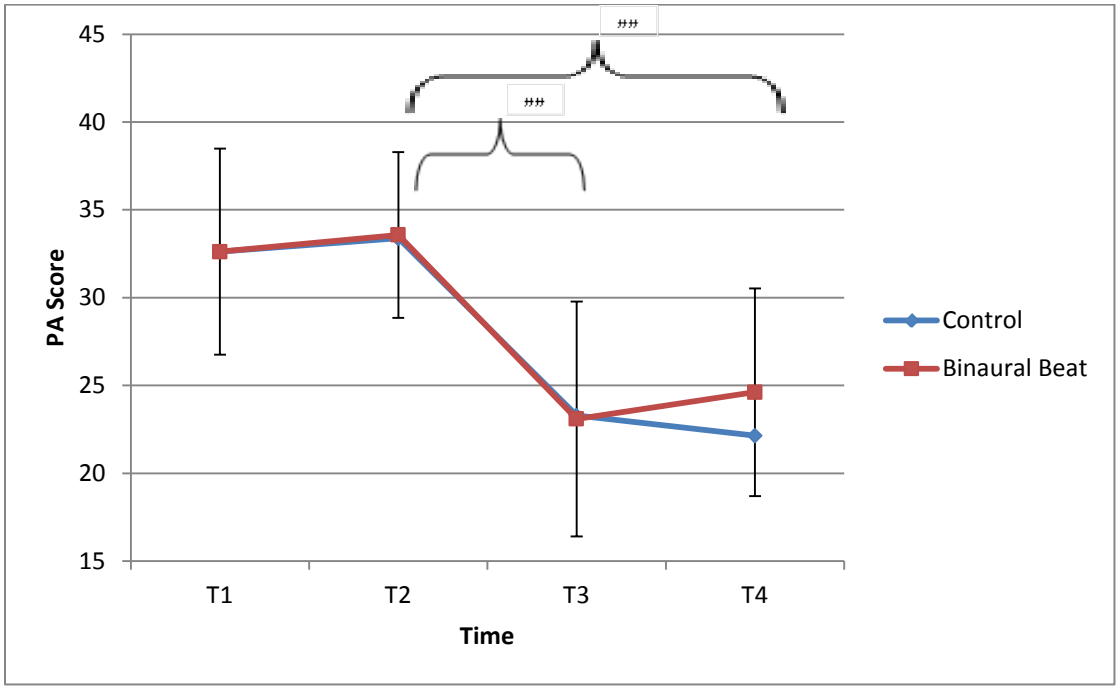

Figure 4.1. Mean positive affect scores at T_1 (baseline/trait), T_2 (post-exercise), T_3 (post-relaxation) and T_4 (post-stress). Error bars represent +/- one standard deviation.
$T_2 > T_3$, $T_2 > T_4$, $p < .001$

Table 4.3

Repeated Measures ANOVA for Negative Affect

Source	SS	*df*	MS	*F*	η^2
Condition	10.50	1.00	10.50	2.46	.11
Error	85.50	20.00	4.28		
Time	917.79	2.23	411.84	33.61**	.63
Error	546.21	44.57	12.26		
Condition x Time	16.07	2.27	7.10	1.21	.06
Error	264.93	45.30	5.85		

** $p < .001$.

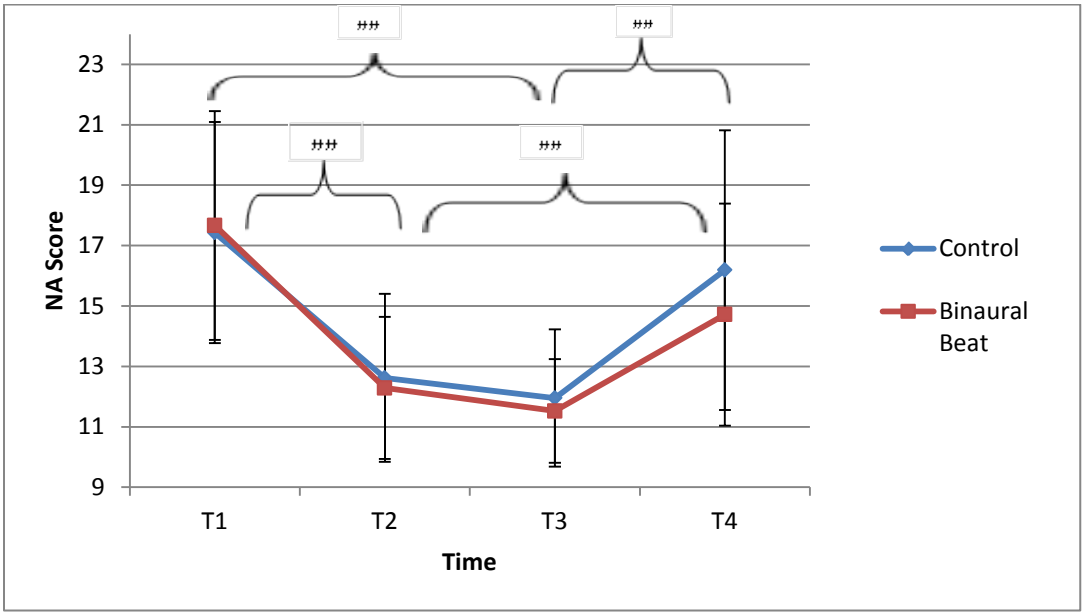

Figure 4.2. Mean negative affect scores at T_1 (baseline/trait), T_2 (post-exercise), T_3 (post-relaxation) and T_4 (post-stress). Error bars represent +/- one standard deviation.
$T_1 > T_2$, $T_1 > T_3$, $T_2 < T_4$, $T_3 < T_4$, $p < .001$

Self-reported relaxation (RELAX). At the end of each session, participants were asked to rate the depth and quality of their relaxation during the 20-min music session on a Likert scale of 1-10, with 10 being the most relaxed and one being the least relaxed. Paired-samples *t*-test showed that while listening to binaural beats, participants reported significantly greater relaxation scores than they did in the control condition (p = .018, one-tailed, r = .45) (Table 4.4). Descriptive statistics for RELAX are found in Table 4.4.

Self-reported stress (STRESS). At the end of each session, participants were asked to rate the intensity of their stress experience during the MA stressor on a Likert scale of 1-10, with 10 being the most stressed and one being the least stressed. Paired-samples *t*-test showed that after listening to binaural beats and being subjected to mental stress, participants reported significantly lower stress scores than they did in the control condition (p = .039, one-tailed, r = .38) (Table 4.5). Descriptive statistics for STRESS are found in Table 4.5.

Physiological Measures

Salivary α-amylase (sAA). Analysis of variance and descriptive statistics for sAA are found in Tables 4.6 and 4.7, respectively. Repeated measures ANOVA (2x4) revealed no significant Condition x Time interaction, no significant main effect of Condition and no significant main effect of Time (Table 4.6). Mauchly's Test of Sphericity was significant for the effect of Time (W = .374, χ^2 (5) = 12.511, $p < .05$), so the Greenhouse-Geisser correction was employed using ε = .596. However, the main effect of Time approached significance (p = .089).

Table 4.4

Paired-Samples t-test for Self-Reported Relaxation Scores

	Condition				
	Control *M*(*SD*)	Binaural Beat *M*(*SD*)	*t*	*df*	*r*
RELAX	8.29 (1.15)	8.86 (1.01)	-2.25 †	20	.45

† $p < .05$, one-tailed

Table 4.5

Paired-Samples t-test for Self-Reported Stress Scores

	Condition				
	Control *M*(*SD*)	Binaural Beat *M*(*SD*)	*t*	*df*	*r*
STRESS	6.19 (2.18)	5.48 (2.14)	1.86 †	20	.38

† $p < .05$, one-tailed

Table 4.6

Repeated Measures ANOVA for Salivary α-amylase

Source	SS	*df*	MS	*F*	η^2
Condition	2.70	1.00	2.70	.00	.00
Error	13404.30	14.00	957.45		
Time	4092.83	1.79	2288.40	2.74	.09
Error	20914.67	25.04	835.28		
Condition x Time	98.57	3.00	32.86	.08	.01
Error	17981.43	42.00	428.13		

Table 4.7

Descriptive Statistics for Physiological Measures over Four Time Periods

	Control	Binaural Beat		
	M (SD)	*M (SD)*	*MM (SD)*	*n*
sAA (kIU/L)				
T_1 Baseline	32.67 (29.27)	34.07 (31.31)	33.37 (29.09)	15
T_2 Post-Exercise	49.00 (47.35)	48.47 (46.23)	48.73 (39.35)	15
T_3 Post-Relaxation	42.40 (40.37)	44.93 (47.95)	43.67 (41.44)	15
T_4 Post-Stress	38.73 (33.56)	36.53 (38.89)	37.63 (34.04)	15
MM (*SD*)	40.70 (33.35)	41.00 (37.30)		
SYS-BP (mmHg)				
T_1 Baseline	111.25 (10.31)	109.05 (10.63)	110.15 (9.88)[#]	20
T_2 Post-Exercise	113.40 (9.76)	113.70 (11.12)	113.55 (9.66)[#]	20
T_3 Post-Relaxation	105.30 (8.75)	104.55 (10.77)	104.93 (9.48)[#]	20
T_4 Post-Stress	107.00 (11.24)	106.40 (8.41)	106.70 (9.17)[#]	20
MM (*SD*)	109.24 (9.53)	108.43 (9.26)		
DIA-BP (mmHg)				
T_1 Baseline	61.30 (8.11)	62.65 (8.92)	61.98 (7.42)	20
T_2 Post-Exercise	61.40 (8.88)	63.50 (7.61)	62.45 (5.63)	20
T_3 Post-Relaxation	60.90 (6.77)	60.25 (8.00)	60.58 (6.40)	20
T_4 Post-Stress	60.95 (6.74)	59.80 (8.79)	60.38 (6.48)	20
MM (*SD*)	61.14 (6.13)	61.55 (6.89)		

Note. sAA = salivary alpha-amylase, kIU/L = kilo international units per liter; mmHg = millimeters of mercury; SYS-BP = Systolic Blood Pressure; DIA-BP = Diastolic Blood Pressure; *MM* = marginal mean; *SD* = standard deviation.

[#] $p < .05$; $T_2 > T_1$, $T_3 < T_2$, $T_4 > T_3$, $T_4 < T_2$

Even though there was a very large amount of variability associated with sAA, the means generally followed the expected trend: rising with exercise and falling with relaxation, but surprisingly continuing to fall after the MA stressor (Table 4.6).

The COCORO Meter™ used in this study was subject to a high degree of potential user error due to the need for very precise timing with the insertion of the salivary test strip, pressing down a lever at the exact right time and then pausing for a brief moment prior to pulling the strip out one notch only. Any mistakes in this process yielded an error message, resulting in lost data for that participant at that time point. Further, since the experimental design was cross-over in nature, if a participant's data were lost at a given time point in one condition, they had to be omitted from analysis in the alternate condition as well. Thus, as Table 4.6 shows, after taking into account sAA error, the analyzable sample size decreased to $n = 15$.

Systolic BP (SYS-BP). Descriptive statistics for SYS-BP are found in Table 4.7. Repeated measures ANOVA (2x4) revealed no significant Condition x Time interaction effect and no significant main effect of Condition (Table 4.8). However, the 2x4 ANOVA revealed a significant main effect of Time ($p < .001$) (Table 4.8). Mauchly's Test of Sphericity was non-significant. Mean SYS-BP increased significantly from T_1 (baseline) to T_2 (post-exercise) ($p < .05$), decreased significantly from T_2 to T_3 (post-relaxation) ($p < .001$) and increased significantly from T_3 to T_4 (post-stress) ($p < .05$) (Figure 4.3). SYS-BP was significantly higher at T_1 than at T_3 ($p < .001$) and at T_2 compared to T_4 ($p < .001$). As expected, SYS-BP was significantly increased through exercise, decreased through relaxation and increased again with the MA stressor (Figure 4.3). One participant's BP data had to be omitted from analysis due to equipment malfunction.

Diastolic BP (DIA-BP). Descriptive statistics for DIA-BP are found in Table 4.7. Repeated measures ANOVA (2x4) revealed no significant Condition x Time interaction effect, no significant main effect of Condition and no significant main effect of Time (Table 4.9). Mauchly's Test of Sphericity was non-significant. Essentially, DIA-BP was constant and not affected significantly by the exercise, relaxation, or mental stress.

Heart Rate Variability (HRV) Measures

Time-domain. Time-domain HRV measures consist of several highly-correlated measures that are derived from either instantaneous heart rate or from the intervals between successive QRS complexes (Task-Force, 1996). Of these measures, SDNN represents "all of the cyclic components responsible for the variability in the period of recording", while RMSSD provides an estimate of the short-term components of variability (Task-Force, 1996). Further, while RMSSD, NN50 and pNN50 are highly correlated and provide information regarding short-term cyclic components of the HR signal, RMSSD is preferred due to better statistical properties (Task-Force, 1996).

Heart rate (HR). Descriptive statistics for HR are found in Table 4.10. Repeated measures ANOVA (2x4) revealed no significant Condition x Time interaction effect and no significant main effect of Condition (Table 4.11). However, the 2x4 ANOVA revealed a significant main effect of Time ($p < .001$) (Table 4.11). Mauchly's Test of Sphericity was significant for the effect of time ($W = .292$, $\chi^2(5) = 23.031$, $p < .001$), so the Greenhouse-Geisser correction was employed using $\varepsilon = .604$. Mean HR differed significantly between all time points ($p < .001$) except for between T_3 (post-relaxation) and T_4 (post-stress) ($p > .05$).

