Learning to Relax: The Impact of Brief Biofeedback Training on Salivary Cortisol Reduction

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LEARNING TO RELAX: THE IMPACT OF BRIEF BIOFEEDBACK TRAINING ON
SALIVARY CORTISOL REDUCTION

A Master’s Thesis

Presented to

The Graduate College of

Missouri State University

In Partial Fulfillment

Of the Requirements for the Degree

Master of Science, Psychology

By

Dallas Robinson

August 2019
LEARNING TO RELAX: THE IMPACT OF BRIEF BIOFEEDBACK TRAINING ON
SALIVARY CORTISOL REDUCTION

Psychology

Missouri State University, August 2019

Master of Science

Dallas Robinson

ABSTRACT

Chronic stress has been linked to several health related and psychological problems. There is evidence that relaxation techniques can be useful in the reduction of stress and psychological complaints. Short-term interventions like computer-based biofeedback could provide a brief and independent way to manage stress and anxiety. The purpose of this study is to examine the impact of the ALIVE biofeedback program (a relatively new software that allows individuals to play a game requiring proper smoothness of breathing and heart rate) on the stress response after the Trier Social Stress Test, a laboratory stress task shown to elicit changes in cortisol levels found in participants' saliva samples. Those in the biofeedback condition had a significantly greater decrease in cortisol after the intervention and at the conclusion of the study.

KEYWORDS: biofeedback, salivary cortisol, stress response, relaxation technique, physiological psychology
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In the interest of academic freedom and the principle of free speech, approval of this thesis indicates the format is acceptable and meets the academic criteria for the discipline as determined by the faculty that constitute the thesis committee. The content and views expressed in this thesis are those of the student-scholar and are not endorsed by Missouri State University, its Graduate College, or its employees.
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I would like to thank my parents for their constant support during the course of my graduate studies. They never stopped believing in me and pushing me to the best person I could be, while still doing my best as a student. I would also like to thank my fiancé, who has been by my side through everything, and the many professors in the psychology department that helped me reach my full academic potential.
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INTRODUCTION

Balance is essential to a healthy lifestyle. College students deal with a variety of stressors, and often struggle with stress and mental health problems (Lipson et al., 2015). Chronic stress has been linked to several health related and psychological problems, and can make maintaining a physiological balance difficult (Carroll & Winslow, 2017). Currently, research is lacking in short term strategies for relaxation and stress reduction that could be useful for college students, as they typically lead very busy lives.

Cortisol and the Stress Response

Self-regulation is the integrative functioning of emotional and physiological regulation to maintain or reestablish homeostasis, where the body’s physiological systems are all at their optimal levels (Sapolsky, 2004). This can be measured through the functioning of the autonomic nervous system (ANS), which is responsible for regulating bodily systems like respiration and heart rate (Crockett, Gill, Cashwell, & Myers, 2017). Maintaining a physiological balance is essential to maintaining good health, and many things, including prolonged activation of the stress response, can disrupt homeostasis (Sapolsky, 2004). The stress response can be defined as a network of physiological responses in reaction to a threat/stressor (any physical or psychological factor that threatens homeostasis) (Crockett et al., 2017; Sapolsky, 2004). A psychological stressor affects a person’s physiology by activating specific cognitive and affective processes even when there is no physiological reality, for example, the stress associated with anticipating speaking in public (Sapolsky, 2004). The thalamus and prefrontal cortex first evaluate sensory information and send emotional responses via the limbic system (Dickerson &
Kemeny, 2004). The limbic system connects to the hypothalamus and serves as the primary pathway to activate the HPA axis, which is essential for supporting normal physiological functions and regulating other systems (Dickerson & Kemeny, 2004). This can lead to an increase in levels of several hormones including cortisol.

Cortisol is a glucocorticoid hormone released when stimuli interpreted as a potential physical or psychological threat activates the HPA axis (Byrd-Craven, Auer, Granger, & Massey, 2012). It plays an important role in maintaining homeostasis by mobilizing energy resources and providing fuel to the body (Dickerson & Kemeny, 2004). Prolonged cortisol activation or chronic dysregulation of cortisol (often referred to as chronic stress) has been shown to have detrimental long-term effects on both physical and mental health (Dickerson & Kemeny, 2004; Raison & Miller, 2003).

Chronic stress has been linked to several health-related problems in areas of fine motor performance, attention, and cognitive functioning (Carroll & Winslow, 2017). When energy is constantly mobilized by the stress response, fatigue occurs because the body is unable to store energy (Sapolsky, 2004). This can be even more detrimental when the stress-response is activated due to an imagined threat or a psychological stressor as opposed to a truly threatening situation or stressor. Stress and anxiety can be accompanied by physiological symptoms such as increased heart rate, sweating, and rapid breathing, as well as distress and an inability to function normally (Tabachnick, 2015).

Research suggests that stressors characterized by social evaluative threat (situations in which one’s social self is threatened by potentially negative feedback) elicit changes in cortisol levels (Dickerson & Kemeny, 2004; Dickerson, Myceck, & Zaldivar, 2008; Zoccola, 2018). Two studies found that participants in a social evaluative threat condition, in which participants had to
deliver a speech in front of an evaluative audience, had greater cortisol responses to the speech stressor compared to those in the non-social evaluative stressor condition (Dickerson et al., 2008; Zoccola, 2018). No effect was shown for increasing the difficulty or cognitive load of the stressor (Zoccola, 2018). Those in a condition giving a speech in front of an inattentive confederate also showed no change in cortisol levels, indicating social presence alone is not enough to change cortisol levels (Dickerson et al., 2008). However, being videotaped can also affect cortisol levels indicating that perceived negative social evaluation is a key aspect of the stressor affecting physiological responses like salivary cortisol levels (Dickerson et al., 2008).

Dickerson and Kemeny’s 2004 meta-analytic review of 208 laboratory stressor studies found that stressors with social-evaluative threat were associated with larger changes in cortisol compared to only verbal (e.g., public speaking) or cognitive tasks (e.g., the Stroop task). A combined verbal/cognitive task combination such as the Trier Social Stress Test (Kirschbaum, Pirke, & Hellhammer, 1993) had the largest effect size, indicating this type of task elicited greater cortisol changes (Dickerson & Kemeny, 2004). This response was even stronger when the social evaluative threat was paired with uncontrollability and also associated with a slower return of cortisol to baseline (Dickerson & Kemeny, 2004).

The Trier Social Stress Test (TSST) has been shown to stimulate the stress response in a research setting. This test has been found to increase levels of salivary cortisol to 2 to 4 elevations above baseline levels (Kirschbaum et al., 1993). The TSST consists of a ten-minute anticipatory period and a ten-minute test period where the participant delivers a free speech (5 minutes) and performs mental arithmetic (5 minutes) in front of an evaluative audience. When this test is administered, salivary cortisol has been shown to peak after ten minutes (Kirschbaum et al., 1993; Gordis, Granger, Susman, & Trickett, 2006). Cortisol levels began to decline after
they peaked, eventually returning to baseline 90 minutes after the start of the TSST (Kirschbaum et al., 1993).

