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Research Article

Effects of Bio-Tuning Intervention on Stress, Depression, and Anxiety During the COVID-19 Pandemic

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Abstract

Bio-tuning has been proposed as a tool for stress reduction. Its protocol uses sound that has been derived from the patient's nervous system using a heart rate variability monitor. Developers at the Center for Neuro acoustic Research (CNR) claim that in addition to being effective, Bio-tuning can help many people as it is non-invasive, and time and cost efficient. This study examined its effectiveness by evaluating data obtained by a certified CNR practitioner. Bio-tuning's treatment effect was evaluated on adult patients' anxiety, depression, sleep quality, perceived quality of life, and stress related to COVID-19. Thirty-four patients completed assessments at the beginning and end of the Bio-tuning treatment period. Patients also completed a sleep questionnaire at mid-treatment to track sleep quality changes. Data was collected between September 2021 and February 2022. At the end of the 21-day treatment period, results showed a significant decrease in anxiety ($p=0.02$, Cohen's $d=0.36$) and depression ($p<0.0001$, Cohen's $d=0.66$), and significant improvements in sleep quality ($p<0.0001$, Cohen's $d=0.89$) and quality of life in the physical ($p=0.002$, Cohen's $d=0.63$) and psychological health ($p=0.02$, Cohen's $d=0.32$) domains. Repeated measures analysis of sleep quality showed significant improvements between Day 1 and Day 11 ($p<0.001$, Cohen's $d=0.83$), suggesting an immediate improvement in sleep quality. These results suggest that Bio-tuning can effectively treat symptoms of depression and anxiety, improve quality of sleep, and improve perceptions of psychological and physical health.

Keywords: Bio-tuning, Stress reduction, Depression, Anxiety, Sound frequencies, Sleep quality, Mind-body, Binaural beats, Brainwave entrainment, COVID-19

Introduction

When our daily lives become challenging, we begin to experience a degree of displeasure that we identify as stress. Cherry and Mattiuzzi defined stress as "A condition in which extreme pressure, hardship, or pain is either suddenly experienced or built up over time" (p.255) [1]. Breedlove and Watson further suggest that stress involves upsetting a person's "homeostatic balance" (p.510) [2]. While these definitions may be vague and somewhat elusive, it is clear that stress involves a reaction to a disruption in a person's life. Thus, as a reaction, stress can be perceived as having an adaptive value. Breedlove and Watson state that in the immediate encounter of a threat, the body's reaction is to mobilize its resources for energy usage at the cost of energy storage [2]. This reaction provides a person with the appropriate resources to respond to a threat for the sake of survival. However, over time, this continued draw on the body's resources can be debilitating and result in health problems [1-3]. Stress is associated with a number of implications for mental health. Among the more prevalent is anxiety. The National Comorbidity Survey revealed that an estimated 31.1% of U.S. adults have experienced some anxiety disorder at some point in their life [4]. The fifth edition of the Diagnostic and Statistical Manual of Mental Disorders (DSM-5) states that among other symptoms, anxiety involves excessive worry and difficulty controlling the worry [5]. As a

result, a person may find it difficult to concentrate on tasks, may be easily fatigued, and may become restless or irritable. Such impairments in a person's functioning may ultimately result with problems in their personal life, such as the loss of a job, disruption of relationships, academic difficulties, etc. Thus, one can easily see how constant worry about the future not only causes distress, but may also exacerbate a person's existing problems, adding to their overall experience of stress. Therefore, the role of stress in a person's life should be considered when examining their level of anxiety.

In addition to anxiety, depression is often associated with high levels of stress. Among its many symptoms, the DSM-5 lists a sad or empty mood, or a lack of interest in all or almost all activities, as hallmarks of a depressive disorder [5]. The National Comorbidity Survey reports that an estimated 19.4% of Americans struggle with depression at some point in their life [4]. Studies show that depression may occur as a result of some stressful life event [6]. Further, studies have also shown that stressful life events were likely to precede the first onset of a person's depression [7-9]. Thus, it is evident that as a moderating variable, stress should also be considered for its role in depression. Stress can also impact sleep. The implications of this are far reaching, ranging from mental health to physical health problems. For instance, one study showed a significant positive correlation between the amount of perceived stress and sleep disturbances among female college students [10]. Additionally, *Stepan, et al.,* [11] showed that poor sleep is associated with problems in memory and attention [11]. Further, *Li, et al.,* [12] found that having a healthy sleep pattern was inversely associated with heart failure [12]. Thus, it is essential to consider the role stress plays when examining one's overall quality of sleep as it may have implications beyond fatigue. Finally, a look at a person's quality of life is necessary when examining the impact of stress. As mentioned, stress can exacerbate a person's experience of depression and anxiety and interfere with healthy sleep patterns. However, the experience of stress may also impair a person's perceived life quality. For instance, a review of studies highlights the negative association between stress and quality of life [13]. As quality of life has been defined as a "perception of [one's] position in life,", it is imperative to recognize how this perception may also impact a person's overall mental health [14]. Moreover, it is necessary to understand the role that stress plays in influencing this perception. In early 2020, people around the world were asked to retreat to their homes as a means of reducing the number of individuals becoming infected with a novel respiratory disease known as coronavirus (COVID-19). As of December 2020, the World Health Organization's (WHO) website reported more than 69 million confirmed cases of individuals affected by the disease and more than 1.5 million related deaths worldwide [15]. Concurrently, in the United States, the CDC reported a total of 15 million total cases and nearly 300,000 related deaths [16]. As reported by the CDC, a large majority of deaths (80%) appear to involve individuals aged 65 or older. This is followed by people aged 50-64 (15%), then people aged 0-49 (<5%) [17].

The impact on people's way of life as a result of the virus has been monumental. Though the rules vary between places (such as countries, states, and cities) and are often changed within that particular place, people are often required to social distance, wear a protective mask any time they are outside their home, limit their interactions with others, and manage their expenses in a time of severe economic uncertainty. This is in addition to the expected fear of contracting the virus, frustration over uncertainty about when a vaccine would be made available, and sadness over the forced isolation and loss that comes with the inability to interact with loved ones. Thus, it is expected that such stressful circumstances would negatively impact the mental health of people. Indeed, a review of studies conducted by *Lakhan, et al.,* [18] showed an increase in levels of depression, anxiety, and stress, as well as sleep problems and psychological distress in the general population during the months of December 2019 through June 2020 [18]. A number of methods may be considered when attempting to manage stress. People may choose to engage in stress-reducing behaviors: calling a friend for support, engaging in physical activity, or practicing relaxation techniques such as deep breathing, meditation, yoga, or tai chi. Simply put, people may engage in these behaviors just because they believe it helps them feel better. Indeed, their hunch about these practices may not be without merit, as meta-analyses show that alternative practices such as yoga and acupuncture have demonstrated a reduction in stress [19-24]. Similarly, mindfulness-based practices, such as meditation and body scanning, have also demonstrated a stress-reducing effect [25-27]. Indeed, such outcomes have not gone unnoticed by clinicians in the psychotherapeutic field. A study conducted by *Michalak, et al.,* [28] found that of the 64 therapists they surveyed, at least 82% had incorporated some type of mindfulness-based practice in their work with patients [28]. This reflects an openness among clinicians to incorporate alternative stress reduction methods into their work.

Mindfulness-based techniques have been embedded into therapy in more profound ways. Specifically, Mindfulness-Based Stress Reduction (MBSR) and its variant, Mindfulness-Based Cognitive Therapy (MBCT), have been shown to reduce stress, depression, and anxiety [29,30]. Developed by *Kabat-Zinn* [31]. MBSR is an 8-week group program targeting various psychological conditions, while mindfulness-based cognitive therapy (MBCT) is a program developed for targeting depression [32]. More importantly, these programs have been subject to empirical study and were shown to be effective in reducing stress [27,33-35]. Although effective, the usefulness of MBSR, MBCT, and related techniques are often limited only to those who have the time available to commit to such a program. For instance, the MBSR program is an 8-week program that requires the active daily home practice of mindfulness meditation [33]. In addition, the program may ask the participant to engage in extra tasks such as attending weekly meetings, day long retreats, and/or a mandatory orientation *(https://www.mindfulleader.org/mbsr-training)*. Given the responsibilities of the average person's life (i.e., work, school, family, etc.), such a commitment may cause

some to rethink their enrollment in such a program. As such, a less demanding method for reducing stress is needed. One such method is Bio-tuning, which was developed by the Center for Neuro acoustic Research (CNR) [36]. The Bio-tuning procedure requires the patient to meet once with a technician who is trained in the method. In this initial 2-hour session, the patient is asked to enter the sound lab where their body's "fundamental frequency" can be found and converted into a tone that will be used for their treatment. The process for identifying one's fundamental frequency is described in more detail later in this document. However, simply stated, finding one's fundamental frequency involves identifying the point at which the parasympathetic nervous system and sympathetic nervous system are at an equilibrium through a biofeedback device known as a heart rate variability monitor. Once identified, this fundamental frequency is converted into a tone, and subsequently, a .wav file that will be played back to the patient. The patient is then asked to lie on a table while listening to this tone over a 30-minute period. At the end of the treatment, the patient is then allowed to remain on the table for a few extra minutes to allow for the gradual shift from a relaxed state to a more consciously aware state. The patient is then given an audio copy (.wav file) of their tone to take home, where they are asked to listen to it nightly over the next 21 days. The overall time of each nightly "listening session" is 30 minutes.

The Center for Neuro acoustic Research (CNR) bases its theory of the utility of Bio-tuning in the physics of acoustics, which suggests that every material substance has a resting resonance frequency [36]. For example, it is this principle that suggests that a glass of wine will vibrate when exposed to a certain tone. In addition, CNR takes advantage of the principle of coupled oscillators, which suggests that elements will resonate together in order to produce the most amount of work possible and expend the least amount of energy. CNR suggests that when a material is exposed to a frequency that matches its own frequency, it begins to "sympathetically vibrate" (i.e., coupled oscillation), and consequently, reaches its highest possible state of resonance. This state is the point at which the cells increase their metabolic state, and thus, incite a "super healing" state (p.1&2). These principles provide the foundation for the development of the Bio-tuning equipment. Using these principles, researchers at CNR report observing a balancing of the parasympathetic and sympathetic divisions of the Autonomic Nervous System (ANS) in their patients after exposing them to their fundamental frequency for a period of time [35]. They argue that the balancing of the autonomic nervous system eliminates the symptoms of stress. Despite these arguments, to date there have been no studies to support them.