Table 4.8

Repeated Measures ANOVA for Systolic Blood Pressure

Source	SS	*df*	MS	*F*	η^2
Condition	26.41	1	26.41	.78	.04
Error	647.47	19	34.08		
Time	1752.27	3	584.09	30.68**	.62
Error	1085.36	57	19.04		
Condition x Time	32.12	3	10.71	.50	.03
Error	1226.51	57	21.52		

**$p < .001$.

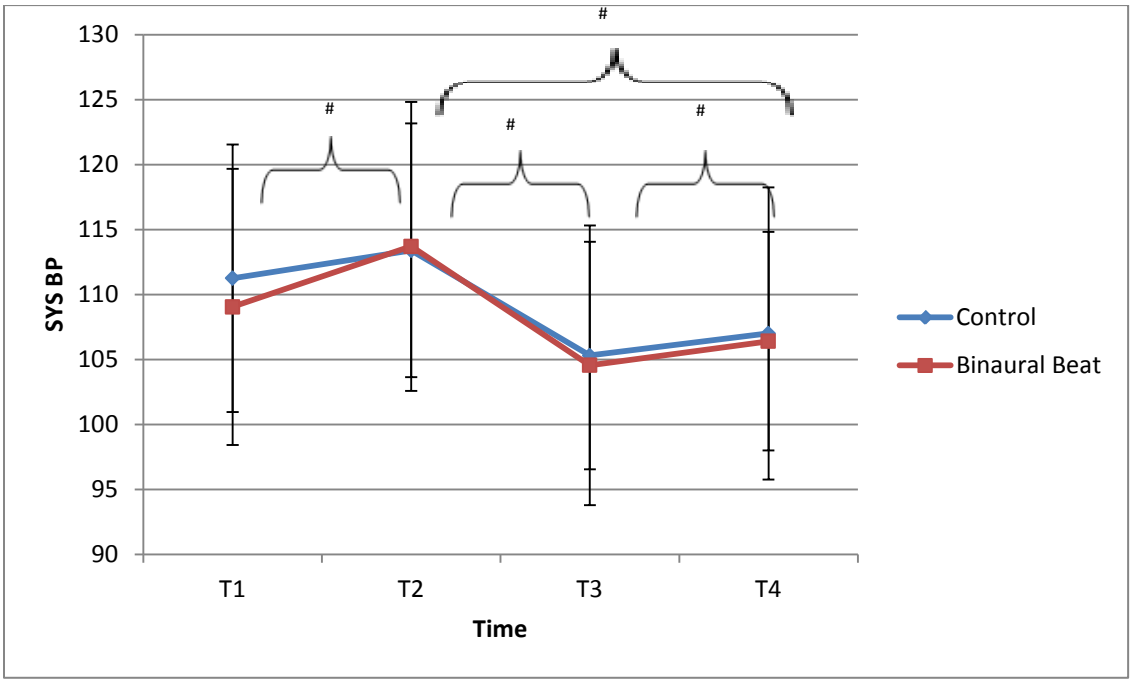

Figure 4.3. Mean Systolic Blood Pressure at T_1 (baseline), T_2 (post-exercise), T_3 (post-relaxation) and T_4 (post-stress). Error bars represent +/- one standard deviation.
$T_2 > T_1$, $T_3 < T_2$, $T_4 > T_3$, $T_4 < T_2$, $p < .05$

Table 4.9

Repeated Measures ANOVA for Diastolic Blood Pressure

Source	SS	*df*	MS	*F*	η^2
Condition	6.81	1	6.81	.07	.00
Error	1939.57	19	102.08		
Time	126.07	3	42.02	1.24	.06
Error	1927.56	57	33.82		
Condition x Time	72.97	3	24.32	1.00	.05
Error	1384.16	57	24.28		

Table 4.10

Descriptive Statistics for Time-Domain Heart Rate Variability Measures over Four Time Periods

	Control	Binaural Beat		
	M (SD)	*M (SD)*	*MM (SD)*	*n*
Heart Rate (bpm)				
T_1 Baseline	72.88 (12.37)	70.60 (12.69)	71.74 (11.91)[#]	21
T_2 Post-Exercise	99.40 (16.72)	98.75 (15.41)	99.08 (15.49)[#]	21
T_3 Post-Relaxation	82.13 (12.82)	82.21 (14.15)	82.17 (13.15)[#]	21
T_4 Post-Stress	81.20 (11.78)	80.23 (13.25)	80.72 (12.05)[#]	21
MM (*SD*)	83.90 (12.79)	82.95 (13.34)		
avRR (ms)				
T_1 Baseline	853.40 (140.47)	883.44 (148.92)	868.42 (137.39)[##]	21
T_2 Post-Exercise	623.46 (112.17)	624.37 (98.710)	623.92 (102.38)[##]	21
T_3 Post-Relaxation	755.86 (120.61)	758.56 (129.21)	757.21 (121.26)[##]	21
T_4 Post-Stress	761.19 (110.00)	773.35 (121.97)	767.27 (111.77)[##]	21
MM (*SD*)	748.48 (25.10)	759.931 (25.78)		
SDNN (ms)				
T_1 Baseline	52.54 (14.56)	54.04 (14.17)	53.29 (12.30)[###]	20
T_2 Post-Exercise	20.85 (15.65)	22.68 (11.38)	21.76 (11.94)[###]	20
T_3 Post-Relaxation	48.70 (17.16)	51.59 (18.50)	50.14 (15.88)[###]	20
T_4 Post-Stress	48.11 (14.93)	52.30 (14.70)	50.21 (12.92)[###]	20
MM (*SD*)	42.55 (12.61)	45.15 (12.12)		

Note. bpm = beats per minute; avRR = average R-R interval; SDNN = standard deviation of N-N intervals; *MM* = marginal mean; *SD* = standard deviation;

[#] $p < .001$; $T_2 > T_1$, $T_3 < T_2$, $T_4 < T_2$, $T_4 > T_1$
[##] $p < .001$; $T_2 < T_1$, $T_3 > T_2$, $T_4 > T_2$, $T_4 < T_1$
[###] $p < .001$; $T_2 < T_1$, $T_3 > T_2$, $T_4 > T_2$

Table 4.10, continued

Descriptive Statistics for Time-Domain Heart Rate Variability Measures over Four Time Periods

	Control	Binaural Beat		
	M (SD)	*M (SD)*	*MM (SD)*	*n*
RMSSD (ms)				
T_1 Baseline	45.42 (18.16)	50.78 (20.59)	48.10 (18.10)[#]	21
T_2 Post-Exercise	12.81 (12.46)	13.46 (8.39)	13.14 (18.15)[#]	21
T_3 Post-Relaxation	36.23 (19.81)	39.39 (19.20)	37.81 (18.15)[#]	21
T_4 Post-Stress	34.25 (15.11)	38.90 (15.96)	36.57 (14.49)[#]	21
MM (*SD*)	32.18 (14.71)	35.63 (14.11)		
NN50				
T_1 Baseline	32.67 (19.31)	39.33 (22.35)	36.00 (19.43)[##]	21
T_2 Post-Exercise	3.67 (11.18)	3.00 (4.86)	3.33 (7.61)[##]	21
T_3 Post-Relaxation	23.10 (19.97)	27.43 (19.61)	25.26 (17.78)[##]	21
T_4 Post-Stress	21.95 (17.73)	28.38 (19.48)	25.17 (7.23)[##]	21
MM (*SD*)	20.35 (14.76)	24.54 (14.48)		

Note. RMSSD = Root mean standard deviation; NN50 = frequency of N-N intervals separated by more than 50 ms; *MM* = marginal mean; *SD* = standard deviation.

[#] $p < .001$; $T_2 < T_1$, $T_3 > T_2$, $T_4 > T_2$, $T_4 < T_1$
[##] $p < .01$; $T_2 < T_1$, $T_3 > T_2$, $T_4 > T_2$, $T_4 < T_1$

Table 4.11

Repeated Measures ANOVA for Heart Rate

Source	SS	*df*	MS	*F*	η^2
Condition	38.17	1.00	38.17	0.49	.02
Error	1569.12	20.00	78.46		
Time	16399.60	1.81	9053.72	141.18**	.88
Error	2323.24	36.23	64.13		
Condition x Time	30.54	3.00	10.18	1.00	.05
Error	608.71	60.00	10.15		

** $p < .001$.

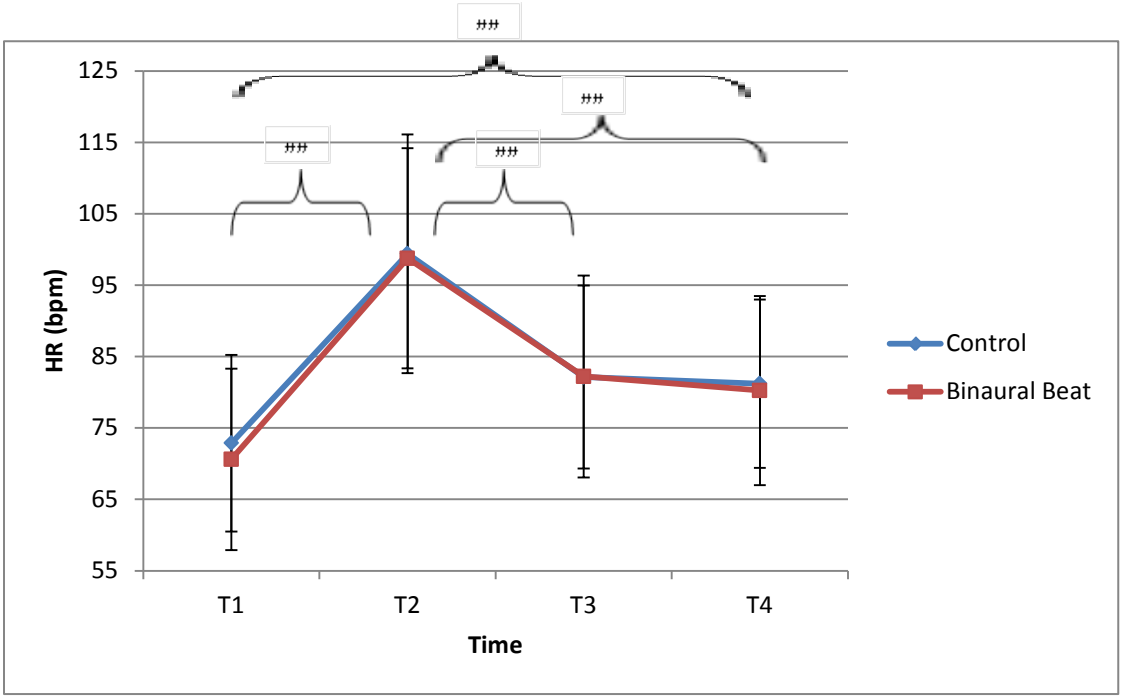

Figure 4.4. Mean Heart Rate at T_1 (baseline), T_2 (post-exercise), T_3 (post-relaxation) and T_4 (post-stress). Error bars represent +/- one standard deviation.
$T_2 > T_1$, $T_3 < T_2$, $T_4 < T_2$, $T_4 > T_1$, $p < .001$

As with sAA, HR generally followed the expected trend; HR increased with exercise and decreased with relaxation. The MA stressor did not have a significant effect on HR (Figure 4.4).

Average R-R interval (avRR). Descriptive statistics for avRR are found in Table 4.10. Repeated measures ANOVA (2x4) revealed no significant Condition x Time interaction effect or main effect of Condition (Table 4.12). However, the 2x4 ANOVA revealed a significant main effect of Time ($p < .001$) (Table 4.12). Mauchly's Test of Sphericity was significant for the effect of time ($W = .224$, $\chi^2 (5) == 27.985$, $p < .001$) and for the Condition x Time interaction effect ($W = .553$, $\chi^2 (5) = 11.106$, $p < .05$), so the Greenhouse-Geisser correction was employed using $\varepsilon = .526$ and $\varepsilon = .739$ respectively. As expected, the R-R intervals followed an inverse pattern to that of HR (Figure 4.5). Specifically, avRR differed significantly between all time points ($p < .001$) except for between T_3 (post-relaxation) and T_4 (post-stress) ($p > .05$) (Figure 4.5).

Standard deviation of R-R intervals (SDNN). Descriptive statistics for SDNN are found in Table 4.10. Repeated measures ANOVA (2x4) revealed no significant Condition x Time interaction effect and no significant main effect of Condition (Table 4.13). However, the 2x4 ANOVA showed a significant main effect of Time ($p < .001$) (Table 4.13). Mauchly's Test of Sphericity was non-significant. Mean SDNN decreased significantly from T_1 (baseline) to T_2 (post-exercise) ($p < .001$), increased significantly from T_2 toT_3 (post relaxation) ($p < .001$) and was significantly lower at T_2 than T_4 (post stress) ($p < .001$), but did not differ significantly between T_1 and T_3, T_1 and T_4, or T_3 and T_4 ($p > .05$) (Figure 4.6).