One group that is more susceptible to chronic activation of the stress response is college students. Research shows that the duration of the stressor strongly influences its impact (Sternberg, Chrousos, Wilder, & Gold, 1992). College is a long-term commitment rife with various stressors such as exams, time demands, financial pressure, poor sleeping and eating habits, increased workload, and a fear of failure (Robotham & Julian, 2006). Stress also leads to increased risky behaviors and a decrease in academic performance (Robotham & Julian, 2006). Perceptions of stress and ability to cope determine an individual’s response to potentially stressful situations (McEwen, 1998). Because individuals may respond to the same stressful situations in different ways, an individual method for stress management would be ideal. Relaxation training is one way to manage stress, as well as resulting in a faster return to baseline cortisol levels (Van Rhenen, Blonk, van der Klink, van Kijk, & Schaufeli, 2005).

**Biofeedback**

The purpose of relaxation skills training is to learn effective skills in order to produce the relaxation response, a set of physiological changes that are the opposite of the stress response (Anstead, 2009). Stress may be inevitable, especially in the life of a student, but learning to monitor and regulate one’s response to stress through effective relaxation techniques may increase one’s ability to maintain homeostasis and overall health, as well as reducing chronic stress. Stress management can be defined as the ability to reduce or cope with stressors by finding a way to control the frequency, intensity, and duration of the stress reaction (Girdano, Everly, & Dusek, 1997). Regular practice of relaxation can also increase one’s resiliency to
stress, as shown in studies by Hoffman, Benson, and Arns (1982) and Lehmann, Goodale, and Benson (1986), by reducing responsiveness to the stress hormone norepinephrine after four to six weeks of daily relaxation practice (1982; 1986).

Most relaxation techniques that aim to reduce the stress response involve breathing. While deep, paced breathing is an effective relaxation technique, it has several limitations. For example, it often lacks proper physiological assessments to ensure one is breathing correctly (Reiner, 2008). Biofeedback is one relaxation technique that provides direct and immediate physiological feedback, bringing about an increased awareness of one’s internal state (Anstead, 2009). This technique has shown efficacy in helping individuals regulate ANS functioning, as well as learning how to become aware of and eventually control physiological processes like muscle tension, breathing, and heart-rate variability that are normally considered automatic (Frank, 2010; Ratanasiripong, Kaewboonchoo, Ratanasiripong, Hanklang, & Chumchai, 2015).

Peripheral biofeedback provides feedback based on activity in the body and can be used to target the physiological symptoms associated with stress and anxiety through a feedback loop in which the individual can use information about his or her experience to promote self-regulation (Crocket et al., 2017). Individuals often lose the ability to cope effectively after long-term exposure to daily hassles or stress (Kotozaki, 2014), and biofeedback is a technique that could help improve individuals’ ability to handle stress in the moment. Biofeedback typically consists of a visual display of heart rate and breathing rate, which could eventually teach individuals to control those processes and improve self-regulation.

Biofeedback has three stages: 1) acquiring awareness of the maladaptive physiological responses 2) learning to control responses using techniques like deep breathing and passive muscle relaxation 3) transferring that control to everyday life (Carroll & Winslow, 2017). This
technique has been shown to be effective in reducing stress associated with everyday hassles and anxiety (Kotozoki, 2014; Carroll & Winslow, 2017). In Carroll and Winslow’s 2017 study, a condensed 90-minute biofeedback training method was used incorporating diaphragmatic breathing and stress inoculation training. The experimental group was compared to a control group using physiological measures of the ANS response and cortisol measures of the stress response, as well as perceived stress levels using the state portion of the State Trait Anxiety Inventory. Those in the biofeedback condition had a statistically significant reduction in cortisol from pre-training to post-training (Carroll & Winslow, 2017).

In another study, participants in the biofeedback condition using a diaphragmatic breathing technique experienced a significant reduction in cortisol from pre-training to post-training, but the control group did not (Carroll & Winslow, 2017). Similarly, a four-week biofeedback intervention showed a significant reduction in perceived stress when compared to the control group (Ratanasiripong et al., 2015).

Heart rate variability (HRV), looking at the beat-to-beat changes in heart rate, is one aspect often evaluated in biofeedback. Low HRV has been linked to psychopathologies like anxiety disorders and substance use disorders during craving states (Henriques, Keffer, Abrahamson, & Horst, 2011; Thurstone & Lajoie, 2013). HRV biofeedback has been shown to reduce stress and mean salivary cortisol levels (Thurstone & Lajoie, 2013).

Research has suggested that computer-based biofeedback with HRV and breathing smoothness may be beneficial for stress management. The use of a computer-based biofeedback program significantly decreased levels of both state and trait anxiety in college students (Henriques et al., 2011). This biofeedback program used a finger pad to track the user’s heart rate and displayed a graph showing the user’s real time heart rate, where a smooth line indicated
relaxation. This intervention lasted four weeks and was individualized so the participants could practice biofeedback on their own.

There is a need for more in-depth research on computer-based biofeedback and its potential benefits, as well as understanding the efficacy of a short-term biofeedback intervention. One of the general guidelines for biofeedback is that participants need to take an active role in practicing, but there is still debate on the amount of time, method of biofeedback, and sessions necessary for efficacy (Carrol & Winslow, 2017).

There is also little research on the use of biofeedback with college students. Research has shown that biofeedback and relaxation training are effective tools to directly modify the stress response, as well as treating psychological disorders such as anxiety, depression, and addictions (Shannon, 2001). The constant stress that students often face has the potential to affect the immune system, allowing greater vulnerability to physical illness and increased mental distress (Sternberg et al., 1992). In one study focusing on the use of biofeedback with college students, the majority of students participated in a single session of biofeedback and relaxation training using Electromyography (EMG) (Anstead, 2009). The EMG detects and measures electrical activity in certain muscles, visually displaying the fluctuating voltage and providing feedback. Data revealed a significant difference pre- to post- EMG sessions, indicating a decrease in tension post-session (Anstead, 2009). This study also found that students reported feeling overwhelmed, feeling anxious, and having difficulty concentrating as the top three stress related symptoms. Another study using college students found that respiratory biofeedback significantly reduced state anxiety (Meier, 2013), providing further support for biofeedback as a stress management tool for college students.
The ALIVE biofeedback software (2019 Somatic Vision, Inc.) used in the current study visually displays the participants’ heart rate and breathing smoothness as well as a “game.” In one of the options, a car is shown in a race. The car only drives if the participant’s breathing is smooth and his or her heart rate is steady. The car slows down and eventually stops if there are too many irregularities. Some games involve stressful situations, e.g. obstacles where a car crashes into other cars, while others show environments like waves on a beach or a plant growing. This aspect of the ALIVE software provides the opportunity for participants to practice self-regulation during the biofeedback session when faced with excitement or stress about the different game options.

**Hypotheses**

In the current study, stress response recovery as measured by participant cortisol levels was examined in relation to the use of biofeedback gaming software to further understand the efficacy of computer-based biofeedback software used in a brief stress reduction intervention.

**Hypothesis 1.** It was predicted that participants would not vary in their cortisol response to the Trier Social Stress Test, and that the cortisol levels of all participants would peak at saliva collection time 2, ten minutes after completion of the stressor.

**Hypothesis 2.** It was also predicted that after the intervention, participants in the biofeedback condition would have a greater reduction in cortisol than those in the control condition.
METHOD

This study was approved on January 17, 2019 by the International Review Board (IRB-FY2019-380). Participants were recruited through psychology courses in a large mid-western university. They were instructed prior to the study to abstain from caffeine, alcohol, and tobacco two hours prior to the study due to potential impact on cortisol concentrations. Students received research credit or extra credit for their participation. The study was conducted in a research lab in the psychology building on campus. Participants were randomly assigned to a control condition, a condition teaching ALIVE biofeedback software, or a condition teaching yoga (used in a separate thesis project). See Figure 1 for a concise method flowchart.