Statement of the Problem

A search was conducted for studies that examine the effects of sound on the mental states of individuals. While their results appear promising with regard to the reduction of stressful symptoms, the number of studies found were relatively few [32-35]. Fortunately, these studies lend support to the brainwave entraining method utilized by the Bio-tuning procedure. As such, the current study serves to add further support to previous studies that have demonstrated an effect of sound on the mental states of individuals. This study also serves to examine the effectiveness of Bio-tuning as a tool for stress reduction. The purpose of this study is to test the effectiveness of Bio-tuning in reducing symptoms of depression and anxiety, improving sleep quality, and improving the overall quality of life among a sample of participants. As it only requires one in-office session, Bio-tuning places less demands on the time of patients compared to other methods of stress reduction. CNR reports that they have seen positive patient compliance as a result of the noninvasive and low-cost method utilized by the Bio-tuning procedure [35]. However, as with its effectiveness, these claims have not been subjected to systematic studies as of yet. Given the challenges posed by the COVID-19 pandemic, a treatment that is effective at reducing stress that is both time and cost efficient desirable. CNR reports that it takes about 21 days of repeated exposure to an experience to develop a "Learned Neurological Response"-that is, a new pattern of functioning within the nervous system [35]. Thus, after about 21 days of repeated exposure to a person's primary frequency, CNR posits that this new frequency pattern will become entrained in their brain, thereby becoming the prominent brainwave frequency [35]. This results in the experience of homeostasis in the ANS, and thus, a reduction of stress-related symptoms. As such, it is hypothesized that the use of the Bio-tuning equipment will result in a significant reduction of stress after a period of use of 21 days. For this study, we defined stress as a person's reaction to a disturbance in their daily life. Stress was measured by changes in sleep quality, quality of life, anxiety, and depression scores. In addition, we also examined the role that the COVID-19 pandemic played in worsening perceptions of stress [37].

The Present Study

Stress is a regular part of our lives. In the short term, it can be beneficial as it may help us be more efficient in our work or perhaps escape danger. However, over time, stress can be taxing and may start to take a toll on our health, both physically and mentally. Given the current worldly state of affairs, stress has made its way into our lives at a more significant degree. As we find ourselves living through a global pandemic, we are challenged in ways that many never thought possible. In our own way, many of us are still adapting to the new reality of life that involves face mask wearing, disinfecting, and social distancing, with the added task of being ordered to stay at home. However, amid these challenges is the toll that this change has taken on our emotional health. In addition to the effort involved with incorporating these new behaviors into our daily lives, the public is also being tasked with managing the distress that results from those changes. As one can imagine, this results in an experience of stress that is beyond our "normal" day-to-day experience. As such, a method for stress reduction that is both effective and efficient is called for. The present study applied a within-subject design in which data was collected at three time points: before and after a 21-day period of Bio-tuning use, as well as mid-treatment measures to assess changes in sleep patterns. Measurements included five dependent variables overall: anxiety, de-

pression, sleep quality, quality of life, and COVID-related stress. The current study examined the effectiveness of Bio-tuning as a stress reduction tool among a random sample of participants over the age of 18. Bio-tuning is a stress reduction tool that works to restore the homeostatic balance between the sympathetic nervous system and the parasympathetic nervous system within the autonomic nervous system. This is done by identifying the body's resonant frequency at which the SNS and PSNS are at an equilibrium, calibrating that frequency to target a specific conscious state, and delivering that frequency as a sustained tone back to the participant. Over time (i.e., a period of 21 days), the body's normal mode of functioning will begin to resonate at this new frequency, thereby resulting in a more efficient use of the body's resources. As a result, the individual will experience a reduction in stress. For this study, stress was measured by symptoms of anxiety and depression, sleep quality, perceived quality of life, and stress resulting from COVID-related concerns. As such, this study examined the following five hypotheses:

1) Hypothesis 1: Using Bio-tuning will result in a reduction of anxious symptoms.
2) Hypothesis 2: Using Bio-tuning will result in a reduction of depressive symptoms.
3) Hypothesis 3: Using Bio-tuning will result in improved sleep quality.
4) Hypothesis 4: Using Bio-tuning will result in an increased perceived quality of life.
5) Hypothesis 5: Using Bio-tuning will result in a reduction of COVID-related stress.

Methods

Participants

The current study analyzed data from a sample of participants who underwent Bio-tuning for a variety of presenting problems in a private clinic setting. The sample consisted of adults over the age of 18 from diverse backgrounds not limited by demographic variables such as gender, ethnicity, income, education, sexual orientation, religion, disability status, etc. This study employed a convenience sampling method to obtain participation from a total of 34 participants who were seeking Bio-tuning treatment and agreed to participate in this study. The number was determined by a power analysis (α=0.05, β=0.08, Correlation among repeated measures: default=0.5.

Dr. Linda Yniguez provided this study's primary author access to data of those clients who agreed to undergo the Bio-tuning procedure and share their data for research purposes. In addition to the pre-, mid-, and post-treatment measures, each participant was provided with a consent form informing them that their data will be used for research purposes.

Measures

Demographics Questionnaire: A demographics questionnaire was provided that contained general information about the participants, such as their health and work history. This questionnaire was also kept anonymous and served to ensure that no patients were under the age of 18.

Beck Anxiety Inventory (BAI): The BAI is a 21-item self-report inventory measuring common cognitive and somatic symptoms of anxiety. Its score is derived through the summation of the scores on the 21 items. Each item requests that the participant answer along a Likert scale ranging of 0-3. Scores are then totaled and evaluated in reference to overall range of 0-63. Total scores will fall into three ranges: minimal anxiety (0-7), mild anxiety (8-15), moderate anxiety (16-25) and severe anxiety (26-63). The BAI has been found to have good discriminant validity and is written for persons with a grade level beyond eighth grade *(Halfaker, et al.,* 2011*; Kabacoff et al.,* 1997*)*. Higher scores are reflective of more severe anxious symptoms.

Beck Depression Inventory, Second Edition (BDI-II): The BDI-II is a common screening tool for symptoms of depression. It is a 21-item multiple choice, self-report questionnaire that measures a person's depression severity. Each item on the BDI-II is designed to assess how the person has been feeling in the last week. Each item consists of four possible answers that range in intensity and are scored accordingly. For example, under sadness a person indicating "I do not feel sad" would obtain a score of 0, while a person reporting "I am so sad or unhappy I can't stand it" would obtain a score of 3. Accordingly, each item is scored on a Likert scale with a range of 0-3. Similar to the BAI, scores from all items are summed resulting in an overall score that ranges from 0-63. Overall scores will fall in one of four ranges: minimal depression (0-13), mild depression (14-19), moderate depression (20-28), and severe depression (29-63). Higher scores are reflective of more severe depressive symptoms.

World Health Organization Quality of Life-BREF (WHOQOL-BREF) Scale: The WHOQOL-BREF scale is a short quality of life assessment based on the comprehensive 100-question quality of life assessment (WHOQOL-100) developed by the World Health Organization (The WHOQOL Group, 1998) [37]. Its development is intended for ease of use in the field. The WHOQOL-BREF is a 26-item self-report questionnaire which examines the subjective quality of the four domains of a person's quality of life (QOL): physical health, psychological health, social relationships, and environment. Additionally, a person's overall QOL and health in general are evaluated. Items are scored on a Likert scale range of 1 to 5. Higher scores indicate a reportedly higher subjective quality of life. Only three of the four domains within the WHOQOL-BREF were used. Indeed, all items within the WHOQOL-BREF were included except for those relating to the domain of the environment.

COVID-19 Student Stress Questionnaire (CSSQ)-Modified: The CSSQ is a 7-item self-report questionnaire used to assess stress related to COVID-19 among university students (*Zurlo, et al.,* 2020). Each item is scored on a Likert scale range of 0-4. A score of 0 indicates that the respondent perceives the item's content as "not

stressful at all," while a score of 4 suggests the item's content is perceived as "extremely stressful." The summation of all items results in a Global Stress score. The Global Stress score is a measure of stress severity about COVID-related stress. This score falls into one of three ranges: Low (0-6), Average (7-15), or High (16-28). A modified version of the CSSQ was used. As this measure was originally designed to assess college students, the content relating to academic life was revised so that it is relevant to both school and work. For instance, item 4 reads "How do you perceive the relationships with your university colleagues during this period of the COVID-19 pandemic?" As such, this question was changed to read "How do you perceive the relationships with your work/school colleagues during this period of the COVID-19 pandemic?"

Sleep quality Questionnaire (SQQ)-Modified: The SQQ is a 10-item measure that assesses sleep quality (Kato, 2014). This measure is comprised of two separate factors. Four items within the measure assess sleep difficulty and six items assess daytime sleepiness. Respondents use a 5-point Likert scale ranging from 0 (strongly disagree) to 4 (strongly agree) to rate each item on the measure. The original instructions asked respondents to answer each item in terms of their experience over the past month. However, this study obtained a version that modified that time span and asked the respondents to answer each item in terms of their experience over the past week. Participants were asked to choose from the following response choices: strongly disagree, disagree, not sure, agree, strongly agree. Higher scores on this measure indicate poorer sleep quality.