Table 4.12

Repeated Measures ANOVA for Average R-R Interval

Source	SS	*df*	MS	*F*	η^2
Condition	5508.60	1.0	5508.60	.86	.04
Error	127704.87	20.0	6385.24		
Time	1268408.45	1.58	803237.93	117.83**	.86
Error	215296.87	31.58	6816.99		
Condition x Time	5606.46	2.22	2530.29	1.88	.00
Error	59635.79	44.32	1345.73		

** $p < .001$.

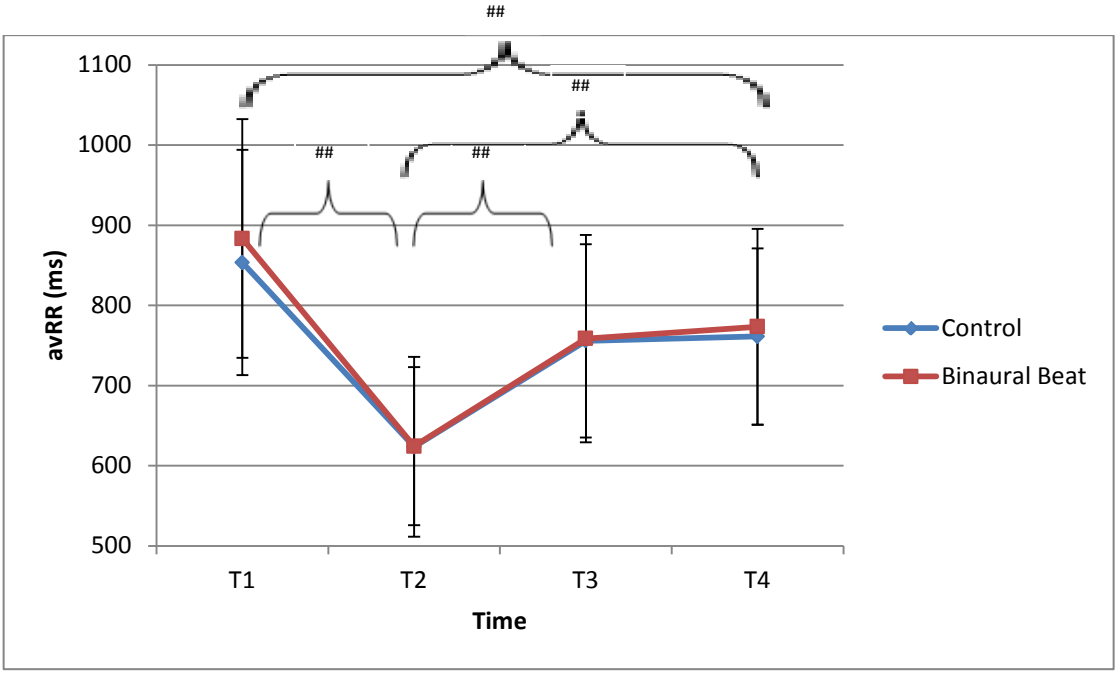

Figure 4.5. Mean R-R interval at T_1 (baseline), T_2 (post-exercise), T_3 (post-relaxation) and T_4 (post-stress). Error bars represent +/- one standard deviation.
$T_2 < T_1$, $T_3 > T_2$, $T_4 > T_2$, $T_4 < T_1$, $p < .001$

Table 4.13

Repeated Measures ANOVA for Standard Deviation of R-R Intervals

Source	SS	*df*	MS	*F*	η^2
Condition	283.66	1.00	283.66	1.43	.07
Error	3958.73	20.00	197.94		
Time	27594.83	3.00	9198.28	68.90**	.78
Error	8009.69	60.00	133.50		
Condition x Time	46.32	3.00	15.44	.26	.01
Error	3533.65	60.00	58.90		

** $p < .001$.

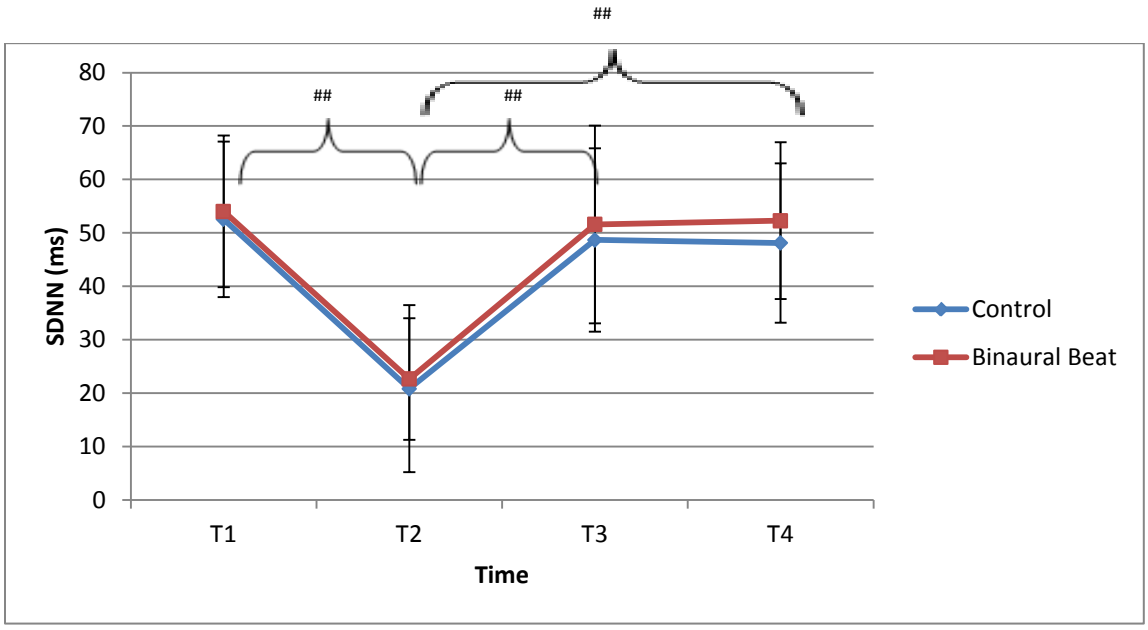

Figure 4.6. Mean SDNN at T_1 (baseline), T_2 (post-exercise), T_3 (post-relaxation) and T_4 (post-stress). Error bars represent +/- one standard deviation.
$T_2 < T_1$, $T_3 > T_2$, $T_4 > T_2$, $p < .001$

As expected, exercise caused a significant decrease in SDNN and relaxation caused a significant increase back to baseline levels. The MA stressor did not significantly affect SDNN.

Root mean square standard deviation of R-R intervals (RMSSD). Descriptive statistics for RMSSD are found in Table 4.10. Repeated measures ANOVA (2x4) revealed no significant Condition x Time interaction effect and no significant main effect of Condition (Table 4.14). Mean RMSSD decreased significantly from T_1 (baseline) to T_2 (post-exercise)($p < .001$), increased significantly from T_2 to T_3 (post relaxation) ($p < .001$) and was significantly lower at T_2 than T_3 and T_4 (post stress) ($p < .001$) (Figure 4.7). RMSSD was significantly higher at T_1 than T_4 ($p < .001$), but did not differ significantly between T_3 and T_4 ($p > .05$). Exercise caused a decrease in RMSSD and relaxation increased RMSSD, but not all the way back to baseline levels.

NN50. Descriptive statistics for NN50 are found in Table 4.12. Repeated measures ANOVA (2x4) revealed no significant Condition x Time interaction effect ($p >$ and no significant main effect of Condition (Table 4.15). However, the 2x4 ANOVA revealed a significant main effect of Time ($p < .001$) (Table 4.15). Mauchly's Test of Sphericity was significant for the effect of time ($W = .431$, $\chi^2 (5) = 15.740$, $p < .05$), so the Greenhouse-Geisser correction was employed using $\varepsilon = .686$. Mean NN50 decreased significantly from T_1 (baseline) to T_2 (post-exercise) ($p < .001$), increased significantly from T_2 to T_3 (post-relaxation) ($p < .001$) and was significantly higher at T_1 than T_4 (post-stress) ($p < .01$) (Figure 4.8). NN50 was significantly lower at T_2 than T_4 ($p < .001$) and not differ significantly between T_3 and T_4 ($p > .05$).

Table 4.14

Repeated Measures ANOVA for Root Mean Square Standard Deviation of R-R Intervals

Source	SS	*df*	MS	*F*	η^2
Condition	500.25	1.00	500.25	2.79	.12
Error	3592.59	20.00	179.63		
Time	27518.84	3.00	9172.95	62.54**	.76
Error	8800.65	60.00	146.68		
Condition x Time	136.82	3.00	45.61	1.13	.05
Error	2415.45	60.00	40.26		

** $p < .001$.

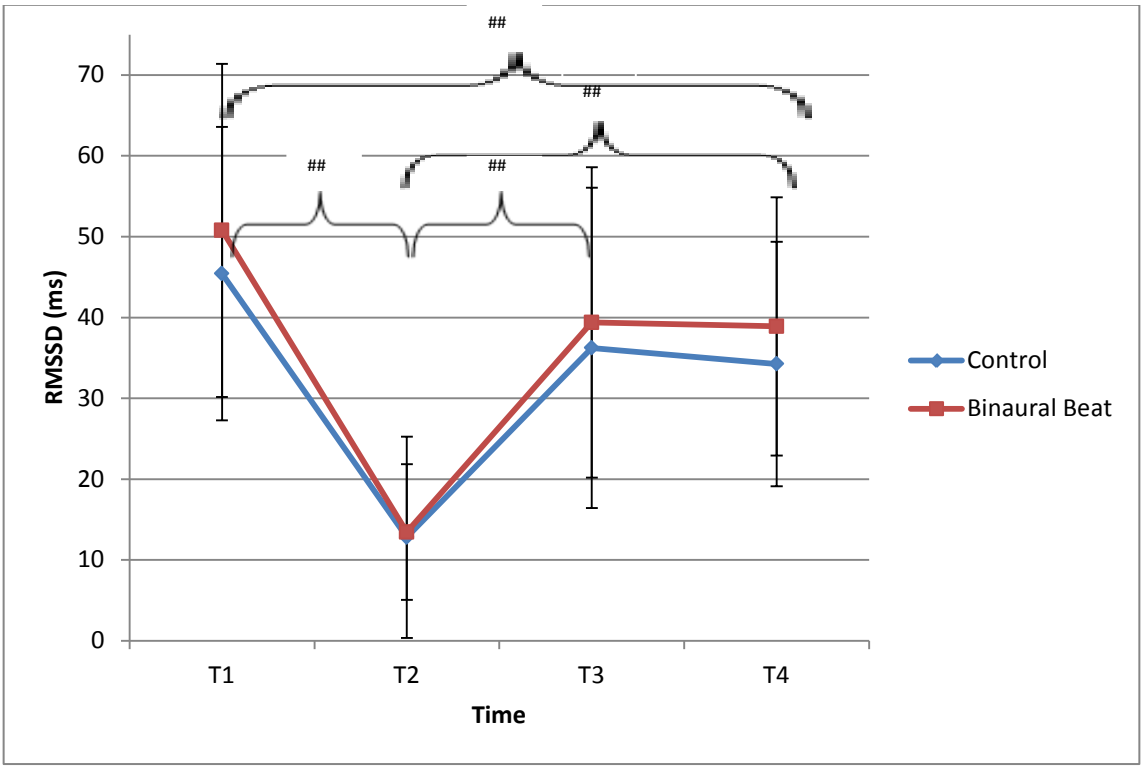

Figure 4.7. Mean RMSSD at T_1 (baseline), T_2 (post-exercise), T_3 (post-relaxation) and T_4 (post-stress). Error bars represent +/- one standard deviation.
$T_2 < T_1$, $T_3 > T_2$, $T_4 > T_2$, $T_4 < T_1$, $p < .001$

Table 4.15

Repeated Measures ANOVA for NN50

Source	SS	*df*	MS	*F*	η^2
Condition	737.52	1.00	737.52	3.77	.16
Error	3917.23	20.00	195.86		
Time	23702.12	2.06	11518.74	40.93**	.67
Error	11583.13	41.15	281.46		
Condition x Time	364.91	2.70	135.25	1.76	.08
Error	4136.35	53.96	76.66		

** $p < .001$.

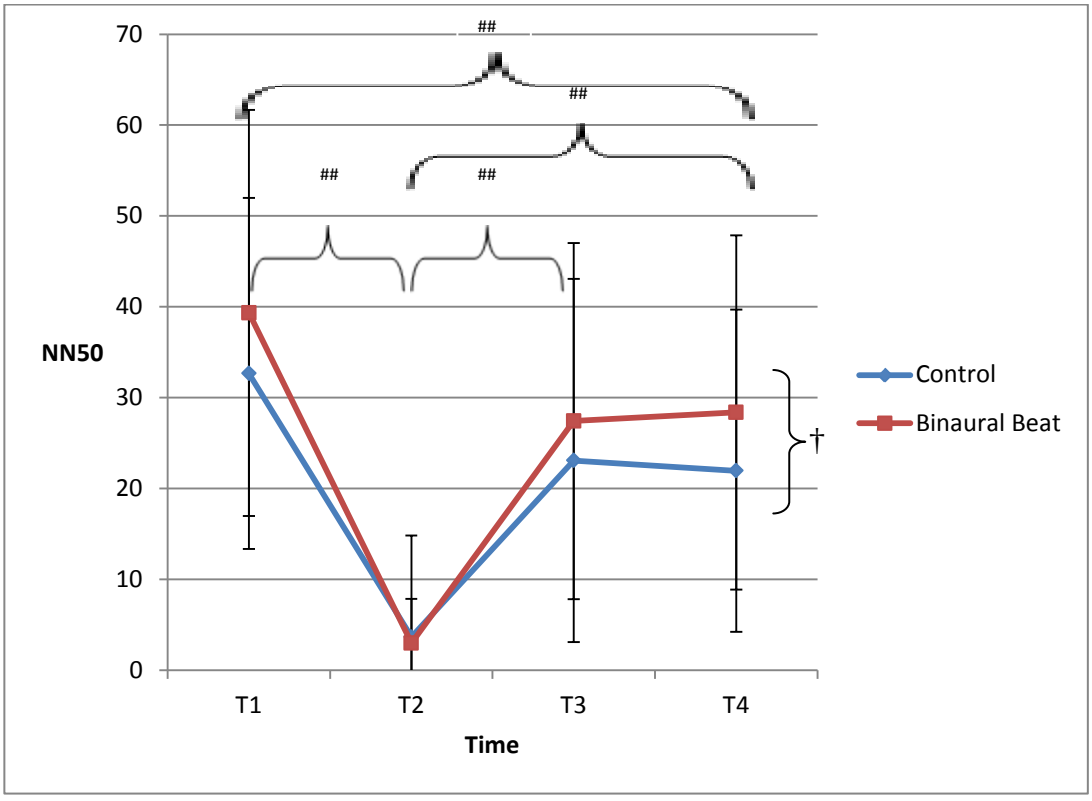

Figure 4.8. Mean NN50 at T_1 (baseline), T_2 (post-exercise), T_3 (post-relaxation) and T_4 (post-stress). Error bars represent +/- one standard deviation.
† $p < .05$, one-tailed; $r = .42$; ## $p < .01$; $T_2 < T_1$, $T_3 > T_2$, $T_4 > T_2$, $T_4 < T_1$

As with the other HRV measures, NN50 was significantly affected by both exercise (it was decreased) and relaxation (it was increased), but was not significantly affected by the MA stressor. At T_4, NN50 values were still significantly lower than baseline levels. It is worth noting that the ANOVA for Condition approached significance ($p = .067$) (Table 4.15). Participants in the binaural beat condition demonstrated higher mean NN50 measurements at both T_1 and T_4. Upon further inspection, paired-samples *t*-test revealed a significant difference between conditions only at T_4 (Table 4.16), indicating a potential reduction in stress responsivity after listening to binaural beats.