For this study, there were two researchers participating, one acting as the judge and the other as administrator, collecting saliva and giving instructions. Several researchers, all trained, rotated roles of judge or administrator. All researchers were trained prior to data collection and used a script to ensure a standardized interaction between researchers and participants. The research room and observation room were connected by one-way glass, to ensure participants were not on their phones or drinking/eating during the study.

Upon arrival, participants were given an informed consent document explaining the study (see Appendix A). Saliva was collected immediately before beginning the Trier Social Stress Test (TSST) to establish a baseline stress response (measured by salivary cortisol levels). Participants were given ten minutes to prepare a speech alone as if on a job interview to build up anticipatory stress, followed by five minutes of evaluation during the participants speech and five minutes of evaluation during a mental mathematical activity where participants verbally counted backwards from 1051 by 7, and were told to start over if they made a mistake. The judge gave
the instructions during the TSST, and that was the only interaction the participant had with the judge. The purpose of a separate researcher acting as judge was to cause additional social evaluative threat, and to avoid attitudes towards the administrator affecting the stress response. Saliva was collected again ten minutes after the completion of the stressor (in order to allow time for cortisol to peak).

Participants were randomly assigned to one of three groups: the ALIVE biofeedback intervention, the yoga intervention, or the control condition for thirty minutes.

Participants in the ALIVE biofeedback condition were first educated on how to use the software. A workshop on ‘Ways to Breathe’ was selected for the participants. The researcher walked the participants through pursed lip breathing, belly breathing, and paced breathing. Each was explained, and the participant was given two minutes to practice deep breathing with the pacer, observing his or her heart rate and attempting to keep his or her breathing smooth. Participants then responded to prompts asking if they were comfortable with their breathing and asking if they felt lightheaded. The basic biofeedback education portion took approximately five minutes. Next, participants were shown the different interactive options available on the ALIVE biofeedback software. Participants all began with the “dream house” option displaying a house that builds only if participants’ breathing is smooth and their heart rates are steady. The researcher remained with the participant until the participant was comfortable with the biofeedback.

Another option was a car that only drove in a race if the participant kept his or her breathing paced and smooth. The car would slow, and the screen would eventually go dark and quiet if the participant did not. Participants were allowed to select different games for approximately 30 minutes and told to stay on the same game for at least five minutes to
maximize time using the biofeedback software. The control group watched a non-stress eliciting nature documentary for 30 minutes, and the yoga group watched and followed along to a 30-minute gentle yoga flow.

Saliva was collected after completion of the intervention. Participants were then given several surveys through Qualtrics; the order in which they were presented was counterbalanced. Saliva was collected for a final time 30 minutes after beginning the surveys. After each of the saliva collections, participants were offered a small drink of water. Approximately 30 minutes passed between each saliva sample collection. Participants were debriefed after giving the final saliva sample, the researcher explaining the TSST was designed to be stressful and evaluative as well as why saliva samples were taken. The saliva samples were labeled by participant number and time collected, and were then frozen at -20C until assayed for cortisol.

**Figure 1.** Method Flow Chart

**Participants**

Participants were undergraduate psychology students from a large Midwestern university. They were granted course credit for their participation. A total of 97 participants completed the study, and 10 were removed as outliers (see analyses section). Of these participants, there were 30 male participants and 57 female participants with a mean age of 19.85 (SD = 3.22).
Approximately 73.60% of participants self reported their racial/ethnic background as White, 6.90% as Black, 5.70% as Hispanic/Latino, 6.90% as Asian/Asian-American, 3.4 as Native American, and 1.10% as Native Hawaiian/ Pacific Islander.

**Materials**

The demographics questionnaire included items to determine participants’ age, relationship status, ethnicity, and grade level. The personality inventory (Big Five – IPIP) is a 100-item assessment contains 20 questions evaluating each of the big five dimensions of personality including: Openness, Conscientiousness, Extraversion, Agreeableness, and Emotional Stability. Overall Cronbach’s alpha: .90, overall Mean Item Intercorrelation: .31 (Goldberg, et al., 2006).

The Acceptance and Action Questionnaire-2 (AAQ-2) consists of seven questions and measures a person’s experiential avoidance and immobility and acceptance and action on a 7-point Likert scale. Higher scores indicate greater levels of psychological inflexibility. The mean alpha coefficient is 0.84. (Bond, et al., in press).

The Depression Anxiety and Stress Scale (DASS-21) is a 21-item self-report questionnaire designed to measure the severity of symptoms commonly seen in depression and anxiety. Participants rate the extent to which they have experienced each symptom over the past week, on a 4-point severity/frequency scale. Summing the scores for the relevant 7 items and doubling that score determines the scores for the DASS-21 scales. Internal consistencies (coefficient alpha) for each scale for the DASS normative sample from the original 42-item scale were: Depression 0.91; Anxiety 0.84; Stress 0.90. (Lovibond & Lovibond, 1995)
These questionnaires were included for future analyses separate from this thesis project.

See Appendix B for full questionnaires.
RESULTS

This study was completed in conjunction with another thesis project, resulting in 3 groups of data. Participants were randomly assigned to a control condition, a biofeedback intervention, and a yoga intervention. For the purposes of this thesis project, only differences between participants in the control condition and the biofeedback intervention were analyzed. Results from the questionnaires will be analyzed at a later date as part of a larger project.

Outliers

In total, 97 individuals participated. Ten participants were removed as outliers resulting in 87 participants: 29 in the control condition, 30 in the biofeedback condition, and 28 in the yoga condition. Cortisol assays were examined for each participant, and if a score was over 2 standard deviations above the mean the participant's scores were removed from further analysis. The 2 standard deviation cut off point was used because it identifies more possible outliers in the data than a 3 standard deviation cut off. In this study we did not determine cortisol-waking time, which can sometimes impact the results. A study by Herbert and colleagues found that only 1% of scores are 3 standard deviations above the mean (2012).

Analyses

After removal of the 10 outliers, cortisol scores were positively skewed. This was corrected by taking the natural log of all cortisol scores to normalize the distributions (Gordis et al., 2006). Change scores were then created to account for individual differences by subtracting the converted cortisol scores of time 1 from time 4, time 2 from time 4, time 3 from time 4, time
1 from time 3, time 2 from time 3, and time 1 from time 2. This resulted in a total of 6 change scores.

**Hypothesis 1.** A one-way between-subjects ANOVA was conducted with a Bonferroni correction to compare the effects of the three conditions on salivary cortisol change scores. It was determined that condition did not have a significant effect on time 2 to 1, indicating that participants in all conditions did not differ in their stress levels, and that all participants had an increase in cortisol levels ($F(2,84) = 2.456, p = 0.092$).

**Hypothesis 2.** Condition did have a significant effect on time 4 to 3 ($F(2,84) = 3.106, p = 0.050$), time 4 to 2 ($F(2, 84) = 7.29, p = 0.001$), and time 3 to 2 ($F(2,84) = 5.423, p = 0.006$) at the $p < 0.05$ level (See Table 1 for ANOVA results).

A series of independent sample t-test were then conducted for post hoc analyses of the significant ANOVA scores. For the current thesis study, t-tests were only conducted between the control condition and the biofeedback condition. Analyses between the two intervention groups will be examined at a later date.