Procedures

Upon meeting at the sound lab, participants were given a consent form for their review and signature. The consent form contained information such as risks and benefits of the Biotuing treatment, contact information, and information regarding the sharing of their response data for research purposes. In addition to consent, the participants were also asked to fill out pre-treatment questionnaires that asked about demographics and established a baseline of their levels of anxiety, depression, quality of life, sleep quality, and COVID-related stress Upon completing this task, participants were then escorted into the sound room by *Dr. Yniguez* for a follow-up interview and treatment. This interview involved a brief physical health intake and allowed time for *Dr. Yniguez* to answer any questions the participant might have had regarding the treatment. At the conclusion of this interview and with the participant's consent, *Dr. Yniguez* started the treatment process. With the participant's permission, Dr. *Yniguez* pasted the sensors of Heart Rate Variability monitor (HRV) to the skin on the back of the participant's wrist. At this point, the HRV began to capture the participant's ANS functioning on a moment-to-moment basis and displayed it on a computer screen. Specifically, the display captured the participant's current SNS/PSNS functioning, in addition to their current heart rate. The participant was then asked to lie on the sound table. The sound table has been specifically designed to receive input from the computer that will instruct it to vibrate mildly at specified frequencies. While lying on the table, the participants began to feel the vibration, which caused their nervous system to resonate sympathetically. The vibrational frequencies ranged from 50Hz to 220Hz. During this initial exposure to the range of vibrational frequencies, the participant's moment-to-moment ANS response was monitored by the HRV. As the participant experienced this sweep of frequencies, their ANS gradually moved from its current resting frequency toward its primary frequency (i.e., the state of perfect balance between both divisions of the ANS).

Through the use of the HRV monitoring system, the participant's primary frequency was captured as a value that was then used to calibrate the tone for the participant's personalized delta wave. This tone was then extracted as a 30-minute .wav file. In addition, primordial sounds (e.g., nature sounds) were overlaid over the delta tone. The primordial sounds were also calibrated to match the participants' delta frequency. At the end of this process, the participant was asked to remain on the table for the remainder of the Bio-tuning treatment. At this point, the participant was given a set of noise-cancelling headphones that were used to play back the tone over the course of the next 30 minutes. The participant remained in this resting state for the remainder of the 30-minute treatment session. At its conclusion, the participant was asked to remove the headphones and was allowed a few extra minutes to gradually shift from a calm and relaxed state to an aware state. *Dr. Yniguez* also remained available to answer any final questions the participant had about the procedure. Finally, the participants were instructed to return to her office on the 10th day of treatment to complete the sleep quality questionnaire so that changes in their sleep quality could be tracked. Accordingly, on the 22nd day after treatment patients were instructed to return to their office to complete post-treatment measures. The post-treatment measures included many of the same items answered on their first day of treatment, with additional items asking about their overall experience and adherence to the Bio-tuning treatment protocol. Upon being released from the sound lab, the participants were provided a set of treatment instructions that they were required to adhere to over the next 21-days. The participants were instructed to find a space in their home that is free of distraction and comfortable that will allow them to relax for the entire 30-minute treatment session. The ideal space would be a safe room where they feel comfortable enough to lie on their back, close their eyes and focus on the sound emanating from the headphones. Accordingly, a functional set of noise cancelling headphones and equipment to play a .wave file were also necessary to participate in the treatment. As such, *Dr. Yniguez* met with each participant to ensure they had the necessary equipment to engage in the treatment. Finally, participants were instructed to listen to their Bio-tuning file lying in bed and preparing to fall asleep. At the end of the 30-minute treatment, the participants were free to remove the headphones and resume their normal nightly activities.

Results

Descriptive Statistics

The sample (N=34) predominantly consisted of women, with

65% (N=22) of the participants being female and 35% (N=12) being male. Participants' ages ranged between 18 and 72. The breakdown was as follows: one in their late teens, seven in their 20s, nine in their 30s, seven in their 40s, two in their 50s, five in their 60s and two in their 70s. Regarding marital status, 38% (N=13) were married, 29% (N=10) were single, and 18% (N=6) were divorced. Nearly 80% of the sample reported having either a high school (N=10), bachelors (N=11), or master's degree (N=6). Over half of the sample (53%; N=18) consisted of persons from the city of Whittier, California. An additional 42% (N=14) reported residing in other areas of Los Angeles County. More than half (65%) reported their annual household income ranging between $50, 000 and $200,000 (N=22). A large majority of participants (85%; N=29) reported their ethnicity as Hispanic. Regarding their expectations of the Bio-tuning procedure actually improving their well-being, 44% (N=15) indicated that they believed the procedure would provide "some" help, while 38% (n=13) reported that they "very much" believed it would help. Finally, regarding adherence, 77% (N=26) of participants reported adhering to the treatment (i.e., listening to their track for the entire 21 days as instructed). The distribution of gender, marital status, education, race, income, treatment adherence, and expectation of treatment effectiveness can be seen in (Table 1a-1g).

Table 1: Descriptive Statistics.

Table 1a:

Gender		
	Frequency	Percent
Male	12	35.3
Female	22	64.7
Total	34	100

Table 1b:

Marital Status		
	Frequency	**Percent**
Married	13	38.2
Single	10	29.4
Divorced	6	17.6
Life partner	1	2.9
Separated	1	2.9
Widowed	3	8.8
Total	34	100

Table 1c:

Education		
	Frequency	**Percent**
Some high school	1	2.9
High school	10	29.4
B.A.	11	32.4
M.A	6	17.6
Doctorate	2	5.9
Trade school	3	8.8
Prefer not to say	1	2.9
Total	34	100

Table 1d:

Ethnicity		
	Frequency	**Percent**
Black-non Hispanic	1	2.9
Hispanic	29	85.3
Asian/pacific islander	1	2.9
White-non Hispanic	2	5.9
Other	1	2.9
Total	34	100

Table 1e:

Income		
	Frequency	**Percent**
<$25,000	1	2.9
$25-50,000	4	11.8
$50-100,000	11	32.4
$100-200,000	11	32.4
>$200,000	1	2.9
Prefer not to say	5	14.7
Total	33	97.1
No data	1	2.9
Total	34	100

Table 1f:

Treatment Adherence		
	Frequency	**Percent**
Yes	26	76.5
No	7	20.6
No Data	1	2.9
Total	34	100

Table 1g:

Expectation of Tx Efficiency		
	Frequency	**Percent**
Little	2	5.9
Some	15	44.1
Very much	13	38.2
No data	4	11.8
Total	34	100

Main Findings

Results from the paired-samples t tests that compared pre-treatment and post-treatment anxiety, depression, sleep quality, perceived COVID-related stress, and perceived quality-of-life

scores are shown in Tables 2a-2i. Statistical significance was found in the variables of anxiety, depression, and sleep quality. Regarding quality of life, statistical significance was found on the physical health and psychological domains of the measure. No significant changes were found in stress related to the COVID-19 pandemic post-treatment.

Table 2: Paired-samples t test.

Table 2a:

Symptoms of An*iety			
	Mean	**SD**	**N**
Pre	8.882353	8.605019	34
Post	6.411765	7.266017	34
t(33)=-2.004, p=.0266, one sided			

Table 2b:

Symptoms of Depression			
	Mean	**SD**	**N**
Pre	12.02941	8.447647	34
Post	6.823529	7.354531	34
t(33)=4.023, p<.001, one sided			

Table 2c:

Sleep Quality			
	Mean	**SD**	**N**
Pre	18.44118	8.773898	34
Post	11.23529	7.356954	34
t(33)=4.051, p<.001, one sided			

Table 2d:

QOL-Health			
	Mean	**SD**	**N**
Pre	26.41176	4.171459	34
Post	28.91176	3.824702	34
t(33)=3.020, p=.002, one sided			

Table 2e:

QOL-Psychological			
	Mean	**SD**	**N**
Pre	20.51515	4.221383	33
Post	21.75758	3.544629	33
t(32)=1.979, p<.05, one sided			

Table 2f:

QOL-Social Relationships			
	Mean	**SD**	**N**
Pre	10.82353	2.081238	34
Post	10.97059	2.56415	34

Table 2g:

QOL-Perceptions on Overall QOL			
	Mean	**SD**	**N**
Pre	4.147059	0.702047	34
Post	4.147059	0.702047	34

Table 2h:

QOL-Perceptions on Overall Physical Health			
	Mean	**SD**	**N**
Pre	3.205882	1.148897	34
Post	3.352941	1.097721	34

Table 2i:

COVID-19-Related Stress			
	Mean	**SD**	**N**
Pre	6.941176	5.902959	34
Post	5.970588	6.575829	34

Hypothesis 1-There is evidence that undergoing the Bio-tuning treatment improves symptoms of anxiety such that participants' mean scores were significantly improved after treatment ($t_{(33)}$=-2.004, p= .0266, one sided). Hypothesis 2-There is evidence that undergoing the Bio-tuning treatment improves symptoms of depression such that participants' mean scores were significantly improved after treatment ($t_{(33)}$=4.203, p<.001, one sided). Hypothesis 3-There is evidence that undergoing the Bio-tuning treatment improves sleep quality such that participants' mean scores were significantly improved after treatment ($t_{(33)}$=4.051, p<.001, one sided). Hypothesis 4-There is evidence that undergoing the Bio-tuning treatment improves perceived quality of life with respect to facets of physical ($t_{(33)}$= 3.020, p=.002, one sided) and psychological ($t_{(32)}$= 1.979, p<.05, one sided) health such that participants' mean scores were significantly improved after treatment; however no evidence was found indicating that undergoing Bio-tuning treatment improved overall perceptions of quality of life ($t_{(33)}$= 0, n.s.) and physical health ($t_{(32)}$= 1.979). There is no evidence that undergoing Bio-tuning treatment significantly improved social relationships ($t_{(33)}$=1.221, n.s). Hypothesis 5-There is no evidence that undergoing Bio-tuning treatment significantly improved symptoms of stress related to COVID-19 ($t_{(31)}$=1.238, n.s.).

Repeated-Measures Analyses

Since the protocol for the Bio-tuning procedure used in this study was set to Delta I (i.e., the protocol that targets deep-restorative sleep), researchers were interested in examining changes in sleep scores over the three-week treatment period. As such, the data were submitted to a one-way repeated measures ANOVA. Sphericity was met and, thus, no correction has been applied. There was a significant effect of time on sleep quality ($F_{(2,60)}$=12.227, p<0.001). Pairwise comparisons indicate a significant improve-

ment in poor sleep quality from pre-treatment (m=17.935, SE= 1.437) to mid-treatment (m=11.452, SE=1.359) and pre-treatment to post-treatment (m=10.355, SE=1.1114). The difference between mid- and post-treatment was not significant. Results examining changes in sleep scores over time are shown in (Figure 1).