Frequency-domain (Fast Fourier Transformed). Power spectral density in the frequency domain can be calculated through both parametric (AR spectrum) and nonparametric (FFT spectrum) analyses. Generally, nonparametric methods are preferred due to their simplicity and speed of processing (Task-Force, 1996).

Low-frequency power (normalized units) (LF). Descriptive statistics for LF are found in table 4.17. Repeated measures ANOVA (2x4) revealed no significant Condition x Time interaction effect and no significant main effect of Condition (Table 4.18). However, the 2x4 ANOVA revealed a significant main effect of Time ($p < .001$) (Table 4.18). Mauchly's Test of Sphericity was significant for the effect of time ($W = .537$, $\chi^2(5) = 11.646$, $p < .05$), so the Greenhouse-Geisser correction was employed using $\varepsilon = .778$. Mean LF significantly increased from T_1 (baseline) to T_2 (post-exercise) ($p < .001$), significantly decreased from T_2 and T_3 (post-relaxation) ($p < .01$) and was significantly lower at T_1 than T_4 (post-stress) ($p < .05$)(Figure 4.9). LF was significantly higher at T_2 compared to T_4 ($p < .05$) and did not change significantly between T_3 and T_4 ($p > .05$).

Table 4.16

Paired-Samples t-test for NN50

	Condition				
	Control *M*(*SD*)	Binaural Beat *M*(*SD*)	*t*	*df*	*r*
NN50-T_1	32.67(19.31)	39.33(22.35)	-1.982	20	.41
NN50-T_4	21.95(17.73)	28.38(19.48)	-2.069†	20	.42

$^{\dagger}p < .05$, one-tailed

Table 4.17

Descriptive Statistics for Frequency-Domain Heart Rate Variability Measures over Four Time Periods

		Control	Binaural Beat		
		M (SD)	*M (SD)*	*MM (SD)*	*n*
Low Frequency Power (n.u.)					
	T_1	70.57 (14.76)	66.07 (20.92)	68.32 (15.40)$^{\#}$	21
	T_2	86.36 (9.78)	85.98 (9.17)	86.17 (7.93)$^{\#}$	21
	T_3	77.33 (12.70)	75.41 (14.36)	76.38 (11.36) $^{\#}$	21
	T_4	78.97 (13.16)	74.86 (14.75)	76.91 (12.74)$^{\#}$	21
		78.31 (9.90)	75.58 (11.78)		
High Frequency Power (n.u.)					
	T_1	33.21 (19.13)	33.93 (20.92)	33.57 (15.53)$^{\#\#}$	21
	T_2	13.64 (9.78)	14.02 (9.17)	13.83 (7.93)$^{\#\#}$	21
	T_3	22.67 (12.70)	24.59 (14.36)	23.63 (11.36)$^{\#\#}$	21
	T_4	21.03 (13.16)	25.14 (14.75)	23.09 (12.34)$^{\#\#}$	21
		22.64 (2.10)	24.42 (2.57)		
LF/HF					
	T_1	3.37 (2.38)	3.40 (2.98)	3.39 (2.15)$^{\#\#\#}$	21
	T_2	10.98 (8.52)	9.86 (7.50)	10.42 (6.83)$^{\#\#\#}$	21
	T_3	6.10 (6.84)	4.62 (3.25)	5.36 (4.49)$^{\#\#\#}$	21
	T_4	6.34 (5.27)	4.98 (4.70)	5.66 (3.85)$^{\#\#\#}$	21
		6.70 (4.08)	5.72 (3.53)		

Note. n.u.= normalized units; LF/HF = Low frequency to high frequency ratio; *MM* = marginal mean; *SD* = standard deviation.

$^{\#}$ $p < .05$; $T_2 > T_1$, $T_3 < T_2$, $T_4 < T_2$, $T_4 > T_1$
$^{\#\#}$ $p < .05$; $T_2 < T_1$, $T_3 > T_2$, $T_4 > T_2$, $T_4 < T_1$
$^{\#\#\#}$ $p < .05$; $T_2 > T_1$, $T_3 < T_2$, $T_4 < T_2$

Table 4.18

Repeated Measures ANOVA for Low Frequency Power (normalized units)

Source	SS	*df*	MS	*F*	η^2
Condition	313.24	1.00	313.24	2.63	.12
Error	2385.16	20.00	119.26		
Time	6710.60	2.34	2873.92	18.92**	.49
Error	7094.88	46.70	151.92		
Condition x Time	117.96	3.00	39.32	.41	.02
Error	5825.98	60.00	97.10		

** $p < .001$.

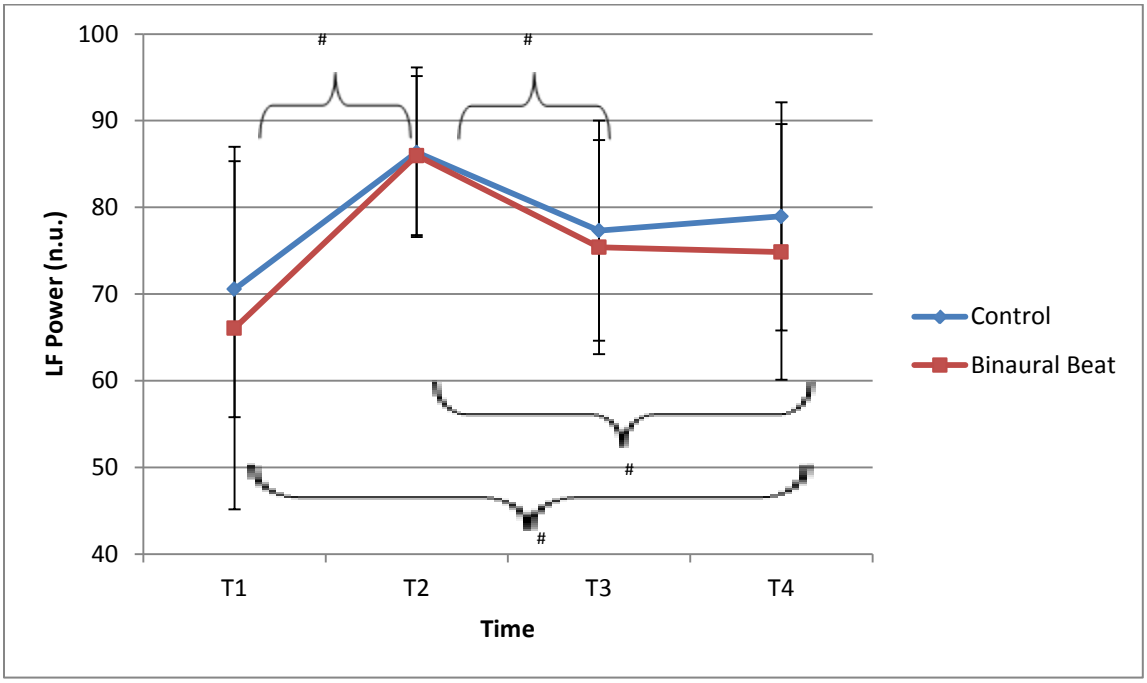

Figure 4.9. Mean Low-Frequency Power at T_1 (baseline), T_2 (post-exercise), T_3 (post-relaxation) and T_4 (post-stress). Error bars represent +/- one standard deviation.
# $p < .05$; $T_2 > T_1$, $T_3 < T_2$, $T_4 < T_2$, $T_4 > T_1$

High-frequency power (normalized units) (HF). Descriptive statistics for HF are found in Table 4.17. Repeated measures ANOVA (2x4) revealed no significant Condition x Time interaction effect and no significant main effect of Condition (Table 4.19). However, the 2x4 ANOVA revealed a significant main effect of Time ($p < .001$) (Table 4.19). Mauchly's Test of Sphericity was significant for the Condition x Time interaction effect ($W = .457$, $\chi^2 (5) = 14.646$, $p < .05$), so the Greenhouse-Geisser correction was employed using $\varepsilon = .800$.

Mean HF decreased significantly from T_1 (baseline) to T_2 (post-exercise) ($p < .001$), increased significantly from T_2 to T_3 (post-relaxation) ($p < .01$) and was significantly higher at T_1 compared to T_4 (post-stress) ($p < .05$) (Figure 4.10). HF was significantly lower at T_2 compared to T_4 ($p < .05$), but did not change significantly from T_3 to T_4 ($p > .05$).

Low-frequency to High-frequency ratio (LF/HF). Descriptive statistics for LF/HF are found in Table 4.17. Repeated measures ANOVA (2x4) revealed no significant Condition x Time interaction effect and no significant main effect of Condition (Table 4.20). However, the 2x4 ANOVA revealed a significant main effect of Time ($p < .001$) (Table 4.20). Mauchly's Test of Sphericity was significant for both the effect of time ($W = .445$, $\chi^2 (5) = 15.167$, $p < .05$) and the Condition x Time interaction ($W = .492$, $\chi^2 (5) = 13.271$, $p < .05$), so the Greenhouse-Geisser correction was employed using $\varepsilon = .646$ and $\varepsilon = .668$ respectively. Mean LF/HF increased significantly from T_1 (baseline) to T_2 (post-exercise) ($p < .001$), decreased significantly from T_2 and T_3 (post-relaxation) ($p < .05$)(Figure 4.11). LF/HF was significantly higher at T_2 compared to T_4 (post-stress) ($p < .001$) and did not differ significantly between T_3 and T_4 ($p > .05$).

Table 4.19

Repeated Measures ANOVA for High Frequency Power (normalized units)

Source	SS	*df*	MS	*F*	η^2
Condition	133.93	1.00	133.93	.92	.04
Error	2897.95	20.00	144.90		
Time	8192.64	3.00	2730.88	19.92**	.50
Error	8225.11	60.00	137.09		
Condition x Time	89.70	1.93	46.47	.22	.01
Error	8140.96	38.60	210.90		

** $p < .001$.

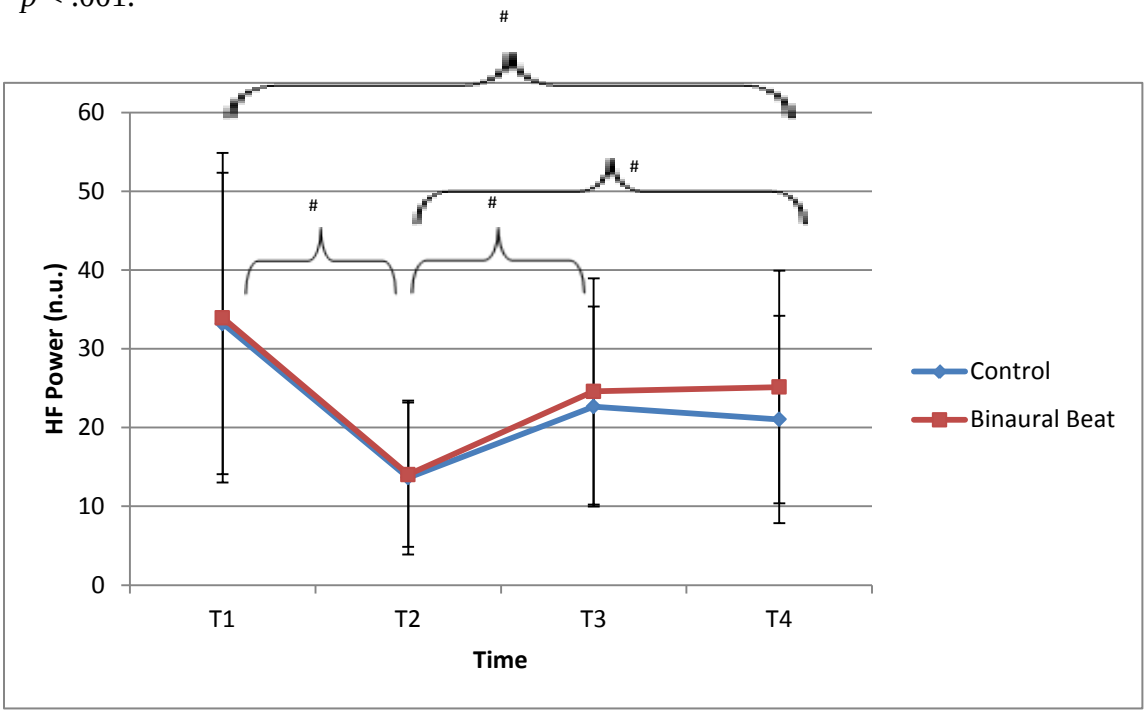

Figure 4.10. Mean High-Frequency Power at T_1 (baseline), T_2 (post-exercise), T_3 (post-relaxation) and T_4 (post-stress). Error bars represent +/- one standard deviation.
# $p < .05$; $T_2 < T_1$, $T_3 > T_2$, $T_4 > T_2$, $T_4 < T_1$,