Results revealed a significant difference between the control and biofeedback conditions from time 4 to 2 ($df(57) = 3.700, p = 0.001$) with scores indicating a greater reduction in cortisol levels for participants in the biofeedback condition ($M = -0.515, SD = 0.467$) compared to the control condition ($M = -0.120, SD = 0.345$). There was also a significant difference from time 3 to 2 ($df(57) = 3.375, p = 0.001$), with scores indicating a greater reduction in cortisol levels for participants in the biofeedback condition ($M = -0.389, SD = 0.364$) compared to the control condition ($M = -0.078, SD = -0.344$). This reduction in salivary cortisol levels indicates a greater reduction in stress levels in participants in the biofeedback condition after the
intervention and upon completion of the study. See Table 1 for a full list of ANOVA results, and Table 2 for a full list of means and standard deviations.

Table 1

ANOVA Results Comparing Salivary Cortisol Change Scores of an ALIVE Biofeedback Intervention, a Yoga Intervention, and a Control Condition.

<table>
<thead>
<tr>
<th>Change Score</th>
<th>Between Groups</th>
<th>Within Groups</th>
<th>Total</th>
</tr>
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<tbody>
<tr>
<td>Time 4 to 3</td>
<td>0.377</td>
<td>5.104</td>
<td>5.481</td>
</tr>
<tr>
<td>Time 4 to 2</td>
<td>2.769</td>
<td>15.956</td>
<td>18.726</td>
</tr>
<tr>
<td>Time 4 to 1</td>
<td>0.367</td>
<td>23.378</td>
<td>23.744</td>
</tr>
<tr>
<td>Time 3 to 1</td>
<td>0.016</td>
<td>28.154</td>
<td>28.170</td>
</tr>
<tr>
<td>Time 3 to 2</td>
<td>1.455</td>
<td>11.271</td>
<td>12.726</td>
</tr>
<tr>
<td>Time 2 to 1</td>
<td>1.740</td>
<td>29.750</td>
<td>31.490</td>
</tr>
</tbody>
</table>

*Indicates statistical significance at the p < 0.05 level
<table>
<thead>
<tr>
<th>Change Score</th>
<th>Group</th>
<th>n</th>
<th>M</th>
<th>SD</th>
<th>SE mean</th>
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</thead>
<tbody>
<tr>
<td>Time 4 to 3</td>
<td>Control</td>
<td>29</td>
<td>-0.043</td>
<td>0.199</td>
<td>0.037</td>
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<tr>
<td></td>
<td>Biofeedback</td>
<td>30</td>
<td>-0.126*</td>
<td>0.250</td>
<td>0.046</td>
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<tr>
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<td>0.345</td>
<td>0.064</td>
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<td>30</td>
<td>-0.515*</td>
<td>0.467</td>
<td>0.085</td>
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<tr>
<td>Time 3 to 2</td>
<td>Control</td>
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<td>-0.078</td>
<td>0.344</td>
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<tr>
<td></td>
<td>Biofeedback</td>
<td>30</td>
<td>-0.389*</td>
<td>0.364</td>
<td>0.067</td>
</tr>
</tbody>
</table>

* indicates a greater decrease in cortisol
DISCUSSION

Conclusion

The purpose of this study was to examine stress response recovery as measured by participants’ cortisol levels in relation to biofeedback gaming software. We hypothesized that participants in the biofeedback condition would have a greater reduction in cortisol after a stressor than those in the control condition after the intervention and upon completion of the study (saliva collection times 3 and 4 respectively). This hypothesis was supported. The participants’ cortisol peaked in both conditions at saliva collection time 2, ten minutes after the completion of the Trier Social Stress Test, and there was no significant difference between the two conditions similar to previous literature using the TSST (Carroll & Winslow, 2017; Gordis et al., 2006). Additionally, those in the biofeedback condition had a significantly greater cortisol reduction from time 2 to time 3, as well as from time 2 to time 4, demonstrating an overall reduction in cortisol production after a stressor. This is consistent with the reduction of cortisol from pre-training to post-training found in Carroll and Winslow’s 2017 study using biofeedback.

Based on the pattern of cortisol levels observed during the study, it appears ALIVE computer-based biofeedback software was a more effective short-term relaxation tool than the control condition (participants sitting while watching an emotionally neutral nature film). During the biofeedback intervention, cortisol levels decreased significantly more than when no intervention was used, though both went through the same research situation characterized by social evaluative threat: the Trier Social Stress Test.

College students are a high-risk population in need of recommendations on handling anxiety and stress, and the results of this study provide further support for biofeedback as an
effective relaxation tool. These results are consistent with what has been found in previous literature on college students and biofeedback used as a short-term intervention (Meier, 2013; Anstead, 2009). This is an important finding because several studies have shown that prolonged cortisol activation or chronic dysregulation of cortisol has detrimental long-term effects, both on physical and mental health (Dickerson & Kemeny, 2004; Raison & Miller, 2003). Learning self-regulation skills that provide immediate feedback, even used briefly, can help reduce levels of cortisol and combat the physiological effects of stress.

Many studies involving biofeedback discussed in this paper involve participants engaging in multiple training sessions over a span of time. In this study, participants were exposed to a short introduction to the use of biofeedback and 30 minutes of biofeedback training. Despite this having been a relatively brief intervention, participants showed a significantly greater reduction in cortisol levels compared to the control condition, indicating that even brief biofeedback can be effective in short-term stress reduction. More research is needed to determine the optimal amount of time for biofeedback training.

**Limitations**

One limitation of this study is that we did not collect pre and post self-report data on participant stress and anxiety levels to determine if participants’ perceived levels of stress reflect the observed cortisol reduction measured in this study. This would be helpful information in assessing how participants perceived the intervention, whether or not they found it useful as a relaxation technique. Another limitation is that the vast majority of researchers in this study were female, so it is unclear if there was a gender difference in how participants reacted to male or female administrators and judges.
Future Directions

It would be beneficial to compare the biofeedback intervention to the yoga intervention to understand the difference in efficacy of two different breathing-based interventions. Research has shown that both of these interventions are effective at reducing cortisol and stress levels, but yoga has the addition of physical movement and postures. It would also be useful to have a condition where only deep breathing is taught to compare the efficacy of teaching deep breathing techniques to monitoring and controlling the breath.

This study allowed participants to practice the first two stages of biofeedback as outlined by Carroll and Winslow: acquiring awareness of the maladaptive physiological responses and learning to control those responses using techniques such as deep breathing and passive muscle relaxation (2017). A future direction for this research would be to allow participants the opportunity to transfer that control to everyday life through multiple sessions of biofeedback training (Carroll & Winslow, 2017). This would allow participants to use the relaxation training to manage everyday stressors and avoid the chronic activation of the stress response that research has shown leads to physical and psychological complications.
REFERENCES


Students in Public Health Nursing. Nursing Research & Practice, 20151-5. doi:10.1155/2015/160746


APPENDICES

Appendix A. Informed Consent

TITLE: Learning to Relax: Changing the Stress Response
INVESTIGATORS: Dallas Robinson, B.S., Missouri State University
Tabetha Hopke, B.S., Missouri State University
Amber Abernathy, Ph.D., Missouri State University

PURPOSE: The purpose of the study is to better understand the stress response and how to reduce it through different relaxation techniques.

VOLUNTARY: This study is completely voluntary. You may refuse to answer any questions or choose to withdraw from participation at any time without any penalty or loss of benefits to which you are otherwise entitled.