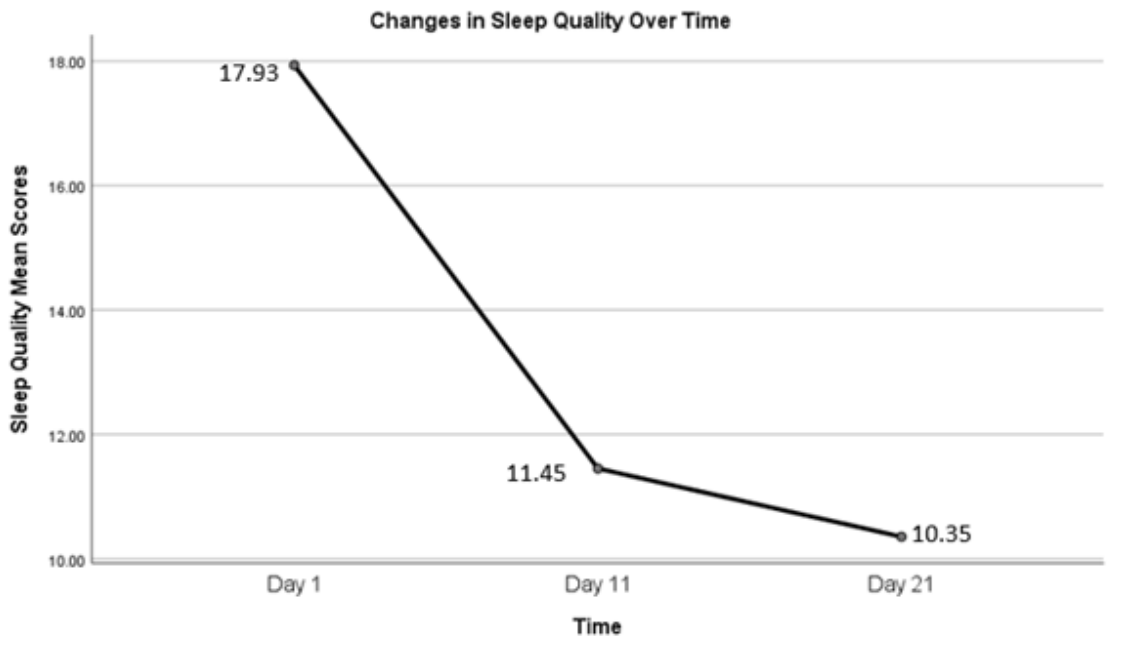

Figure 1: One-way repeated measures ANOVA.

Correlational Analyses

Table 3: Pearson Correlation Matrix.

		Physical Health Perception (post-tx)	Psychological Health Perception (post-tx)	Social Relationships Perception (post-tx)	COVID-19 Stress (post-tx)	Sleep Quality (post-tx)	Sleep Quality (mid-tx)	Depression (post-tx)	Anxiety (post-tx)	Expectation that Blotuning will work (pre-tx)	Belief that Biotuning has helped (mid-tx)	Expectation that Bictuning will continue to help moving forward (mid-tx)
Physical Health Perception (post-tx)	Pearson Correlation	1	.599**	.528**	-0.217	-.537**	-0.135	-.449**	-.318**	-0.129	0.22	0.105
	Sig (1-tailed)		0	0.001	0.117	0.001	0.234	0.004	0.033	0.249	0.13	0.295
	N	34	33	34	32	34	31	34	34	30	28	29
Psychological Health Perception (post-tx)	Pearson Correlation	.599**	1	.632**	-.453**	-.470**	-.506**	-.535**	-.554**	-0.264	.336*	122
	Sig (1-tailed)	0		0	0.005	0.003	0.002	0.001	0	0.083	0.043	269
	N	33	33	33	31	33	30	33	33	29	27	28

Social Relationships Perception (post-tx)	Pearson Correlation	.528**	.632**	1	-.479**	-0.263	-.309*	-.407**	-.417**	-0.073	0.165	0.128
	Sig (1-tailed)	0.001	0		0.003	0.066	0.045	0.008	0.007	0.35	0.201	0.254
	N	34	33	34	32	34	31	34	34	30	28	29
COVID-19 Stress (post-tx)	Pearson Correlation	-0.217	-.453**	-.479**	1	0.214	.631**	.495**	.669**	.398*	-0.198	0.76
	Sig (1-tailed)	0.117	0.005	0.003		0.12	0	0.002	0	0.018	0.166	0.353
	N	32	31	32	32	32	29	32	32	28	26	27
Sleep Quality (post-tx)	Pearson Correlation	-.537**	-.470**	-0.263	0.214	1	.443**	0.276	0.204	0.018	-.423*	-0.285
	Sig (1-tailed)	0.001	0.003	0.066	0.12		0.006	0.057	0.123	0.463	0.012	0.067
	N	34	33	34	32	34	31	34	34	30	28	29
Sleep Quality (mid-tx)	Pearson Correlation	-0.135	-.506**	-.309*	.631**	.443**	1	.429**	.536**	.492**	-0.236	0.031
	Sig (1-tailed)	0.234	0.002	0.045	0	0.006		0.008	0.001	0.004	0.114	0.436
	N	31	30	31	29	31	31	31	31	28	28	29
Depression (post-tx)	Pearson Correlation	-.449**	-.535**	-.407**	.495**	0.276	.429**	1	.514**	.440**	-0.085	0.032
	Sig (1-tailed)	0.004	0.001	0.008	0.002	0.057	0.008		0.001	0.008	0.333	0.434
	N	34	33	34	32	34	31	34	34	30	28	29
Atxliets (post-tx)	Pearson Correlation	-.318*	-.554**	-.417**	.669**	0.204	.536**	.514**	1	.456**	-0.232	0.085
	Sig (1-tailed)	0.033	0	0.007	0	0.123	0.001	0.001		0.006	0.118	0.331
	N	34	33	34	32	34	31	34	34	30	28	29
Expectation that Biotuning wil, work(pre-tx)	Pearson Comellation	-0.129	-0.264	-0.073	.398*	0.018	.492**	.440**	.456**	1	0.127	.362*
	Sig (1-tailed)	0.249	0.083	0.35	0.018	0.463	0.004	0.008	0.006		0.263	0.029
	N	30	29	30	28	30	28	30	30	30	27	28
Belief that Bictuning has helped (mid-tx)	Pearson Correlation	0.22	.336*	0.165	-0.198	-.423*	-0.236	-0.085	-0.232	0.127	1	.613**
	Sig (1-tailed)	0.13	0.043	0.201	0.166	0.012	0.114	0.333	0.118	0.263		0
	N	28	27	28	26	28	28	28	28	27	28	28
Expectation that Biotuning will continue to help moving forward (mid-tx)	Pearson Comelation	0.105	0.122	0.128	0.076	-0.285	0.031	0.032	0.085	.362*	.613**	1

	Sig (1-taled)	0.295	0.269	0.254	0.353	0.067	0.436	0.434	0.331	0.029	0	
	N	29	28	29	27	29	29	29	29	28	28	29

Note*: **Correlation is significant at the 0.01 level (1- Tailed).

*Correlation is significant at the 0.05 level (1- Tailed).

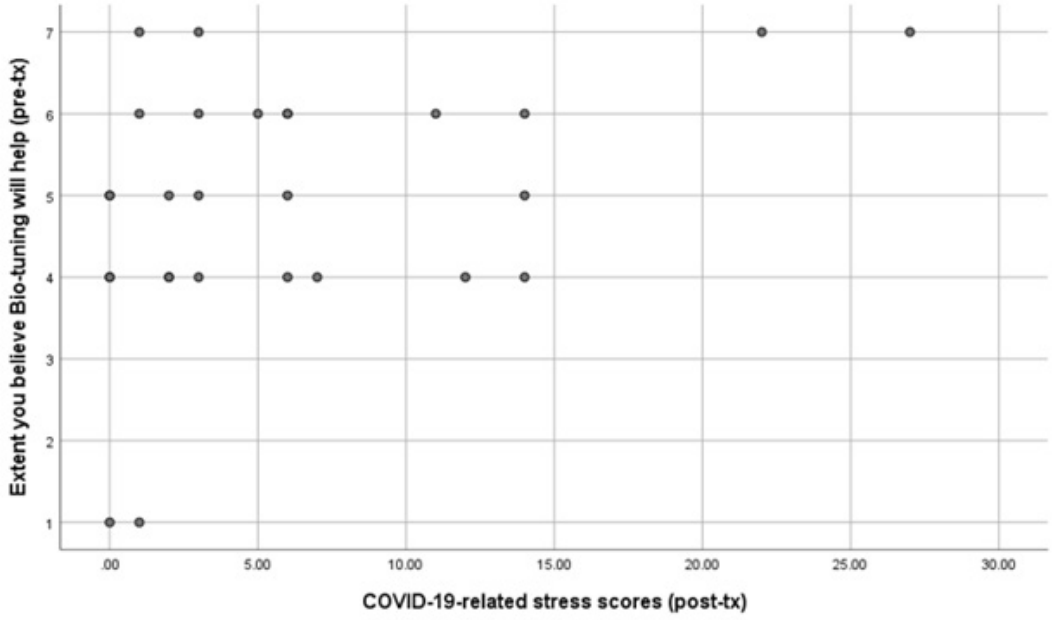

Figure 2a: Pearson Correlations.