Table 4.20

Repeated Measures ANOVA for Low Frequency to High Frequency Ratio

Source	SS	*df*	MS	*F*	η^2
Condition	40.23	1.00	40.23	1.70	.08
Error (Condition)	474.76	20.00	23.74		
Time	1122.03	1.94	579.01	13.81**	.41
Error (Time)	1624.80	38.76	41.92		
Condition x Time	14.97	2.01	7.47	.28	.01
Error (Condition x Time)	1071.20	40.11	53.56		

** $p < .001$.

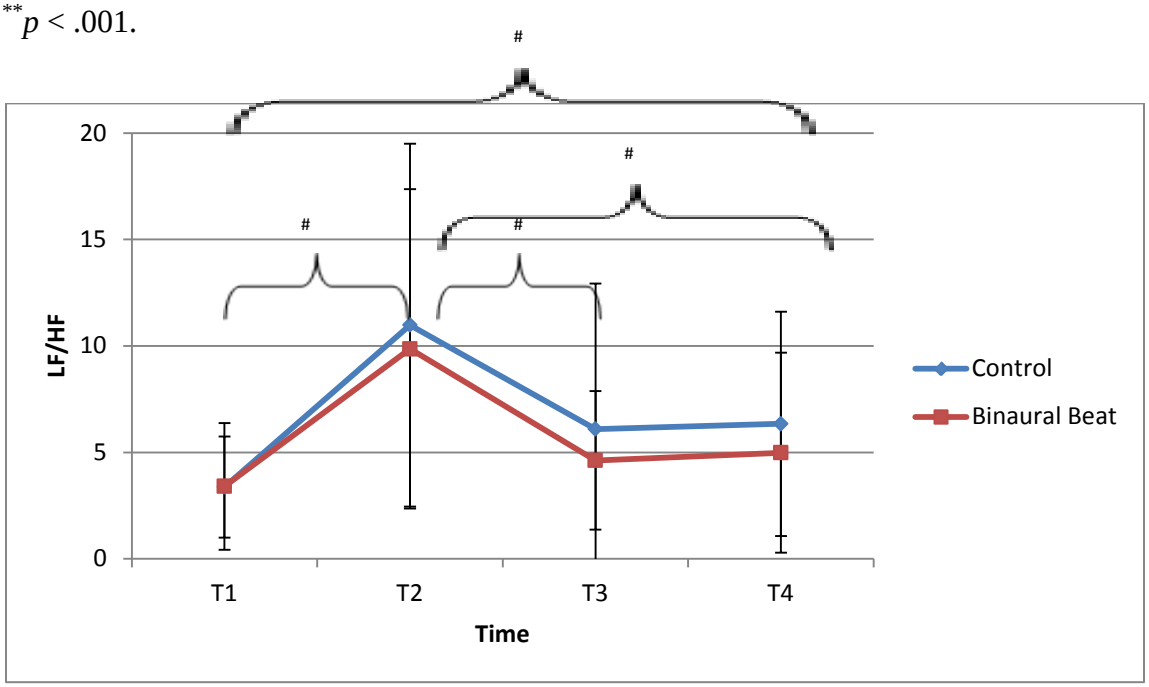

Figure 4.11. Mean Low-Frequency to High-Frequency Power Ratio at T_1 (baseline), T_2 (post-exercise), T_3 (post-relaxation) and T_4 (post-stress). Error bars represent +/- one standard deviation.

# $p < .05$; $T_2 > T_1$, $T_3 < T_2$, $T_4 < T_2$, $T_4 > T_1$

LF/HF followed the anticipated trend, significantly increasing with exercise and decreasing with relaxation. The mental stress test had no significant effect on LF/HF.

HRV measures during 20-min relaxation protocol. All HRV indices were recorded during the 20-min relaxation period and compared between conditions. Paired-samples *t*-tests were run to assess differences between binaural beat and control conditions on each of the HRV (time-domain and frequency-domain) measures during the 20-min relaxation protocol (Table 4.21). No significant differences were found for any HRV indices during the relaxation protocol.

Summary

Effects of exercise. Exercise for 20-min at approximately 70% of $V0_2max$ resulted in a large significant decrease in NA ($p < .05$, mean $r = .80$) from baseline levels. Salivary α-amylase was increased by exercise, as expected, but this effect did not reach statistical significance due to large amounts of variability in the measure. Exercise produced a typical cardiovascular response, significantly increasing HR, SYS-BP, LF power and LF/HF, while decreasing HF power, avRR, RMSSD, SDNN and NN50.

Effects of relaxation. There were no significant differences in HRV indices between the control and binaural beat relaxation protocols during the 20-min relaxation session or immediately after (i.e., at T_3). In both control and binaural beat conditions relaxation decreased PA by 30.2% and 31.2% respectively. NA was unaffected by relaxation. Most physiological variables showed the anticipated response: decreased SYS-BP, HR, LF power and LF/HF, along with increased HF power, avRR, RMSSD, SDNN and NN50.

Table 4.21

Paired-Samples t-tests for HRV Indices during 20-min Relaxation Protocol

	Condition					
	Control *M* (*SD*)	Binaural Beat *M* (*SD*)	*t*	*df*	*p*	*n*
HR (bpm)	80.05 (15.15)	79.62 (15.57)	.25	20	.40	21
avRR (ms)	782.55 (154.91)	787.28 (154.90)	-.31	20	.38	21
SDNN (ms)	38.80 (17.00)	39.70 (15.81)	-.26	20	.40	21
RMSSD (ms)	33.79 (19.22)	35.25 (18.35)	-.39	20	.35	21
NN50	208.14 (198.92)	223.81 (184.30)	-.38	20	.35	21
LF (n.u.)	70.33 (15.09)	69.72 (17.4)	.29	20	.39	21
HF (n.u.)	29.67 (15.09)	30.28 (17.4)	-.29	20	.39	21
LF/HF	3.22 (2.01)	3.58 (2.90)	-.92	20	.19	21

Note. HR = Heart Rate; bpm = beats per minute; avRR = average R-R interval; SDNN = standard deviation of N-N intervals; RMSSD = Root mean standard deviation; NN50 = frequency of N-N intervals separated by more than 50 ms; LF = Low frequency; HF = High frequency; n.u.= normalized units; LF/HF = Low frequency to high frequency ratio; *MM* = marginal mean; *SD* = standard deviation.

Effects of mental stress. The MA stressor significantly increased NA in both conditions but did not significantly affect PA. Systolic, but not diastolic, blood pressure increased significantly with mental stress. Participants in the binaural beat condition demonstrated significantly higher NN50 values at T_4 than the control group. Of the physiological variables, only SYS-BP and SDNN returned to baseline levels.

At the end of each experimental session, when asked to provide subjective ratings (Likert scale of 1-10) of perceived stress during the mental stress test and perceived relaxation during the relaxation protocol, participants in the binaural beat condition reported 11.5% less perceived stress (p = .039, one-tailed, r = .38) and 6.4% more perceived relaxation (p = .018, one-tailed, r = .45) than they did in the control condition.

CHAPTER 5

DISCUSSION

Finding its theoretical basis in the auditory driving and ERTAS-arousal modulating hypotheses, the present study investigated the effectiveness of theta-frequency binaural beats in facilitating the post-exercise relaxation response and attenuating a subsequent stress response. No other study to date has examined the effects of binaural beats on HRV measures or sAA, so comparisons to relevant literature are limited. The experimental findings generally do not provide sufficient evidence to reject the null hypotheses and therefore do not support a beneficial entrainment effect of theta-frequency binaural beats post-exercise, during the 20-min relaxation protocol, post-relaxation, or post-stress.

Effects of Exercise and Relaxation

Consistent with previous literature, exercise served as an ergotropic stimulus which increased SNS activity as measured through increased HR (decreased HRV in the time-domain and increased LF power) as well as elevated BP and salivary sAA concentrations (Allgrove, Gomes, Hough, & Gleeson, 2008; Boettger et al., 2010; Bricout et al., 2010; Mendonca, Fernhall, Heffernan, & Pereira, 2009; Nater & Rohleder, 2009; Santaella et al., 2006). The idea was that through initially evoking an ergotropic response, it would be easy to gauge the effectiveness of a trophotropic stimulus (i.e., the binaural beat treatment) through a deepening and quickening of the post-exercise relaxation response.

Presently, it seems that this assumption may have been unfounded. Instead of facilitating the intrinsically potent post-exercise relaxation response, any effects of the

binaural beat treatment may have been obscured by normal shifts back to parasympathetic dominance (Raglin & Morgan, 1987; Santaella et al., 2006). This was reflected in the results which demonstrated, in both conditions, a relaxation-induced decrease in PA, SYS-BP, HR, LF Power and LF/HF ratio, with a concomitant increase in time-domain HRV variables (avRR, SDNN, RMSSD and NN50). All of these experimental results are characteristic of the relaxation response (Lucini et al., 1997; Peng et al., 2004; Sakakibara et al., 1994; Tang et al., 2009; Terathongkum & Pickler, 2004), yet no effect of the binaural beats was evident.

It is plausible that the potency of the post-exercise relaxation response trumped any potential facilitatory effects of the binaural beat treatment (i.e., a 'wash-out' effect). Other evidence for this suggested exercise 'wash-out' effect can be found in the experimental findings of Kubitz and Pothakis (1997). They demonstrated that exercise reduced beta power and increased both theta and alpha power in participants. If this also occurred in the present study, participants may have been unaffected by any potential, yet subtle, effects of the binaural beat treatment. Alternatively, the binaural beats might have been completely ineffective at promoting a deeper relaxation response.

Supporting the anxiolytic role of exercise, the present study demonstrated 27.6% and 30.4% reductions in NA post-exercise in both the control and binaural beat groups, respectively. This is somewhat in conflict with previous literature that shows acute exercise-related changes in positive affect (increased PA) and state anxiety, but not negative affect (McIntyre & Watson, 1990; Petruzzello, Jones, & Tate, 1997). However, training history, exercise duration and exercise intensity all seem to play important roles in exercise-induced changes in perceived affect. A study comparing untrained and

trained runners demonstrated similar findings to the present study (decreased NA) in untrained, but not trained runners (Boutcher, McAuley, & Courneya, 1997). Another study found that 15-min of running (at 75% VO_{2max}) caused no change in NA, while 30-min of running did (Petruzzello & Landers, 1994b).

Relaxation had no effect on NA, but decreased PA by 30.2% and 31.2% in both the control and binaural beat groups. This is unusual, as tropotrophic practices typically are cited as increasing PA (Davidson et al., 2003). This potentially highlights the inadequacy of the PANAS scale to differentiate between the valence (positive or negative) and arousal (high or low) components of affect (Lindquist et al., 2012). Alternatively, given the convenient student sample selected for the present study, participants may have become bored and/or anxious with the relaxation protocol. This may have been reflected in the decreased PA scores and also potentially in the lack of a significant binaural beat treatment effect. In short, the effects of exercise and the convenient student sample may both have independently confounded the effects of the relaxation protocol and the binaural beat treatment.

Multiple studies have demonstrated significant trophotrophic ((Brady & Stevens, 2000; Dabu-Bondoc et al., 2010; Kliempt et al., 1999; Lavallee, Koren, & Persinger, 2011; Le Scouarnec et al., 2001; Lewis et al., 2004; Padmanabhan et al., 2005; Wahbeh, Calabrese, & Zwickey, 2007) and ergotropic (Kennerly, 1994; Lane et al., 1998) binaural beat effects. Critically, the participants in the Brady and Stevens (2000) study (which shared the same experimental stimuli as the present study) were all given prior information regarding theta-frequency binaural beat sound stimulation as well as the general purpose of the experimental sessions. Conversely, in the Lane et al. (1998) study,

which demonstrated a seemingly effective beta-frequency binaural beat entrainment, participants were blinded to the true purpose of the study and naïve to the presence of binaural beats. This disparity can be partially explained by the role attention plays in entrainment to acoustic stimuli (Lazzouni, Ross, Voss, & Lepore, 2010; Saupe et al., 2009; Schwarz & Taylor, 2005).