WHAT DO YOU DO? Participants will be taking part in a mock job interview and a math-based activity. They will then be randomly selected to learn a computer-based relaxation technique, a yoga-based relaxation technique, or be allowed to relax on their own. Saliva will be collected four times to assess stress levels. Participants will then complete a series of questionnaires through Qualtrics, an online computer program.

RISKS: This project contains minimal risks. There may be some discomfort during the mock interview and math activity.

BENEFITS: Participants will learn and practice different ways to relax, which may lead to a reduction in overall stress levels and an increase in ability to handle stress. Participants will receive compensation in the form of research credit or extra credit.

CONFIDENTIALITY: Your answers are entirely confidential, and will not be revealed to anyone other than the researchers conducting the study. Only your arbitrary participant identification number will link you to data you provide. Your confidentiality will be maintained in that your name will not appear on the survey or in the published study itself. Any written results will discuss group findings and will not include information that will identify you. Research records will be stored securely and only researchers and individuals responsible for research oversight will have access to the records. Data will be stored on a secure computer with file encryption software for three years after the study completion.

CONTACTS: If you have any questions regarding the study, please contact Dallas Robinson at Robinson772@live.missouristate.edu

You have read and fully understand the consent form. You attest that you sign it freely and voluntarily.
Appendix B. Questionnaires

Acceptance and Action Questionnaire II (AAQ-II): Below you will find a list of statements. Please rate how true each statement is for you by using the scale below to fill in your choice.

<table>
<thead>
<tr>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
</tr>
</thead>
<tbody>
<tr>
<td>Never True</td>
<td>Very Seldom True</td>
<td>Seldom True</td>
<td>Sometimes True</td>
<td>Frequently True</td>
<td>Almost Always True</td>
<td>Always True</td>
</tr>
</tbody>
</table>

1. My painful experiences and memories make it difficult for me to live a life that I would value.
2. I’m afraid of my feelings.
3. I worry about not being able to control my worries and feelings.
4. My painful memories prevent me from having a fulfilling life.
5. Emotions cause problems in my life.
6. It seems like most people are handling their lives better than I am.
7. Worries get in the way of my success.

Depression, Anxiety, and Stress Scale (DASS 21): Please read each statement and circle a number 0, 1, 2 or 3, which indicates how much the statement applied to you over the past week. There are no right or wrong answers. Do not spend too much time on any statement.

<table>
<thead>
<tr>
<th>0</th>
<th>1</th>
<th>2</th>
<th>3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Never</td>
<td>Sometimes</td>
<td>Often</td>
<td>Almost Always</td>
</tr>
</tbody>
</table>

1. found it hard to wind down
2. I was aware of dryness of my mouth
3. I couldn’t seem to experience any positive feeling at all
4. I experienced breathing difficulty (eg., excessively rapid breathing, breathlessness in the absence of physical exertion)
5. I found it difficult to work up the initiative to do things
6. I tended to over-react to situations
7. I experienced trembling (eg., in the hands)
8. I felt that I was using a lot of nervous energy
9. I was worried about situations in which I might panic and make a fool of myself
10. I felt that I had nothing to look forward to
I found myself getting agitated.
I found it difficult to relax.
I felt down-hearted and blue.
I was intolerant of anything that kept me from getting on with what I was doing.
I felt I was close to panic.
I was unable to become enthusiastic about anything.
I felt that I wasn’t worth much as a person.
I felt that I was rather touchy.
I was aware of the action of my heart in the absence of physical exertion (eg., sense of heart rate increase, heart missing a beat).
I felt scared without any good reason.
I felt that life was meaningless.

The International Personality Item Pool (IPIP) 50 item version: Please rate your agreement with the following items.

<table>
<thead>
<tr>
<th>Neuroticism</th>
<th>Very Inaccurate</th>
<th>Moderately Inaccurate</th>
<th>Neither Accurate nor Inaccurate</th>
<th>Moderately Accurate</th>
<th>Very Accurate</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Often feel blue.</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>2. Dislike myself.</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td></td>
</tr>
<tr>
<td>3. Am often down in the dumps.</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>4. Have frequent mood swings.</td>
<td>4</td>
<td>5</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5. Panic easily.</td>
<td>5</td>
<td>6</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6. Seldom feel blue.</td>
<td>6</td>
<td>7</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>7. Feel comfortable with myself.</td>
<td>7</td>
<td>8</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>8. Rarely get irritated.</td>
<td>8</td>
<td>9</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>9. Am not easily bothered by things.</td>
<td>9</td>
<td>10</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10. Am very pleased with myself.</td>
<td>10</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Extraversion
1. Feel comfortable around people.
2. Make friends easily.
3. Am skilled in handling social situations.
4. Am the life of the party.
5. Know how to captivate people.
6. Have little to say.
7. Keep in the background.
8. Would describe my experiences as somewhat dull.
9. Don’t like to draw attention to myself.
10. Don’t talk a lot.

Openness
1. Believe in the importance of art.
2. Have a vivid imagination.
3. Tend to vote for liberal politician candidates.
4. Carry the conversation to a higher level.
5. Enjoy hearing new ideas.
6. Am not interested in abstract ideas.
7. Do not like art.
8. Avoid philosophical discussions.
9. Do not enjoy going to at museums.
10. Tend to vote for conservative political candidates.

Agreeableness
1. Have a good word for everyone.
2. Believe that others have good intentions.
3. Respect others.
4. Accept people as they are.
5. Make people feel at ease.
6. Have a sharp tongue.
7. Cut others to pieces.
8. Suspect hidden motives in others.
9. Get back at others.
10. Insult people.

Conscientiousness
1. Am always prepared.
2. Pay attention to details.
3. Get chores done right away.
4. Carry out my plans.
5. Make plans and stick to them.
7. Find it difficult to get down to work
8. Do just enough work to get by.
9. Don’t see things through.
10. Shirk my duties.

Demographic Questionnaire: Please select the answers below that best represent you currently.

1.) Your age: ___
2.) Your gender: ___Male   ___Female
3.) Relationship Status:
Married               Cohabitating                 Divorced                  Widowed
Committed Relationship          Dating               Single

4.) Your ethnicity:
   ___Caucasian ___American Indian ___African-American/Black ___Biracial
   ___Hispanic/Latino ___Asian/Asian-American ___Other

5.) Your current grade level (select one):
   ___Freshman ___Sophomore             ___Junior             ___Senior
   ___Other             ___Graduate student ___Not applicable

6.) Please estimate your income:
   ___$0 - $10,000 ___$10,000 - $20,000 ___$20,000 - $30,000
   ___$30,000 - $40,000 ___$40,000 - $50,000 ___$50,000 - $60,000
   ___$60,000 - $70,000 ___$70,000 - $80,000 ___$80,000 – 90,000
   ___$90,000 - $100,000 ___$100,000-$110,000 ___$ Over $110,000

7.) Do you smoke or use nicotine?     ____Yes       ____No

8.) How long ago did smoke or use nicotine?
   ___0 – 30 min ___30 – 60 min ___1 – 2 hours ___2 – 3 hours
   ___3 – 4 hours ___4 – 5 hours ___5 – 6 hours ___6 – 7 hours ___7+ hours

9.) How long ago did you have caffeine?
   ___0 – 30 min ___30 – 60 min ___1 – 2 hours ___2 – 3 hours
   ___3 – 4 hours ___4 – 5 hours ___5 – 6 hours ___6 – 7 hours ___7+ hours