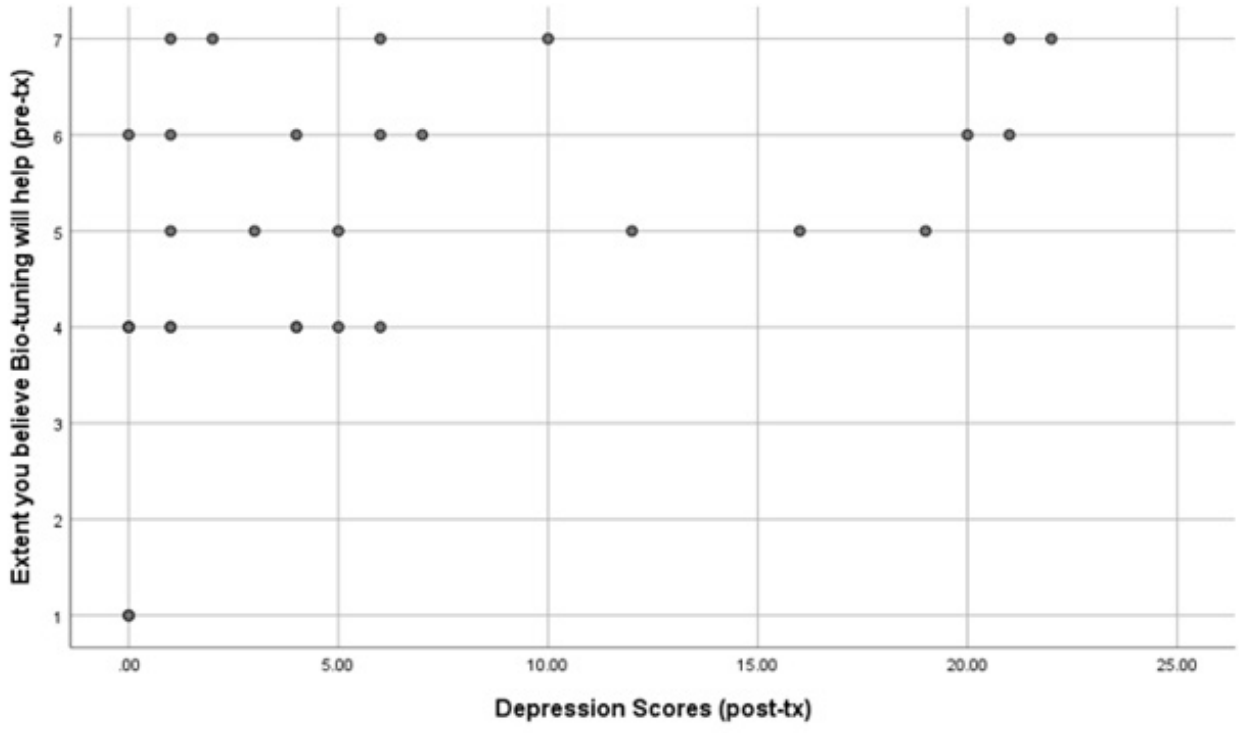

Figure 2b

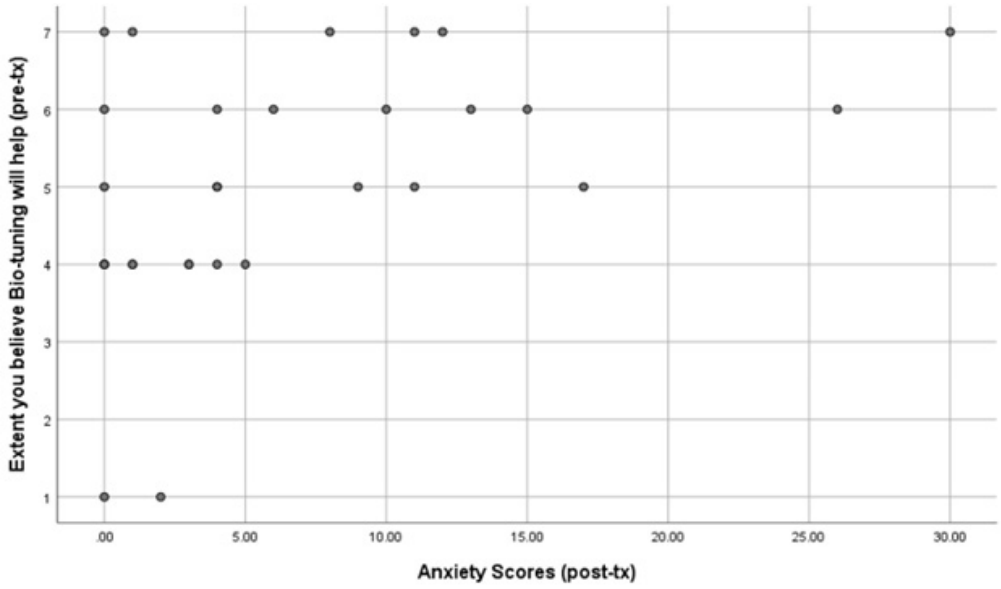

Figure 2c

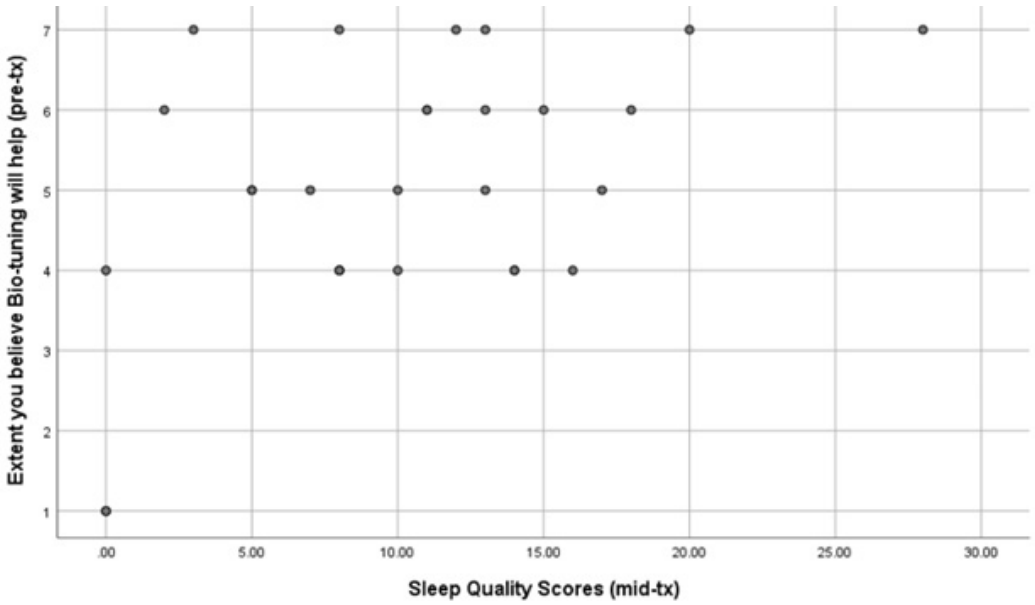

Figure 2d

The Pearson correlations were computed to examine relationships among the hypothesized variables. These analyses can be seen in Table 3. Analyses revealed significant relationships between patient expectations of the Bio-tuning procedure, sleep quality and COVID-related stress and our hypothesized variables. Those correlations are described in further detail below. Regarding expectations, analyses revealed a significant positive correlation between pre-treatment expectation scores and post-COVID-19 stress scores (r_p=.398, p =.018, one tailed), post-treatment depression scores (r_p=.440, p =.008, one tailed), post-anxiety scores (r_p=.456, p =.006, one tailed), and sleep quality at mid-treatment (r_p=.492, p =.004, one tailed). Results comparing expectation to post-COVID-19 stress, post-depression, post-anxiety, and mid-treatment sleep quality scores can be seen in Figures 2a-2d. Analyses also revealed significant relationships between participants' belief that the treatment was working to expectations and sleep quality. A significant positive relationship was found between participant reports of Bio-tuning helping to reduce stress and expectations that Bio-tuning would continue to help reduce stress, at mid-treatment (r_p=.613, p<.0001, one tailed). However, a significant inverse relationship was revealed between reports of Bio-tuning helping with stress reduction at mid-treatment and poor sleep quality at post-treatment (r_p= -0.423, p =.012, one tailed). Thus, any improvement they may have experienced was not as pronounced at mid- to post-treatment,

as compared to pre- to mid-treatment. Results comparing the relationship between participants' belief that Bio-tuning has helped at mid-treatment and expectations that it will continue to help (beyond mid-treatment) can be seen in Figure 3a, while results comparing the relationship between participants' belief that Bio-tuning has helped at mid-treatment and participants' sleep quality at post-treatment can be seen in Figure 3b.

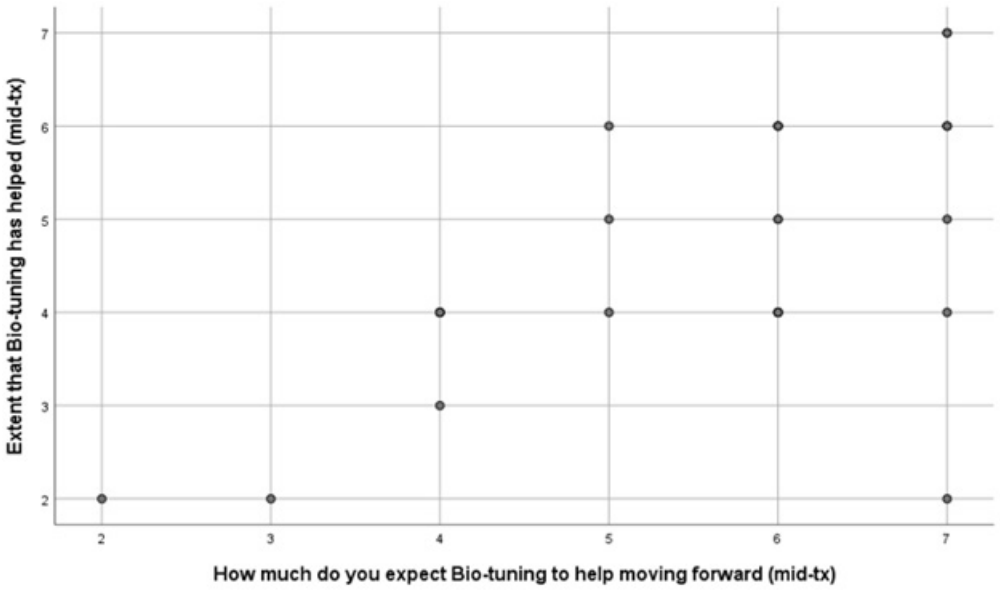

Figure 3a: Pearson Correlations.