In the present study, attention during the relaxation protocol was inadequately controlled for with the generalized instruction to "close your eyes, focus attentively on the music and relax as deeply as you can". One major advantage of the Duke binaural beat study was their use of beta-frequency binaural beats (Lane et al., 1998); it is easier to control for attention during ergotropic activities than it is during trophotropic activities. The "Inverted-U" hypothesis shows us that as arousal decreases, attention broadens (Landers & Arent, 2010). As Lazzouni et al. (2010), Saupe et al. (2009) and Schwarz and Taylor (2005) demonstrated, attention plays a pivotal role in the perception of a binaural beat and the subsequent neural generation of an ASSR. Additionally, as trance theorist G. Rouget stressed, the individual must be willing to become absorbed in the trance state and must also have a specific aim as well as adequate intellectual preparation (Fachner & Rittner, 2011). Participants in the present study may not have been provided with a specific enough aim nor adequate intellectual preparation.

One way around this problem would be to recruit participants with a significant amount of meditative experience or other form of attentional training. This indirect form of control would allow for a greater degree of confidence that participants were actually able to follow instructions. Alternatively, participants could be fitted with a handheld device that cued them every five minutes to rate their level of arousal, ensuring that they

did not fall asleep. Obviously, this has the potential of detracting from the absorption of attention into the activity at hand, which is the real point of the relaxation exercise. Another way around the arousal/attentional control problem would be to provide participants with a generalized relaxation response training session at the beginning of the study and instruct them to elicit the response while focusing on binaural beats or a placebo-control tape.

It is important to note here that the Hemi-Sync® process is a training system, as are all forms of secular and non-secular meditative, hypnotic and yogic practice. The binaural beat tapes used in the present study were designed to increase theta power and hypnotic susceptibility, but the real purpose of binaural beat use is to facilitate long-term practice in altering states of consciousness at will (Atwater, 2004). Our subjects did not have multiple opportunities to train with binaural beats. Skip Atwater, head of TMI's research division, informs us that "passively listening to Hemi-Sync® binaural beats may not automatically engender a focused state of consciousness…we all maintain a psychophysiological momentum, a homeostasis that may resist the influence of binaural beats" (Atwater, 2004).

The findings of a recent EEG study of the effects of binaural beats on meditative ability are consistent with this claim (Lavallee et al., 2011). They may also explain the lack of significant findings during and immediately after the relaxation protocol utilized in the present study. Lavallee et al. (2011) studied novice and experienced mediators who listened to hindering (15 Hz) and facilitative (7 Hz) binaural beats during a 15-min mindfulness meditation (Lavallee et al., 2011). Only experienced meditators demonstrated increased theta power during meditation when listening to facilitative

binaural beats, while novice mediators did not. Recent advances in EEG technology would easily allow for cost-effective inclusion of a simplified EEG device to directly measure theta-band power in subsequent binaural beat studies.

Effects of Stress

At the end of each session, participants were asked to rate (on a Likert scale of 1-10) their level of relaxation during the relaxation protocol and their level of stress during the mental arithmetic stress test. While these are not empirically validated measures, it was a simple task; the findings were statistically significant and worth reporting. Participants in the binaural beat condition reported a 6.4% increase in perceived relaxation during the relaxation session and 11.5% less stress during the mental arithmetic stressor. This provides some support for the hypothesis that the binaural beats were effective in promoting perceived relaxation and mediating subsequent stress. As mentioned, other psychophysiological measures in the present study were not so supportive of these relaxation or stress-blunting effects. Out of all the physiological variables, only SYS-BP was increased significantly in response to the MA stressor. This finding is contrary to the literature, which cites robust physiological effects of the MA stressor (e.g., increased HR, BP and decreased HRV) (Sharpley et al., 2000).

Measuring subjective stress was limited due to the fact that participants were not asked to rate their subjective stress and relaxation immediately post-relaxation and post-stress, but instead were asked at the end of the study, after the mental arithmetic stress test. The effects of the stress may have biased responses. There is some precedent in the literature for using a Likert visual analog scale to measure perceived relaxation (Agras, Horne, & Taylor, 1982; Lee et al., 2011). Future studies should consider using a visual

analog scale to assess perceived stress and relaxation immediately after stress and relaxation protocols.

Another glimmer of support for a beneficial effect of binaural beats on the post-exercise stress response was found in the time-domain HRV measures post-stress. Participants in the experimental condition demonstrated significantly higher NN50 measurements (potentially indicating increased vagal modulation). However, it is important to note that these findings may be suspect. There was also an approach towards significant differences in NN50 at baseline. It becomes unclear if the findings post-stress were a result of binaural beat exposure or an artifact of non-significant baseline variability. In any event, the NN50 findings represent only one variable in 13 psychophysiological measures that provided a bit of support for a binaural beat entrainment effect. Since NN50 typically covaries consistently with other time-domain HRV measures (e.g., SDNN, avRR, RMSSD), it is likely that the significant finding for NN50 is a Type 1 error and not supportive of a binaural beat entrainment effect.

At first it was thought that the lack of significant HRV findings may be a result of using an overly conservative artifact correction algorithm in the HRV data processing. This may have sacrificed true variance along with error variance, lowering the statistical power of the study. However, after completing the first round of data analysis, the HRV data were re-processed using a less conservative artifact correction algorithm and the results were unaffected. Despite the observed effect of binaural beats on self-reported perceived stress and relaxation and post-stress NN50, there was little additional evidence across other variables to support a positive impact of the treatment.

Limitations and Directions for Future Research

A major methodological limitation was that the present study lacked a true control group. This was largely due to a lack of resources and fear of attrition in a sample coming exclusively from a student population. Given that each session represented a 90-min time commitment, the decision was made to directly compare a binaural beat condition to a placebo condition to assess effects before taking on a larger scale study. As a follow-up, it may be interesting to reduce the time commitment of each session to 45 minutes or less (relaxation protocol plus mental arithmetic stress, excluding exercise) and including additional control conditions. This might allow for differentiation of potential entraining effects of the pink noise, carrier tones and binaural beats while also eliminating the hypothesized "wash-out" effect of execise.

A second limitation involved the decision to use the hand-held Cocoro sAA meter. It was subject to frequent error messages, ultimately resulting in the loss of sAA data for six participants. The large amounts of interpersonal and intrapersonal variability observed in this variable made it impossible to detect any real differences through statistical analysis with the present sample size. Previous research with the Cocoro sAA meter and traditional bench-top sAA methods reported baseline, exercise and post-exercise sAA values with standard deviations approximately equal to the mean (Nater & Rohleder, 2009; Rohleder & Nater, 2009; Shimazaki et al., 2008; Strahler et al., 2010). Due to the high variability in sAA measurements (along with the tendency for the sAA data to be positively skewed), it has been suggested that data distributions be normalized through the use of logarithmic or square root transformations (Rohleder & Nater, 2009). These transformations were not performed in the present study. It is also worth

mentioning that the measures taken to reduce variability (e.g., no teeth-brushing, gum-chewing, drinking, eating, *etc*.) may not have been adhered to in the student sample. Future binaural beat research should omit this variable or consider using traditional bench-top methods of sAA analysis.

Methodological limitations aside, this study made contributions to the limited research on the psychophysiological effects of binaural beats and the effectiveness of frequency-entrainment. It is clear that simply listening to theta-frequency binaural beats for 20-min post-exercise was not effective in promoting a significantly greater relaxation response in a student population. This may be a result of a near-maximal post-exercise trophotropic response negating any effect of binaural beat entrainment; however, consumers should be wary about grandiose claims made by some companies marketing binaural beat products. Given the findings of previous research and the limited findings of the present study, empirical study of the causal mechanisms underlying tempo and frequency-entrainment using sound, as well as any potential psychophysiological or behavioral effects, should not be abandoned.

CHAPTER 6

SUMMARY, CONCLUSIONS and RECOMMENDATIONS

Summary

Binaural beats are auditory illusions experienced by the listener when pure tones of similar frequencies are presented separately to each ear via stereo headphones. There is evidence to support the auditory-driving hypothesis, which holds that neural firing patterns (and thus psychological state) can be entrained through exposure to rhythmic auditory stimuli. Binaural beats, if effective as entraining stimuli, would operate through the principle of frequency-entrainment. While tempo-entrainment is supported in the literature, frequency-entrainment remains controversial. There is evidence to support the role of binaural beats in both trophotropic (decreasing arousal) and ergotropic (increasing arousal) state induction. However, the studies that generated this evidence failed to isolate the binaural beats from other potentially entraining stimuli (e.g., music, verbal guidance, nature sounds). Research from the exercise science literature has recently demonstrated evidence that a combined exercise and relaxation training protocol may be superior to either approach alone. Given the secular nature of binaural beats, it seemed logical to evaluate their effectiveness in promoting a relaxation response in a combined exercise-relaxation framework. Therefore, the present study examined the effects of binaural beats, combined with only pink noise (sound over the entire audible frequency range), on the post-exercise relaxation response and subsequent stress response.

College-aged students (n = 21), free from any medical diagnoses or substance use, completed two experimental sessions in which baseline, post-exercise, post-relaxation and post-stress measures were obtained (HR, BP, sAA, PA & NA and HRV measures).

Participants exercised for 20-min at 70% of their predicted VO_{2max}, with a 5-min warm-up and cool-down at 50% of their predicted exercise workload. After exercising, participants listened to either binaural beats or a placebo control while relaxing alone in a quiet, low-light environment. Finally, participants completed a short, timed 20-question mental arithmetic battery designed to elicit a stress response. Participants returned within a two-week period to complete the alternate condition.

A series of *t*-tests and 2x4 repeated measures ANOVAs (Condition x Time) were performed to assess main effects and interaction effects. Significant main effects of Time were found for all variables of interest except for sAA and DBP ($p < .05$) due to the effects of exercise. There were no significant main effects of Condition nor were there any significant interaction effects. Paired sample *t*-tests showed no significant differences in any measure of HRV during the 20-min relaxation protocol. Paired-sample *t*-tests showed a significant difference in reported stress and relaxation, with the binaural beat group reporting significantly less stress and more relaxation. However, given the lack of empirical validation of the measures through which subjective stress and relaxation were assessed, these results must be interpreted cautiously. Theta-frequency binaural beats did not appear to measurably affect the post-exercise relaxation response or attenuate a subsequent stress response. Future research in a more homogenous sample of trained individuals focusing on the effects of binaural beats on the facilitation of a relaxation response from an ordinary state of consciousness is warranted.

Conclusions

Based on the findings of the study, the following conclusions were drawn:

1. Theta-frequency binaural beats failed to significantly affect the depth of a 20-min post-exercise relaxation session in comparison to a placebo-control audio tape, as measured through HRV variables.
2. Theta-frequency binaural beats failed to significantly facilitate post-exercise recovery in comparison to a placebo-control audio tape, as measured through psychophysiological variables. However, theta-frequency binaural beats did increase subjective ratings of perceived relaxation.
3. Theta-frequency beats failed to significantly affect a post-relaxation stress response in comparison to a placebo-control audio tape, as measured through psychophysiological variables. However, theta-frequency binaural beats did decrease subjective ratings of perceived stress.

Recommendations

The results of the present study do not support the role of binaural beats in facilitating a post-exercise relaxation response or mediating a subsequent stress response. However, increases in subjective ratings of relaxation and decreases in subjective ratings of perceived stress, along with some evidence for increased vagal modulation in the binaural beat group post-stress, warrants further research and consideration of methodological improvements. Recommendations for future areas of research include the following:

1. Allow the use of more diverse empirically validated psychometric measures (e.g., the Trier Social Stress Test, Visual analog relaxation/stress scales).
2. To better isolate the effects of binaural beats, avoid the use of exercise/ergotropic activities.

3. Include a true control group (i.e., no audio) to better gauge the relative effects of a binaural beat group and a placebo control group.
4. Use benchtop sAA measures rather than a handheld dry-chemistry device which provides rapid but questionable results.
5. Include relaxation-response training during orientation or a trained sample with meditative experience to more adequately control for attentional factors.
6. Incorporate simplified EEG technology to assess alterations in frequency-band power due to binaural beat treatment.

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APPENDIX A

RECRUITMENT SCRIPT

I am conducting a research project examining the relationship between exercise, stress, music and relaxation and would like to ask for volunteers to participate. I hope that your involvement in this study will teach you something positive about research and may provide you with novel ideas about stress management.

Participation will require attendance at two hour and a half sessions scheduled at the same time of day within a two-week period. During the course of the study, if you decide that you do not want to continue, you may leave at any time. Your participation is voluntary and you may withdraw without penalty.

If you volunteer to participate in the study you will be asked to perform moderate exercise on a treadmill for 20 minutes. You will then be asked to practice relaxing while listening to music. This study will require wearing a heart rate monitor chest-strap and will also require saliva samples, which will involve you placing a test-strip in your mouth, under your tongue, for 30 seconds. Lastly, you will need to refrain from eating or drinking (anything but water) for one hour prior to the session, brushing your teeth with toothpaste or consuming caffeine for three hours prior and avoiding alcohol, medication and exercise for 24 hours prior.

You must be at least 18 years of age to participate. You should not participate if you are currently taking any form of contraceptive; have any history of epilepsy or diagnosis of metabolic, cardiovascular, or psychiatric disorders, substance dependency (tobacco, alcohol, or other illicit drugs), auditory disorders or hearing loss, or present musculoskeletal injury that would interfere with your ability to do the treadmill exercise.

If you are interested in participating, please put your name and email address on the form that I will be passing around. Data collection times will be from 1-3 and 3-5, Monday through Friday and 11-1 also on Tu/Th. Please check the time slots that would work best for your schedule. **Keep in mind that participation will require you attending *two* sessions, scheduled at the same time of day, within a two-week period.** For example, if you sign up for Monday at 1pm, you will need to come back the following Monday at 1pm.