10.) How long ago did you eat?
    ___0 – 30 min ___30 – 60 min ___1 – 2 hours ___2 – 3 hours
    ___3 – 4 hours ___4 – 5 hours ___5 – 6 hours ___6 – 7 hours ___7+ hours

11.) How many days/week do you typically exercise?
     ___1 ___2 ___3 ___4 ___5 ___6 ___7

12.) Do you practice any form of meditation or mindfulness? ___Yes ___No
Appendix C. Human Subjects IRB Approval

IRB #: IRB-FY2019-380

Title: Learning to Relax: Stress Reduction Strategies
Creation Date: 11-30-2018
End Date: 1-17-2020
Status: Approved
Principal Investigator: Amber Abernathy
Review Board: MSU
Sponsor:

Study History
Submission Type Initial Review Type Expedited Decision Approved

Key Study Contacts
Member Amber Abernathy Role Principal Investigator
Contact amberabernathy@missouristate.edu
Member Dallas Robinson Role Primary Contact
Contact Robinson772@live.missouristate.edu
Member Dallas Robinson Role Co-Principal Investigator
Contact Robinson772@live.missouristate.edu
Member Tabetha Hopke Role Co-Principal Investigator
Contact tabetha6789@live.missouristate.edu

Initial Submission

1A. What is the full title of the research protocol?
Learning to relax: Examining the impact of stress reduction techniques on salivary cortisol

1B. Abstract/Summary
Please provide a brief description of the project (no more than a few sentences).
College students have a high prevalence of mental health problems and that number seems to be increasing (Lipson et al., 2015; Reetz et al., 2017). According to the Healthy Minds Study, 34.4% of college students surveyed (n = 42,210) had at least one mental health problem like anxiety, depression, or serious thoughts of suicide (Lipson et al., 2015). There is evidence that relaxation techniques can be useful in overall stress management (Van Rhenen et al., 2005), and short-term interventions like computer-based biofeedback or yoga could be ways to give students a time friendly and independent way to manage the physiological symptoms of stress and anxiety. The purpose of this study is to compare the impact of a biofeedback intervention, a yoga intervention, and a control on the stress response as measured by salivary
cortisol levels and self-report variables like anxiety and distress, as well as examining factors that may influence participant response to the intervention.

1C. Who is the Principal Investigator?
This MUST be a faculty or staff member.
Name: Amber Abernathy
Organization: Psychology
Address: 901 S National Ave, Springfield, MO 65897-0027
Phone: 
Email: amberabernathy@missouristate.edu

1D. Who is the primary study contact?
This person may be the Principal Investigator or someone else (faculty, staff, or student). This person, in addition to the PI, will be included on all correspondence related to this project.
Name: Dallas Robinson
Organization: Psychology
Address: 901, S. National Avenue, Springfield, MO 65897-0027
Phone: 
Email: Robinson772@live.missouristate.edu

1E. Select the Co-Principal Investigator(s).
This MUST be a faculty or staff member. Persons listed as Co-PIs will be required to certify the protocol (in addition to the PI). This person will also be included on all correspondence related to this project.
Name: Dallas Robinson
Organization: Psychology
Address: 901, S. National Avenue, Springfield, MO 65897-0027
Phone: 
Email: Robinson772@live.missouristate.edu
Name: Tabetha Hopke
Organization: Psychology
Address: 901, S. National Avenue, Springfield, MO 65897-0027
Phone: 
Email: tabetha6789@live.missouristate.edu

1F. Select the Investigator(s).
An investigator may be faculty, staff, student, or unaffiliated individuals.
If you could not locate personnel using the "Find People" button, please request access at Cayuse Logon Request
For additional help, email irb@missouristate.edu.

Describe the proposed project in a manner that allows the IRB to gain a sense of the project including:
the research questions and objectives,
key background literature (supportive and contradictory) with references, and the manner in which the proposed project will improve the understanding of the chosen topic.

Cortisol and the Stress Response

Self-regulation is the integrative functioning of emotional and physiological regulation to maintain or reestablish homeostasis, where the physiological systems in the body are all at their optimal levels (Sapolsky, 2004). This can be measured through the functioning of the autonomic nervous system (ANS) (Crockett et al., 2011). The ANS is responsible for regulating bodily systems like respiration, heart rate, etc. Maintaining a physiological balance is essential to maintaining good health, and homeostasis can be disrupted by prolonged activation of the stress response, which can be defined as a network of physiological responses in reaction to a threat or stressor, whether real or perceived (Sapolsky, 2004; Crockett et al., 2011). This prolonged activation can lead to chronic stress and other associated problems.

Chronic stress has been linked to several health related problems in areas of fine motor performance, attention, and cognitive function (Carroll & Winslow, 2017). When energy is constantly mobilized in the stress response, fatigue occurs because the body is unable to store energy (Sapolsky, 2004). This can be even more detrimental when the stress-response is activated due to an imagined threat or a psychological stressor. There are several components to psychological stressors that contribute to anxiety, such as a lack of predictability, lack of control, and an inappropriate interpretation of the stressor (Sapolsky, 2004). Anxiety can be accompanied by physiological symptoms like increased heart rate, sweating, and rapid breathing, as well as distress and an inability to function. (Tabachnick, 2015).

One of the ways the stress response can be measured is through the analysis of cortisol levels from participant saliva samples. Psychological stressors cause the activation of the hypothalamus-pituitary-adrenal (HPA) axis, increasing levels of several hormones including cortisol. Cortisol is a glucocorticoid hormone released when stimuli interpreted as a potential physical or psychological threat stimulates the HPA axis (Byrd-Craven et al., 2012). Chronic dysregulation or excessive secretion of cortisol has been shown to have detrimental long-term effects on both physical and mental health (Raison & Miller, 2003).

The Trier Social Stress Test (TSST) has been shown to stimulate the stress response in a research setting. This test has been found to increase levels of salivary cortisol to 2 to 4 elevations above baseline levels (Kirschbaum et al., 1993). The TSST typically consists of a ten-minute anticipation period and a ten-minute test period where the participant delivers a free speech and performs mental arithmetic in front of an audience. When this test is administered, salivary cortisol has been shown to peak ten minutes after administration (Kirschbaum et al., 1993; Gordis et al., 2012).
Biofeedback
Biofeedback is a method of helping individuals regulate ANS functioning and learn how to become aware of physiological processes like muscle tension, breathing, heart-rate variability, etc., and then work to change them (Ratanasiripong, 2009; Ratanasiripong, 2015). Peripheral biofeedback provides feedback based on activity in the body, while neurofeedback provides feedback based on activity in the brain (Crockett et al., 2011). Peripheral biofeedback can be used to target the physiological symptoms associated with anxiety through a feedback loop in which information about the participant’s experience can be used to inform the client’s self-regulation (Crockett et al., 2011). This could eventually teach individuals to control and reduce those symptoms.

Biofeedback typically has three stages: 1) acquiring awareness of the maladaptive physiological responses 2) learning to control those responses using techniques like deep breathing and passive muscle relaxation 3) transferring that control to every day life (Carroll & Winslow, 2017).

This technique has been shown to be effective in reducing stress associated with every day hassles and anxiety (Carroll & Winslow, 2017). In Carroll and Winslow’s 2017 study, a condensed 90-minute biofeedback training method was used incorporating diaphragmatic breathing and stress inoculation training. The experimental group was compared to a control group using physiological measures of the ANS response and cortisol measures of the stress response, as well as perceived stress levels using the state portion of the State Trait Anxiety Inventory. Those in the biofeedback condition had a statistically significant reduction in cortisol, but not in ANS stress response or perceived stress from pre-training to post-training. These results support the efficacy of a condensed biofeedback method, but also show the importance of additional individual practice time (Carroll & Winslow, 2017).