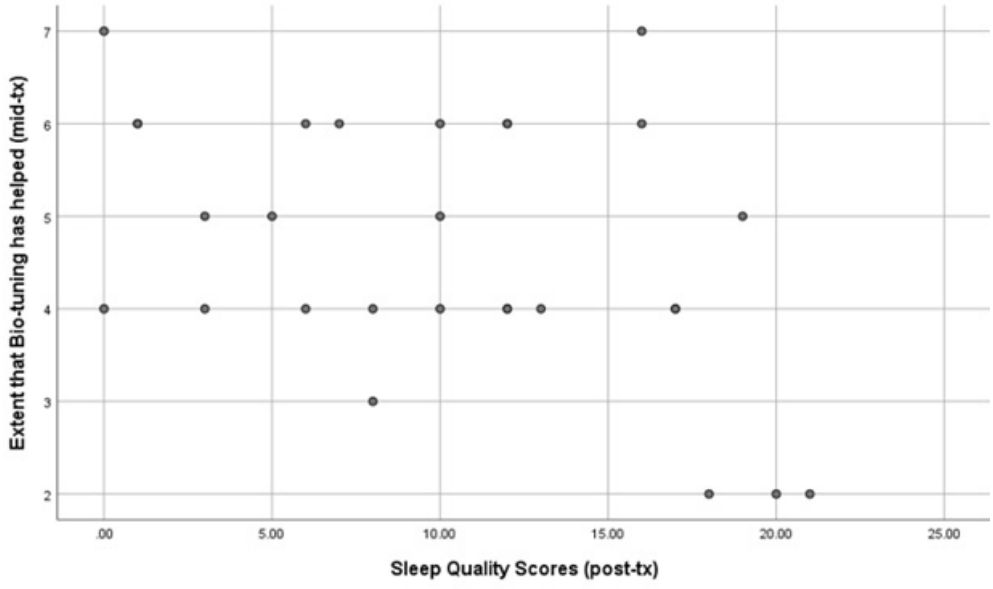

Figure 3b

Analyses revealed significant relationships between sleep quality and COVID-related stress, quality of life, depression and anxiety. Significant inverse relationships were found between poor sleep quality at mid-treatment and perceived psychological health (r_p= -.506, p =.002, one tailed) and social relationships (r_p= -.309, p =.045, one tailed) at post-treatment, as well as significant positive relationships between poor sleep quality at mid-treatment and COVID-related stress (r_p=.631, p<.0001, one tailed), poor sleep quality (r_p=.443, p =.006, one tailed), depression (r_p=.429, p =.008, one tailed), and anxiety (r_p=.536, p =.001, one tailed) at post-treatment. These results suggest that as sleep quality improved at mid-treatment, improvements were also shown in perceived psychological health and social relationships, depression, anxiety, and COVID-related stress at post-treatment. Further, significant inverse relation-

ships were found between poor sleep quality at post-treatment and quality of life in both the physical (r_p= -.537 p =.001, one tailed) and psychological (r_p= -.470, p =.003, one tailed) health domains, suggesting that as sleep quality improved at the end of treatment, perceptions of physical and psychological health also improved. Although improvements are shown in both mid- and post-treatment regarding poor sleep quality and its associated variables, this data supports previously mentioned analyses which shows significant improvements occurring between pre- and mid-treatment. Results comparing relationships between poor sleep quality, COVID-related stress, quality of life, depression, and anxiety can be seen in Figures 4a-4h.

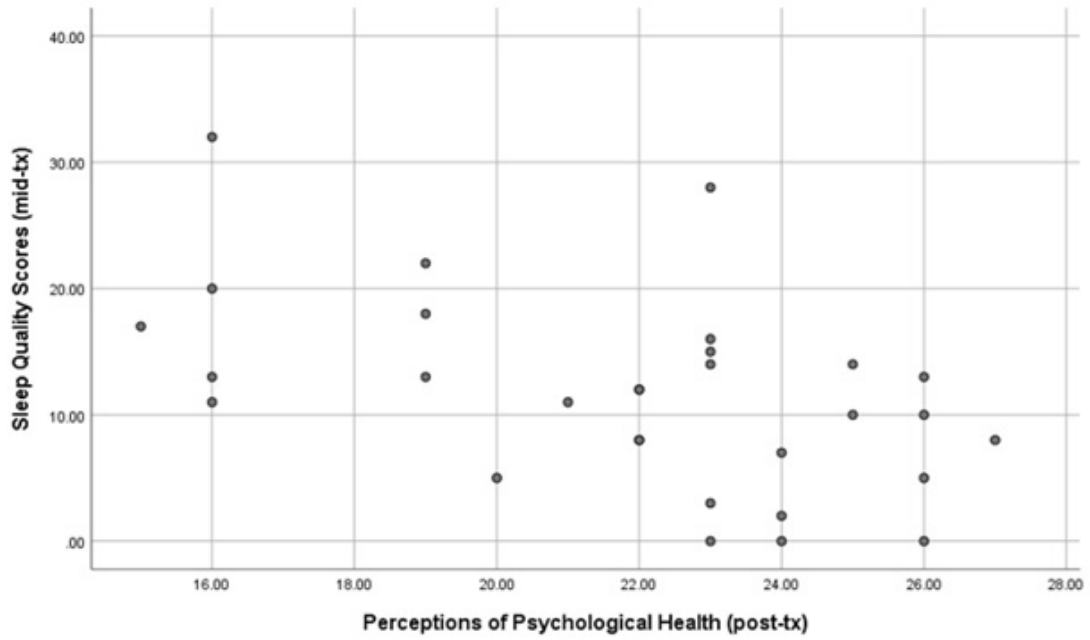

Figure 4a: Pearson Correlations.