APPENDIX B

INFORMED CONSENT

1. **Purpose of the Study**
   The purpose of this study is to examine the relationship between cardiovascular exercise, music, stress and relaxation.
2. **Benefits of the Study**
   a. You will learn about the relationship between stress and relaxation and will get practice in learning how to voluntarily elicit the relaxation response, using music as a tool to do so. Learning how to effectively manage stress is an invaluable skill for college students to master and participation in this study can help you on your way.
   b. The beneficial effects of cardiovascular exercise and relaxation training are well known, but few studies have examined their effects when combined with each other. Preliminary evidence has provided support for this combined framework in reducing blood pressure responses to stress and further research using multiple variables is warranted.
3. **What You Will Be Asked to Do**
   a. *Amount of time it will take*:
      i. Participation in this study will require approximately 3 hours of your time; 1.5 hours each for two experimental sessions scheduled at the same time of day within a 2-week period.
   b. *Tasks and procedures*:
      i. You will be asked to exercise on a treadmill for 20 minutes at a moderate pace. You will then be asked to relax for 20 minutes in a quiet room while listening to music. Measurement of this study's variables will require you wearing a heart rate monitor chest strap. For this, women will need to wear a sports' bra underneath a t-shirt. We will also collect periodic saliva samples by having you place a test-strip under your tongue for about a minute. Lastly, you will be asked to perform some mental math problems.
   c. *Exclusionary criteria*:
      i. You should not participate if you take any form of contraceptive; have any history of epilepsy or diagnosis of metabolic, cardiovascular, or psychiatric disorders, substance dependency (tobacco, alcohol, or other illicit drugs), auditory disorders or hearing loss, or present musculoskeletal injury that would interfere with your ability to perform the treadmill exercise.
4. **Risks:** During exercise there is always a risk of musculoskeletal injury. For participants meeting the inclusion criteria, this risk will be minimal. Listening to the music we play might pose a risk to individuals who have a history of epilepsy,

APPENDIX B (continued)

5. cardiac arrhythmia, psychiatric, or auditory disorders and those persons should not participate in this study.
6. **Compensation for Injury:** If you suffer an injury that requires any treatment or hospitalization as a direct result of this study, the cost for such care will be charged to you. If you have insurance, you may bill your insurance company. You will be responsible to pay all costs not covered by your insurance. Ithaca College will not pay for any care, lost wages, or provide other financial compensation.
7. **If You Would Like More Information about the Study:** If you have any questions at any time during this study, please don't hesitate to ask the researcher. If you have questions or concerns prior to the start of the study or after the completion of the study, you may contact the primary researcher (Patrick McConnell) at E: pmcconn1@ithaca.edu, Cellular Phone: (862)266-7014 or my faculty advisor (Dr. Sforzo) at sforzo@ithaca.edu.
8. **Withdrawal from the Study:** You have the right to withdraw from this study at any point, for any reason, with no negative repercussions whatsoever. Simply notify the experimenter and they will give you a full debriefing and allow you to leave. You may also choose to leave any questionnaire items unanswered if they make you feel uncomfortable for whatever reason, however your full participation is encouraged.
9. **Confidentiality:** Your name will not appear on any of the data or any of the forms used in this study, nor will it be connected with any information you provide. You will be identified solely by your Student ID number (first six digits only). After you submit your responses there will be no way for anyone to identify you with your responses. All the information you provide will be confidential. All data will be stored in a locked filing cabinet at the researcher's private residence and will be retained for a period of 10 years, after which it will be destroyed.

I have read the above and I understand its contents. I agree to participate in the study. I acknowledge that I am 18 years of age or older.

_____

Print or Type Name

_____ _____

Signature Date

APPENDIX C

PRE-PARTICIPATION SCREENING QUESTIONNAIRE (PPSQ)

1st six digits of Student ID Number: _____
Date:_____

Date of Birth: _____ Age: _____ Sex: M / F
Please complete this form to the best of your knowledge.

	Yes	**No**	
1.	0	0	Have you had a heart attack, stroke, or heart surgery?
2.	0	0	Has your doctor said that you have cardiovascular, pulmonary, metabolic or other significant disease?
3.	0	0	During or right after exercise, do you have pains or pressure in the chest area, neck, shoulder or arm?
4.	0	0	Have you experienced any unusual leg pain upon exertion?
5.	0	0	Has your doctor said that you have a heart murmur or irregular heart beat (arrhythmia)?
6.	0	0	Do you have insulin-dependent diabetes or take medication to control your blood sugar?
7.	0	0	Do you experience shortness of breath at rest or with mild exertion?
8.	0	0	Has your doctor said you have high blood pressure ($\geq$ 140/90) or are you on medication for your blood pressure?
9.	0	0	Do you experience dizziness/fainting spells at rest or with exertion?
10.	0	0	Are you currently pregnant or within six weeks postpartum?
11.	0	0	Are you currently taking any contraceptives (i.e. oral, patch)?
12.	0	0	Are you currently taking prescription medication for a medical condition? (i.e. heart, lung, GI, blood, nervous system, psychiatric, or other medical condition)
13.	0	0	Do you have a chronic or acute orthopedic or other health condition that you or your physician feel will be affected by exercise? (i.e. bursitis, arthritis, osteoporosis, neck or back injury, past surgery, *etc*.)
14.	0	0	Do you have a medical condition not mentioned here, which might affect your ability to participate in an exercise program? (i.e. seizures, epilepsy, emphysema, asthma, *etc*.)
15.	0	0	Does anyone in your family have a history of epilepsy or seizures?
16.	0	0	Do you smoke or have you smoked within the last 6 months?
17.	0	0	Have you ever been diagnosed with a psychiatric disorder? (i.e., major depression, ADHD, bipolar disorder, anxiety, substance dependency, *etc*.)
18.	0	0	Do you consume more than 2 alcoholic drinks per day?

APPENDIX C (continued)

Do you have any exercise limitations not previously discussed? (i.e. recent injuries, *etc*.) **Y or N**

If yes, please explain: _____
_____

Please list any other pertinent health/medical information I should be aware of:
_____
_____

What is your ethnicity?
- o Black / African American
- o White / Caucasian
- o Asian
- o Hispanic / Latino
- o American Indian or Alaska Native
- o Native Hawaiian or Other Pacific Islander
- o Other

How much sleep do you regularly get each night?
- o Less than four hours
- o Four to six hours
- o Six to eight hours
- o More than eight hours

**Please note that one of the variables we will be measuring for this study, salivary α – amylase, is sensitive to hormonal changes. We ask that women take this into consideration when signing up for their first session, scheduling it at a point as close to the beginning of their menstrual cycle as possible.

I understand that the completion of this form will not result in any type of diagnosis of disease and that it is not intended as a substitute for consultation with my personal physician. I must consult my own personal physician for any evaluation of my health status.

I hereby certify that I have read and understand all questions on this health and exercise history questionnaire and that all questions have been answered truthfully to the best of my knowledge. I agree to notify Patrick McConnell, Primary Researcher, if there are any changes in the information that I have provided herein.

Signature_____ Date:_____

Witness_____ Date:_____

APPENDIX D

PHYSICAL ACTIVITY RATING (PA-R)

Select the number that best describes your overall level of physical activity for the previous 6
MONTHS:

0 = avoid walking or exertion; e.g., always use elevator, drive when possible instead of Walking
1 = **light activity:** walk for pleasure, routinely use stairs, occasionally exercise sufficiently to cause heavy breathing or perspiration
2 = **moderate activity:** 10 to 60 minutes per week of moderate activity; such as golf, horseback riding, calisthenics, table tennis, bowling, weight lifting, yard work, cleaning house, walking for exercise
3 = **moderate activity:** over 1 hour per week of moderate activity as described above
4 = **vigorous activity:** run less than 1 mile per week or spend less than 30 minutes per week in comparable activity such as running or jogging, lap swimming, cycling, rowing, aerobics, skipping rope, running in place, or engaging in vigorous aerobic-type activity such as soccer, basketball, tennis, racquetball, or handball
5 = **vigorous activity:** run 1 mile to less than 5 miles per week or spend 30 minutes to less than 60 minutes per week in comparable physical activity as described above
6 = **vigorous activity:** run 5 miles to less than 10 miles per week or spend 1 hour to less than 3 hours per week in comparable physical activity as described above
7 = **vigorous activity:** run 10 miles to less than 15 miles per week or spend 3 hours to less than 6 hours per week in comparable physical activity as described above
8 = **vigorous activity:** run 15 miles to less than 20 miles per week or spend 6 hours to less than 7 hours per week in comparable physical activity as described above
9 = **vigorous activity:** run 20 to 25 miles per week or spend 7 to 8 hours per week in comparable physical activity as described above
10 = **vigorous activity:** run over 25 miles per week or spend over 8 hours per week in comparable physical activity as described above

Source: Bradshaw, D. I. (2003). *An accurate v02max non-exercise regression model for 18 to 65 year-old adults.* Master of Science, Brigham Young University.

Modified from Jackson et al., (1990). Prediction of functional aerobic capacity without exercise testing. *Medicine and Science in Sports and Exercise, 22*(6), 863-870.

APPENDIX E

PERCEIVED FUNCTIONAL ABILITY (PFA)

Suppose you were going to exercise continuously on an indoor track for **1 mile**. Which exercise pace is just right for you—not too easy and not too hard?

Circle the appropriate number (any number, 1 to 13).

1. Walking at a *slow* place (18 minutes per mile or more)
2.
3. Walking at a *medium* pace (16 minutes per mile)
4.
5. Walking at a *fast* pace (14 minutes per mile)
6.
7. Jogging at a *slow* pace (12 minutes per mile)
8.
9. Jogging at a *mediu*m pace (10 minutes per mile)
10.
11. Jogging at a *fast* pace (8 minutes per mile)
12.
13. Running at a *fast* pace (7 minutes per mile)

How fast could you cover a distance of **3-miles** and NOT become breathless or overly fatigued? Be realistic.

Circle the appropriate number (any number, 1 to 13).

1. I could walk the entire distance at a *slow* pace (18 minutes per mile or more)
2.
3. I could walk the entire distance at a *mediu*m pace (16 minutes per mile)
4.
5. I could walk the entire distance at a *fast* pace (14 minutes per mile)
6.
7. I could jog the entire distance at a *slow* pace (12 minutes per mile)
8.
9. I could jog the entire distance at a *mediu*m pace (10 minutes per mile)
10.
11. I could jog the entire distance at a *fast* pace (8 minutes per mile)
12.
13. I could run the entire distance at a *fast* pace (7 minutes per mile or less)

Source: Bradshaw, D. I. (2003). *An accurate v02max non-exercise regression model for 18 to 65 year-old adults.* Master of Science, Brigham Young University.
Modified from George et al., (1997). Non-exercise VO2max estimation for physically active college students. *Medicine and Science in Sports and Exercise, 29*(3), 415-423.

APPENDIX F

DEBRIEFING STATEMENT

Thank you for your participation in this study. My primary aim was to investigate the efficacy of theta-frequency (4-7 Hz) binaural beats in facilitating the post-exercise relaxation response. Binaural beats are an auditory illusion experienced when two or more pure tones of variant frequencies are presented dichotically. Theoretically, these beats, when presented to a listener in frequency ranges corresponding to human brainwaves, can help to facilitate entry into different states of consciousness (i.e. relaxation, alertness, sleep, creativity, *etc*).

Additionally, I wanted to examine the beneficial effects of exercise combined with relaxation training on stress reactivity and specifically whether or not binaural beats improved this benefit. I hope that you enjoyed your participation in this study and have learned something about your ability to produce states of increased alertness and relaxation in yourself. Stress-management is a vital skill for college students to master and hopefully this study has kindled an interest in you to cultivate your relaxation skills.

Contact information:

If you have any concerns or questions, please don't hesitate to contact me:

Patrick McConnell

(862)266-7014

pmcconn1@ithaca.edu

Provided below is the contact information for Ithaca College's Center for Counseling and Psychological Services if you experience any negative repercussions from participating in this study.

Business hours: 8:30 am to 5:00 pm, Monday through Friday.

Phone Number: (607)274-3136

APPENDIX G

DAVID BRIAN BRADY PROJECT

Brian Brady's project involves an attempt to encourage the development of theta brainwave frequencies. The study hypothesizes that increased theta activity will result in increased hypnotic susceptibility. The stereo presentation of pink sound (white noise equalized for human hearing) mixed with wide-band theta binaural beating is to be used as a stimulus environment to induce the development of theta brainwaves. Results using other tape formats not containing binaural beats will be compared. The sensation of auditory binaural beating occurs when two coherent sounds of nearly similar frequencies are presented one to each ear with stereo headphones or speakers. Originating in the brainstem's superior olivary nucleus, the site of contralateral integration of auditory input (Oster 1973), the audio sensation of binaural beating is neurologically conveyed to the reticular formation[1] (Swann et al. 1982) and the cortex where it can be observed as a frequency-following response. A cortical frequency-following response to auditory stimulus has been objectively demonstrated in various studies (Oster 1973; Smith, Marsh, & Brown 1975; Marsh, Brown & Smith 1975; Smith et al. 1978; Hink et al. 1980). The frequency-following response provides proof that the sensation of binaural beating has a neurological efficacy. The binaural beats are designed to influence ongoing brainwave states by providing *information* to the brain's reticular activating system (RAS). If internal stimuli, feelings, attitudes, beliefs and external sensory stimuli are not in conflict with this *information*, the RAS will alter brainwave states to match the binaural-beat provocation; in this case a theta state of increased hypnotic susceptibility.