In another study, participants in the biofeedback condition using a diaphragmatic breathing technique experienced a significant reduction in cortisol from pre-training to post-training, but the control group did not (Carroll & Winslow, 2017). In a different study, a four-week biofeedback intervention showed a reduction in both stress and depression, and a statistically significant reduction in anxiety when compared to the control group (Ratanasiripong, 2015). Those in the control condition actually had a slight increase in reported depression and stress.

Heart rate variability (HRV) is one method of biofeedback, looking at the beat-to-beat changes in heart rate. Low HRV has been linked to psychopathologies like anxiety disorders and substance use disorders during craving states (Henriques et al., 2011; Thurstone & Lajoie, 2013). HRV
biofeedback has been shown to reduce stress and mean salivary cortisol levels, one of the more common ways of measuring the stress response (Thurstone & Lajoie, 2013). Research has suggested that computer-based biofeedback may be beneficial for stress management. The use of a computer-based biofeedback program significantly decreased levels of both state and trait anxiety in college students (Henriques et al., 2011). This biofeedback program used a finger pad to track the user’s heart rate and displayed a graph showing the user’s real-time heart rate, where a smooth line indicated relaxation. This intervention lasted four weeks and was individualized so the participants could practice biofeedback on their own. There is a need for more in-depth research on computer-based biofeedback and its potential benefits, as well as how effective a short-term intervention is.

One of the general guidelines for biofeedback is that to develop the skill it is necessary for participants to take an active role in practicing, but there is still debate on the amount of time, method of biofeedback, and sessions necessary for efficacy (Carrol & Winslow, 2017).

Yoga

In the broadest sense of the term, yoga could be considered a practice that involves postures (poses) of the physical body (asanas) and controlled breathing (pranayama). Gentle yoga, which will be the focus of this study, is a low-intensity yoga practice. While this type of class still uses active poses, these poses are not meant to be physically demanding or increase heart rate. Gentle yoga classes are fairly slow-paced, maybe taking several breaths per pose instead of the “one breath one movement” style of a power vinyasa practice, and are meant to be accessible to students with a wide range of physical activity levels.

Many times, there is a mindfulness meditation component to yoga practice. According to Kabat-Zinn (1982), meditation is the practice of directing the thoughts, while mindfulness meditation is the practice of disconnecting from thoughts and simply noticing them without judgment. This mindfulness meditation component is what fosters the mind-body connection that is traditionally associated with yoga, and on its own has been shown to be beneficial in stress reduction (Carlson, Speca, Patel, & Goodeyhe, 2003). Although mindfulness meditation component of yoga classes can also vary greatly, classes frequently involve a focus on breath and connecting breath with postures in specific ways which can assist students in disconnecting from their thoughts.

Implementation of a yoga practice has previously been shown to decrease psychological stress measures as well as biological stress markers (Riley & Park, 2015; Ross & Thomas, 2010). It has also been shown that one session of Hatha yoga immediately decreased salivary cortisol, perceived stress, and negative affect (West et al., 2004). The duration and type of yoga practice used in studies involving yoga however varies greatly. This study aims to add to the existing base of research by examining whether one session of gentle yoga could assist in recovery of the stress
response system as assessed through measurement of salivary cortisol.

2B. Check all research activities that apply:

- ✔ Audio, video, digital, or image recordings
- Biohazards (e.g., rDNA, infectious agents, select agents, toxins)
- ✔ Biological sampling (other than blood)
- Blood drawing
- Class Protocol (or Program or Umbrella Protocol)
- Data, not publicly available
- Data, publicly available
- Deception
- ✔ Devices
- ✔ Diet, exercise, or sleep modifications
- Drugs or biologics
- Focus groups
- ✔ Internet or email data collection
- Materials that may be considered sensitive, offensive, threatening, or degrading
- Non-invasive medical procedures
- ✔ Observation of participants
- Oral history
- Placebo
- Record review
- Specimen research
- Surgical procedures
- ✔ Surveys, questionnaires, or interviews (one-on-one)
- Surveys, questionnaires, or interviews (group)
- Other

Describe the procedures and methods planned for carrying out the study. Make sure to include the following:

- site selection,
- the procedures used to gain permission to carry out research at the selected site(s),
- aaqii-7-item-scoring-original-working-3-reader.pdf
- C - DASS 21.pdf
- IPIP formatted right.docx
- Demographic Questionnaire - B.docx

2C.

- data collection procedures,
- and an overview of the manner in which data will be analyzed.

Provide all information necessary for the IRB to be clear about all of the contact human participants will have with the project.

Participants will be recruited from the community through psychology courses in a large mid-western university. Students will receive course credit or extra credit for their participation. The study will be conducted in a research lab in the psychology building on campus. Participants will be randomly
assigned to a control condition or a condition teaching ALIVE biofeedback software. The biofeedback software will measure heart rate and breathing smoothness through a finger pad. Participants will be given an informed consent document explaining the study upon entry. Saliva will be collected immediately before beginning the Trier Social Stress Test (TSST) to establish a baseline stress response measured by cortisol levels. There will be two researchers in the room, one acting as the judge and the other collecting saliva and giving instructions. The TSST consists of a ten minute anticipatory period where participants will prepare a speech as if on a job interview followed by five minutes of the interview and five minutes of a mathematical activity. Saliva will be collected again ten minutes after the completion of the stress test. Participants will then be assigned to receive the ALIVE biofeedback intervention, the yoga intervention, or the control condition for thirty minutes. Saliva will be collected ten minute after completion of the intervention. Then, participants will be given several surveys through Qualtrics; the order in which they will be presented will be counterbalanced. Saliva will be collected for a final time 30 minutes after beginning the surveys.

2D. Attach surveys, questionnaires, and other social-behavioral measurement tools, if applicable.

3A. Specify the participant population(s). Check all that apply.

✔ Adults
Children (<18 years)
Adults with decisional impairment
Non-English speaking
✔ Student research pools (e.g. psychology)

Specify:
students enrolled in select courses in the psychology department
Pregnant women or fetuses
Prisoners
Unknown (e.g., secondary use of data/specimens, non-targeted surveys, program/class/umbrella protocols)

3B. Specify the age(s) of the individuals who may participate in the research.
Participants will be age 18 and older.

3C. Describe the characteristics of the proposed participants, and explain how the nature of the research requires/justifies their inclusion.
Participants will be college students from psychology classes. They will receive course credit for their participation, and potentially a useful intervention meant to decrease stress levels.

3D.
Provide the total number of participants (or number of participant records, specimens, etc.) for whom you are seeking Missouri State IRB approval.

100 participants.

3F. 
Estimate the time required from each participant, including individual interactions, total time commitment, and long-term follow-up, if any.
Total: Approximately 1.5-2 hours of participation.

3G. 
Describe how potential participants will be identified (e.g., advertising, individuals known to investigator, record review, etc.). Explain how investigator(s) will gain access to this population, as applicable.
Participants will be identified based on their enrollment in psychology courses.

3H. 
Describe the recruitment process; including the setting in which recruitment will take place. Provide copies of proposed recruitment materials (e.g., ads, flyers, website postings, recruitment letters, and oral/written scripts).
Participants will be able to sign up for this study online and select from a variety of different research projects and timeslots available.
3H.1. Attach recruitment materials, if applicable.