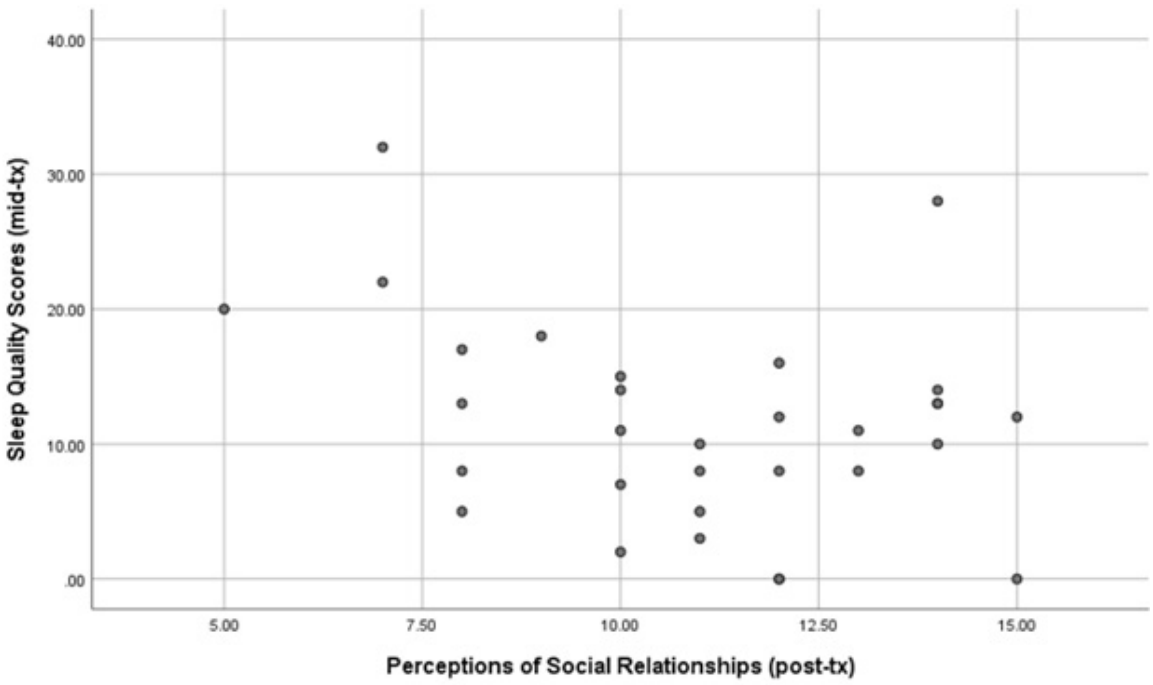

Figure 4b

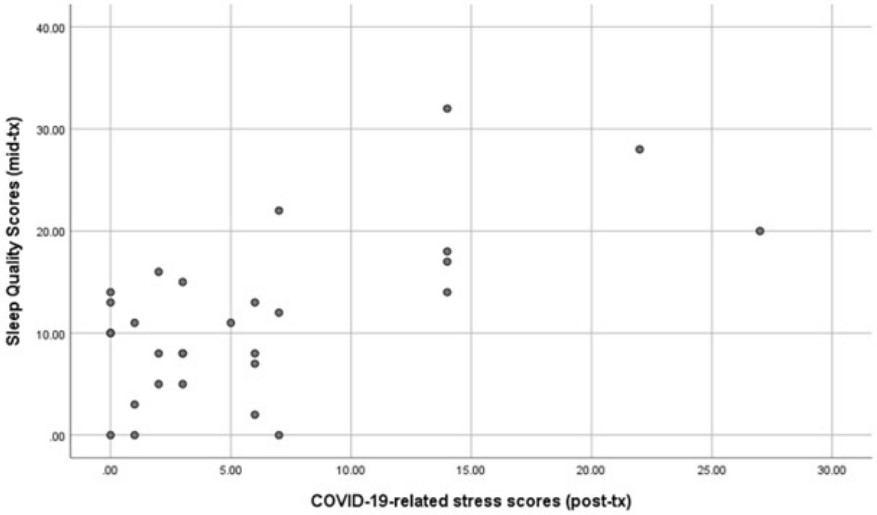

Figure 4c

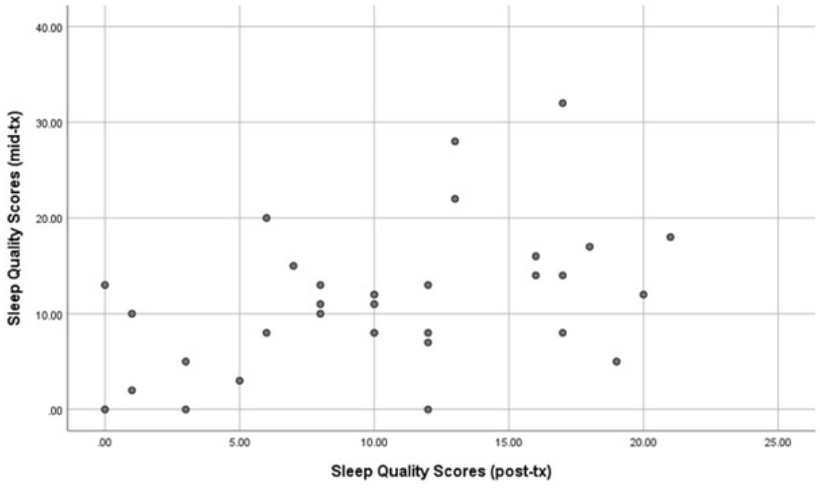

Figure 4d

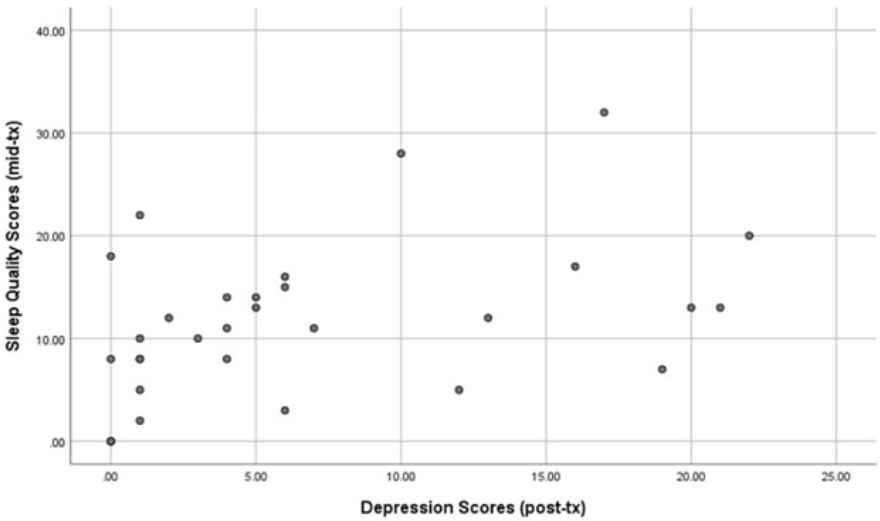

Figure 4e

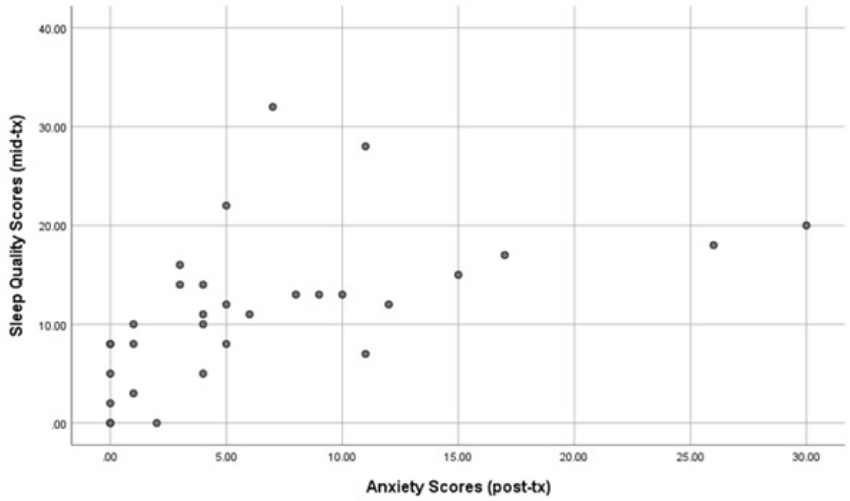

Figure 4f

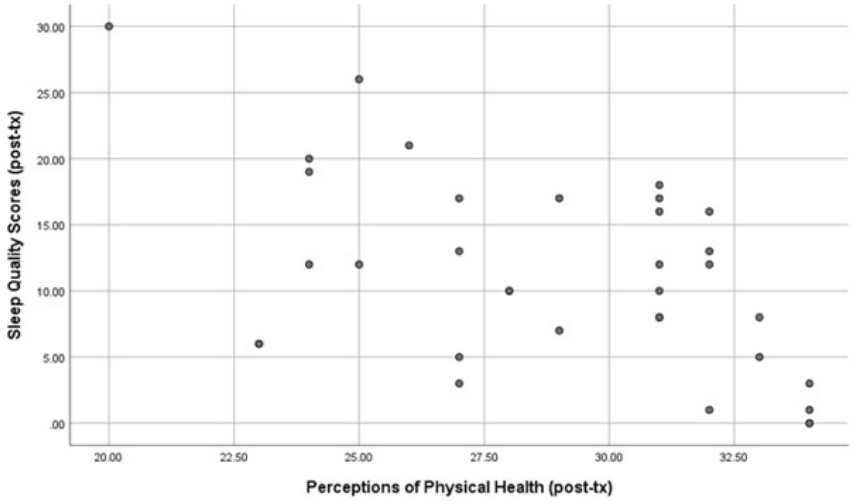

Figure 4g

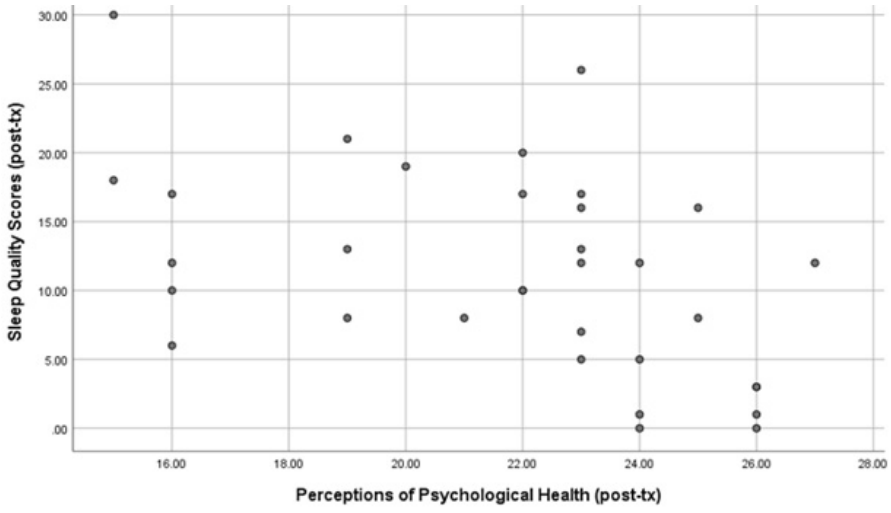

Figure 4h

A final note regarding the COVID-related stress variable. Although no significance was found between pre- and post-treatment measures of COVID-related stress, this variable was shown to be significantly correlated with several other variables. Namely, significant inverse relationships were found between COVID-related stress and quality of life in the domains of psychological health (r_p= -.453 p =.005, one tailed) and social relationships (r_p= -.479 p =.003, one tailed) at post-treatment, suggesting that as COVID-related stress decreased, perceptions of psychological health and social relationships improved. Similarly, significant positive relationships were found between post-treatment COVID-related stress and sleep quality at mid-treatment (r_p= .631 p<.0001, one tailed), as well as depression (r_p= .495 p<.0002, one tailed) and anxiety (r_p= .669 p<.0001, one tailed) at post-treatment, suggesting that as COVID-related stress improved, depression, anxiety, and sleep quality improved as well. Results comparing the relationships between COVID-related stress, quality of life, sleep quality, depression and anxiety can be seen on Figures 5a-5e.

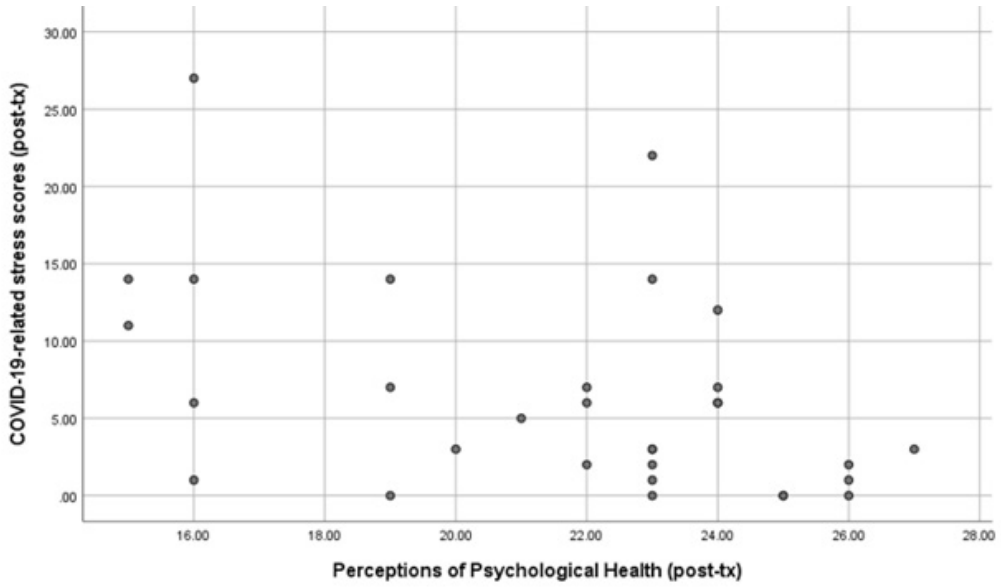

Figure 5a: Pearson Correlations.