The following audiocassette tapes, prepared for Brian Brady's use, consist of pink sound as a format. The experimental tape was imbedded with audio tones or *carriers* that gave rise to a binaural beat being perceived by the listener. The carrier tones on the tapes were changed periodically in an attempt to encourage vigilance on the part of the listener. Stimulus sequence for experimental tape

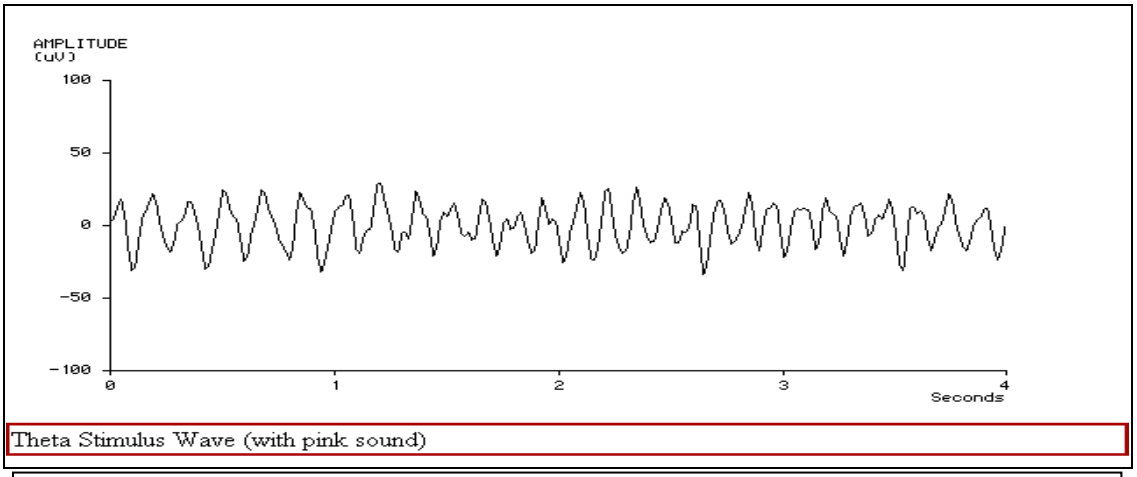
[1] The word reticular means "net-like" and the neural reticular formation itself is a large, net-like diffuse area of the brainstem (Anch et al. 1988). The RAS regulates cortical EEG (Swann et al.1982) and controls arousal, attention and awareness – the elements of consciousness itself (Tice & Steinberg 1989; Empson 1986). How we interpret, respond and react to *information* (internal stimuli, feelings, attitudes and beliefs as well as external sensory stimuli) is managed by the brain's reticular formation stimulating the thalamus and cortex and controlling attentiveness and level of arousal (Empson 1986).

Time	Binaural Beat	Carrier Tones
0:00 - 3:00 min.	*wide-band theta*	C-Major-7th (292 Hz, 330 Hz, 392 Hz, 466 Hz)
3:00 - 5:00 min.	*wide-band theta*	C-Major (292 Hz, 330 Hz, 392 Hz, 523 Hz)
6:00 - 10:00 min.	*wide-band theta*	G-Major (196 Hz, 247 Hz, 294 Hz, 392 Hz)
10:00 - 15:00 min.	*wide-band theta*	D-Minor (294 Hz, 349 Hz, 440 Hz)
15:00 - 20:00 min.	*wide-band theta*	C-Major (292 Hz, 330 Hz, 392 Hz, 523 Hz)

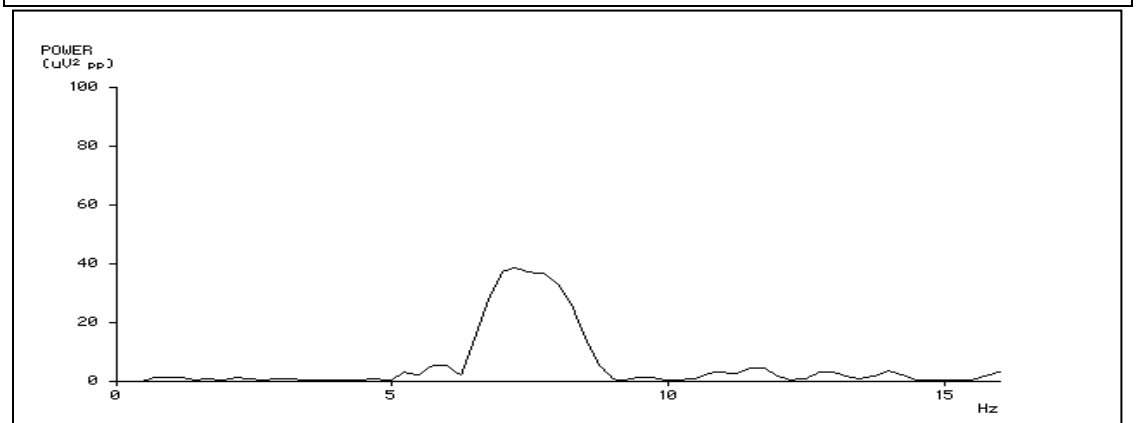
The *wide-band theta* effect for the experimental tape was generated by continuously (smoothly) varying a 7 Hz (theta) left-right frequency difference by plus or minus 1.5 Hz over a period of four seconds. The theta stimulus therefore changed continuously from 5.5 Hz to 8.5 Hz and back to 5.5 Hz over a period of four seconds. Each carrier looped for the above designated time periods. The carrier tones changed from one to another in seamless ten-second cross fades.

Instrumentation was used to objectively display the binaural-beat components imbedded in the experimental tape. This instrumentation was designed to detect right-left differences in stereo audio signals and, through the use of the NRS-24's FFT utility, display frequency spectra (0 Hz to 16 Hz) of these audio differences. Of interest here were the coherent audio signals that would give rise to the desired auditory phenomena known as the binaural beat. Instrumentation revealed clear evidence of the imbedded binaural-beat component on the experimental tape. Strictly speaking, the included graphs do not display *binaural beats* because binaural beats are defined as an auditory phenomenon. Listeners throughout the experimental tape can readily hear a binaural beat.

APPENDIX G (continued)



Theta Stimulus Wave (with pink sound)



Wide-Band Theta Stimulus (with pink sound)

Two additional tapes were prepared. One (training) tape was recorded with pink sound only. The other (control tape) contained pink sound and the same tones as the experimental tape but without binaural beats. The tapes were labeled so as not to reveal which was the experimental tape and which were the alternate tapes. Following was the label coding:

The tape labeled *George* is the experimental tape with pink sound and theta binaural beats.
The tape labeled *Raymond* is the one with pink sound and tones without binaural beats.
The tape labeled *Alfred* is the one with pink sound only.

APPENDIX G (continued)

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APPENDIX H

MENTAL ARITHMETIC PROBLEMS

MA Sequence A

1. 23 + 93 - 47 = 69
2. 31 – 95 + 68 = 4
3. 99 – 78 + 43 = 64
4. 62 – 80 + 45 = 27
5. 95 + 94 – 75 = 114
6. 79 – 37 + 52 = 94
7. 96 + 95 – 94 = 97
8. 48 – 24 + 47 = 71
9. 43 + 21 – 57 = 7
10. 24 + 83 – 55 = 52
11. 28 – 39 + 46 = 35
12. 92 – 39 + 12 = 65
13. 27 + 87 - 49 = 65
14. 17 + 52 – 32 = 37
15. 76 – 13 + 23 = 86
16. 77 – 64 + 89 = 102
17. 55 + 85 – 36 = 104
18. 76 – 23 + 94 = 147
19. 81 + 95 – 18 = 158
20. 59 + 89 – 56 = 92

APPENDIX H (continued)

MA Sequence B

1. 52 + 76 – 69 = 59
2. 16 + 40 – 20 = 36
3. 60 – 50 + 78 = 88
4. 26 + 30 – 47 = 9
5. 83 – 40 + 99 = 142
6. 83 – 46 + 96 = 133
7. 13 + 50 – 60 = 3
8. 31 + 60 – 75 = 16
9. 83 – 10 + 31 = 104
10. 51 - 15 - 95 = 131
11. 29 + 84 – 86 = 27
12. 84 – 11 + 73 = 146
13. 34 – 20 + 52 = 66
14. 75 – 18 + 20 = 77
15. 25 + 83 – 19 = 89
16. 19 + 80 – 10 = 89
17. 76 + 79 – 18 = 137
18. 71 – 24 + 11 = 58
19. 80 – 33 + 31 = 78
20. 59 – 44 + 57 = 72

APPENDIX I

MENTAL ARITHMETIC ANSWER SHEET

Please listen to the following mental arithmetic problems and do your best to answer correctly as many as possible.

1. _____
2. _____
3. _____
4. _____
5. _____
6. _____
7. _____
8. _____
9. _____
10. _____
11. _____
12. _____
13. _____
14. _____
15. _____
16. _____
17. _____
18. _____
19. _____
20. _____

APPENDIX J

POSITIVE AND NEGATIVE AFFECT SCHEDULE - TRAIT (PANAS-T)

This scale consists of a number of words that describe different feelings and emotions. Read each item and then list the number from the scale below next to each word.

Indicate to what extent you have felt this way over THE PAST WEEK:

1	2	3	4	5
Very Slightly or Not at All	A Little	Moderately	Quite a Bit	Extremely

_____ 1. Interested	_____ 11. Irritable
_____ 2. Distressed	_____ 12. Alert
_____ 3. Excited	_____ 13. Ashamed
_____ 4. Upset	_____ 14. Inspired
_____ 5. Strong	_____ 15. Nervous
_____ 6. Guilty	_____ 16. Determined
_____ 7. Scared	_____ 17. Attentive
_____ 8. Hostile	_____ 18. Jittery
_____ 9. Enthusiastic	_____ 19. Active
_____ 10. Proud	_____ 20. Afraid

Source: Watson, D., Clark, L. A., & Tellegen, A. (1988). Development and validation of brief measures of positive and negative affect: The panas scales. *Journal of Personality and Social Psychology, 54*(6), 1063.

APPENDIX J (continued)

POSITIVE AND NEGATIVE AFFECT SCHEDULE – STATE (PANAS-S)

This scale consists of a number of words that describe different feelings and emotions. Read each item and then list the number from the scale below next to each word.

Indicate to what extent you feel this way RIGHT NOW, that is, at the PRESENT MOMENT:

1	2	3	4	5
Very Slightly or Not at All	A Little	Moderately	Quite a Bit	Extremely

_____ 1. Interested	_____ 11. Irritable
_____ 2. Distressed	_____ 12. Alert
_____ 3. Excited	_____ 13. Ashamed
_____ 4. Upset	_____ 14. Inspired
_____ 5. Strong	_____ 15. Nervous
_____ 6. Guilty	_____ 16. Determined
_____ 7. Scared	_____ 17. Attentive
_____ 8. Hostile	_____ 18. Jittery
_____ 9. Enthusiastic	_____ 19. Active
_____ 10. Proud	_____ 20. Afraid

Source: Watson, D., Clark, L. A., & Tellegen, A. (1988). Development and validation of brief measures of positive and negative affect: The panas scales. *Journal of Personality and Social Psychology*, *54*(6), 1063.

APPENDIX K

POST-PARTICIPATION SCREENING QUESTIONNAIRE (POST-PSQ)

1. I am familiar with binaural beat technology.

1	2	3	4	5	6	7
Not at all true	Not very true	Moderately not true	Somewhat true	Moderately true	True	VeryTrue

2. I am experienced with meditation.

1	2	3	4	5	6	7
Not at all true	Not very true	Moderately not true	Somewhat true	Moderately true	True	Very True

3. I have heard of The Monroe Institute before.

1	2	3	4	5	6	7
Not at all true	Not very true	Moderately not true	Somewhat true	Moderately true	True	Very True

4. I am an experienced musician.

1	2	3	4	5	6	7
Not at all true	Not very true	Moderately not true	Somewhat true	Moderately true	True	Very True

5. I was able to tell the difference between the music in each experimental session.

1	2	3	4	5	6	7
Not at all true	Not very true	Moderately not true	Somewhat true	Moderately true	True	Very True

6. The mental arithmetic test made me feel stressed out during the first session.

1	2	3	4	5	6	7
Not at all true	Not very true	Moderately not true	Somewhat true	Moderately true	True	Very True

7. The mental arithmetic test made me feel stressed out during the second session.

1	2	3	4	5	6	7
Not at all true	Not very true	Moderately not true	Somewhat true	Moderately true	True	Very True

8. I liked the effects of exercise followed by relaxation training.

1	2	3	4	5	6	7
Not at all true	Not very true	Moderately not true	Somewhat true	Moderately true	True	Very True

9. I felt equally relaxed by the music in each session.

1	2	3	4	5	6	7
Not at all true	Not very true	Moderately not true	Somewhat true	Moderately true	True	Very True

APPENDIX L

24-HOUR HISTORY QUESTIONNAIRE

1. Have you eaten any food or drank anything (except water) within the last hour? ○ Yes ○ No
2. Have you consumed any alcohol within the last 24 hours?
3. Have you taken any medication within the last 24 hours? ○ Yes ○ No
4. Have you engaged in strenuous exercise within the last 24 hours? ○ Yes ○ No
5. Have you consumed any caffeine within the last 3 hours? ○ Yes ○ No
6. Did you brush your teeth with toothpaste within the last 3hrs? ○ Yes ○ No
7. Are you experiencing any unusual stress in your life at the moment? ○ Yes ○ No
8. How much sleep did you get last night? _____
9. At what time did you wake up? _____
10. Is this a normal amount of sleep for you? ○ Yes ○ No