3I. 
Will participants receive compensation or other incentives (e.g., free services, cash payments, gift certificates, parking, classroom credit, travel reimbursement, etc.) to participate in the research study?

✔ Yes

Describe the incentive, including the amount and timing of all payments.
Participants in an introductory psychology course will receive 4 research credits (1 per half hour of participation). Participants in other psychology classes will be offered extra credit.

4A. 
From the list below, indicate how consent will be obtained for this study. Check all that apply.
✔ Written/signed consent by the subject
Written/signed consent (permission) for a minor by a Parent or Legal Guardian
Written/signed consent by a Legally Authorized Representative (for adults incapable of consenting).
Request for Waiver of Documentation of Consent (e.g. Verbal Consent, Anonymous Surveys, etc.)
Waiver of parental permission
Consent will not be obtained from subjects (Waiver of Consent)

4B. 
Describe the consent process including where and by whom the subjects will be approached, the plans to ensure the privacy of the subjects and the measures to ensure that subjects understand the nature of the study, its procedures, risks and benefits and
that they freely grant their consent. Participants will be presented with a hard copy of the informed consent document and given time to read it. All participant information (survey responses, saliva samples) will be identified with a number to ensure privacy. The method of the study will be described, as well as any potential risks and benefits. Finally, they will sign the document if they wish to continue their participation, and if not, they will be excused from the study.

4B.1. Attach all copies of informed consent documents (written or verbal) that will be used for this study.
Sample documents: Informed Consent Examples

4B.2. Attach all copies of assent documents that will be used for this study, if applicable.
Sample documents: Assent Examples

5A. Describe all reasonably expected risks, harms, and/or discomforts that may apply to the research. Discuss severity and likelihood of occurrence.
Consider the range of risks - physical, psychological, social, legal, and economic.
To elicit the stress response, participants will take the Trier Social Stress Test. This may cause mild discomfort or distress in some participants, but the components are not unlike what students are required to do in class: public speaking and performing mental arithmetic. Participants in the yoga group will be performing light physical activity but the activity is not meant to be challenging or increase heart rate.

5B. Describe the steps that will be taken to minimize risks and the likelihood of harm.
Participants will be informed prior to arriving for the study that they may be asked to take part in a yoga practice or use biofeedback software. They will be given instructions during the test, and the researchers will explain the nature of the TSST to participants upon completion of the study.

5C. List the potential benefits that participants may expect as a result of this research study.
State if there are no direct benefits to individual participants.
The biofeedback and yoga interventions have been shown to reduce stress levels and feelings of anxiety.

5D. Describe any potential indirect benefits to future subjects, science, and society.
This research will further the understanding of short-term biofeedback and yoga interventions and the impact they have on stress response recovery and self-report variables.

5E.
Discuss how risks to participants are reasonable when compared to the anticipated benefits to participants (if any) and the importance of the knowledge that may reasonably be expected to result. The TSST is a small stressor that most will have experienced before. If participants can better regulate their stress response when presented with this stressor, it is possible that ability to cope will translate into their every day lives making the use of the test reasonable.

Missouri State University is committed to keeping data and information secure. Please review the Missouri State Information Security policies. Discuss your project with the MSU Information Security Office or your College's IT support staff if you have questions about how to handle your data appropriately.

6A. Statement of Principal Investigator Responsibility for Data
The principal investigator of this study is responsible for the storage, oversight, and disposal of all data associated with this study. Data will not be disseminated without the explicit approval of the principal investigator, and identifying information associated with the data will not be shared.

✔ By checking this box, all personnel associated with this study understand and agree to the Statement of Principal Investigator Responsibility for Data.

6B. How will the data for this study be collect/stored?
Check all that apply.

✔ Electronic storage format

On paper

Describe where the data will be stored (e.g., paper forms, flash drives or removable media, desktop or laptop computer, server, research storage area network, external source) and describe the plan to ensure the security and confidentiality of the records

6C.
(e.g., locked office, locked file cabinet, password-protected computer or files, encrypted data files, database limited to coded data, master list stored in separate location).
At minimum, physical data should always be secured by lock and key when stored. Electronic data should be stored on University secure servers whenever possible (Office 365 or other secure campus server). If data has to be stored off campus, the file should be encrypted and the device password protected. Additionally, any data to be shared outside the University network will require a SUDERS request be filed and approved.
See https://mis.missouristate.edu/Central/suders/creat...

Saliva samples will be stored in a refrigerator located in a private research lab on the fourth floor of the psychology building. Only the primary investigator and members of the research team will have access to this room. For questionnaires, the online survey system Qualtrics will be used. Participants will enter a participant number, the same number that will be linked to the saliva samples so that the participants name will only be found on the informed consent document. Informed consent forms
will be stored in a locked cabinet in the research lab.

6D. Describe how data will be disposed of and when disposal will occur. At minimum, Federal regulations require research records to be retained for at least 3 years after the completion of the research (45 CFR 46). Research that involves identifiable health information is subject to HIPAA regulations, which require records to be retained for at least 6 years after a participant has signed an authorization. Finally, funded research projects may require longer retention periods, you may need to follow the sponsoring agency guidelines. Informed consent documents will be kept in a locked cabinet in the research lab for three years after the completion of the study and then will be shredded.

7A. Is this study externally funded? For example, this research is funded by a source outside Missouri State; a federal agency, non-profit organization, etc.
Yes ✅ No
Potentially (this study is being submitted for funding, but has not yet been awarded)

7B. Is this study internally funded? For example, this research is funded by a source inside Missouri State; departmental funds, the Graduate College, etc.
✅ Yes
Please list the internal funding source.
Missouri State University Graduate College
No
Potentially (this study is being submitted for funding, but has not yet been awarded)

8A. Does your study contain protected health information (PHI)? PHI is any information in a medical record or designated record set that can be used to identify an individual and that was created, used, or disclosed in the course of providing a health care service, such as a diagnosis or treatment.
Yes ✅ No
A -Consent .docx
Dallas CITI.pdf
Dallas CITI 3.pdf
Dallas CITI 2.pdf
CITI training.pdf
1abernathyciti.pdf
2abernathyciti.pdf
3abernathyciti.pdf
9A. Human Subjects Training Certificates
Attach human subjects training certificates for all listed personnel. To access your training documents, please go to CITI Training.

9B. HIPAA Training Certificates
Attach HIPAA training certificates for all listed personnel, if applicable. To get more information about HIPAA training and/or to access your training documents, please go to HIPAA Information for Researchers.

9C. Informed Consent Documents
Attach all copies of informed consent documents (written or verbal) that will be used for this study.
Sample documents: Informed Consent Examples
Assent Documents
aaqii-7-item-scoring-original-working-3-reader.pdf
C - DASS 21.pdf
IPIP formatted right.docx
Demographic Questionnaire - B.docx

9D. Attach all copies of assent documents (written or verbal) that will be used for this study.
Sample documents: Assent Examples

9E. Recruitment Tools
Attach copies of proposed recruitment tools.

9F. Surveys/Questionnaires/Other Social-Behavioral Measurement Tools
Attach surveys, questionnaires, and other social-behavioral measurement tools.

9G. Other Documents
Attach any other documents that have not been specified in previous questions, but are needed for IRB review.

10A. Would you like to add additional information?
Yes ☑ No