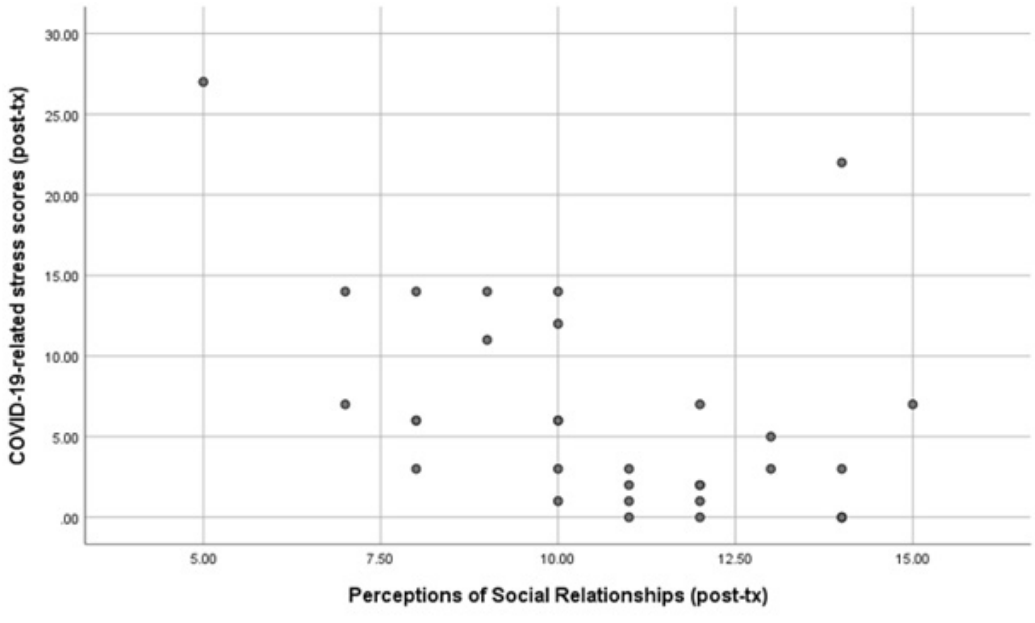

Figure 5b

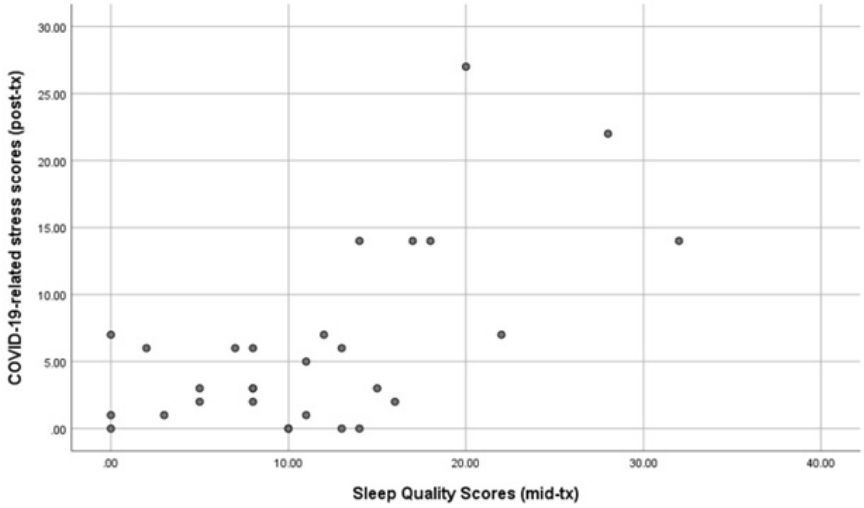

Figure 5c

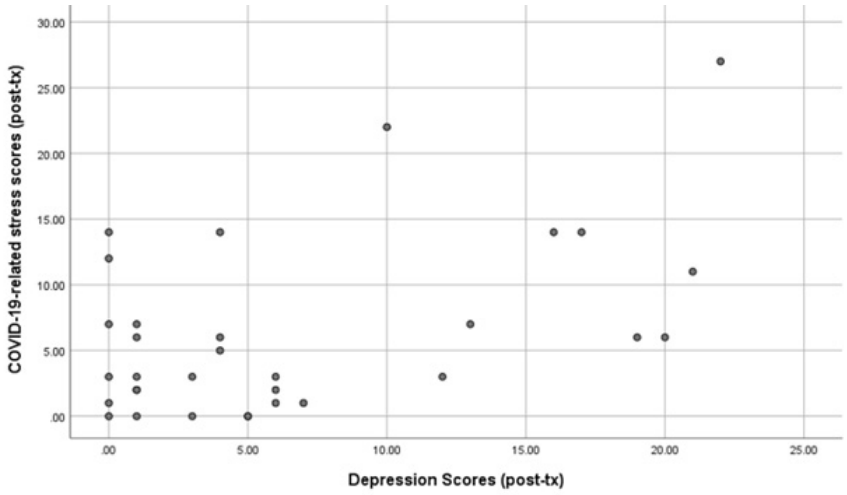

Figure 5d

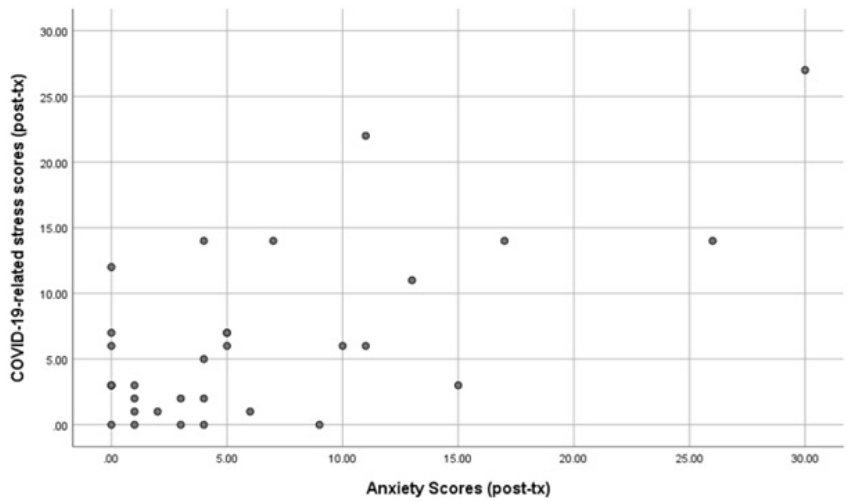

Figure 5e

Mixed Analyses

A mixed ANOVA was used to examine differences among those who adhered to the treatment as instructed and those who did not.

Regarding anxiety, there was a main effect of time of treatment ($F_{(1,31)}$=8.452, p=0.007), such that participants' anxiety scores improved from pre-treatment (m=9.78, SE=1.85) to post-treatment (m=5.67, SE=1.40) regardless of adherence. No main effect was found on adherence ($F_{(1,31)}$=0.268, n.s.), and no interaction was found between time of treatment and adherence ($F_{(1,31)}$=2.068, n.s.).

Regarding depression, there was a main effect of time of treatment ($F_{(1,31)}$=10.427, p=0.003), such that participants' depression scores improved from pre-treatment (m=11.67, SE=1.85) to post-treatment (m=6.60, SE=1.58) regardless of adherence. No main effect was found on adherence ($F_{(1,31}$)=0.213, n.s.), and no interaction was found between time of treatment and adherence ($F_{(1,31)}$=0.002, n.s.).

Regarding sleep quality, sphericity was not met, and no correction was applied. There was a main effect of time of treatment ($F_{(2,56)}$=4.310, p=0.02). Pairwise comparisons indicate that sleep quality improved significantly from pre-treatment (m=18.08, SE=2.02) to mid-treatment (m=13.14, SE=1.81) and post-treatment (m=11.76, SE=1.404). The difference between mid- and post-treatment was not significant. There was no main effect on adherence ($F_{(1,28)}$=2.479, n.s.), and no interaction between adherence and time of treatment ($F_{(2,56)}$=.905, n.s.).

Regarding quality of life, this variable was measured in three separated domains: perceptions of physical health, perceptions of psychological health, and perceptions of social relationships. Additionally, two independently scored items measured the patient's overall perception of their quality of life and the patient's overall perception of their health. The data were submitted to a mixed-measures ANOVA.

Regarding the domain of physical health, there was a main effect on time of treatment ($F_{(1,31)}$=4.78, p=0.036), such that patients reported an improvement in the perception of their health from pre-treatment (m=26.799, SE=0.88) to post-treatment (m=29.00, SE=0.833). No main effect was found on adherence ($F_{(1,31)}$=0.432, n.s.), and no interaction between time of treatment and adherence was found ($F_{(1,31)}$=0.827, n.s.).

Regarding the domain of psychological health, no main effect was found on time of treatment ($F_{(1,30)}$=1.12, n.s.) or adherence ($F_{(1,30}$)=0.221, n.s.), and no significant interaction between time of treatment and adherence ($F_{(1,30)}$=0.768, n.s.) was found.

Regarding the domain of social relationships, no main effect was found on time of treatment ($F_{(1,31)}$=1.40, n.s.) or adherence ($F_{(1,31)}$=1.51, n.s.), and no significant interaction between time of treatment and adherence ($F_{(1,31)}$=0.003, n.s.) was found.

Regarding overall quality of life, no main effect was found on time of treatment ($F_{(1,31)}$=0.39, n.s.) or adherence ($F_{(1,31)}$=0.123, n.s.), and no significant interaction between time of treatment and adherence ($F_{(1,31)}$=0.039, n.s.) was found.

Regarding overall physical health, no main effect was found on time of treatment ($F_{(1,30)}$=0.100, n.s.) or adherence ($F_{(1,30)}$=3.81, n.s.), and no significant interaction between time of treatment and adherence ($F_{(1,30)}$=1.56, n.s.) was found.

Regarding COVID-related stress, there was a main effect on adherence ($F_{(1,29)}$=6.46, p=0.017), such that those who adhered to treatment as instructed reported less COVID-related stress (m=5.29, SE=1.10) than those who did not adhere to treatment as instructed (m=11.21, SE=2.05). No main effect on time of treatment was found ($F_{(1,29)}$=1.26, n.s.), and no significant interaction was found between time of treatment and adherence ($F_{(1,29)}$=0.008, n.s.).

Discussion

In this study, a quasi-experimental design was used to examine the effects of Bio-tuning-a stress reduction tool-on the stress levels of a small sample of participants. The researcher analyzed pre- and post- treatment self-reports of participants' symptoms of anxiety, depression, quality of life, sleep quality, and COVID-related stress. As hypothesized, participants reported an improvement in several of the evaluated factors. Indeed, participants reported an improvement in symptoms of depression and anxiety. Additionally, participants reported a significant improvement in their quality of life, specifically in the domains of perceived physical and psychological health status, as well as an improvement in sleep quality. Participants did not report a significant change regarding stress related to the COVID-19 pandemic. These findings are consistent with previous studies that showed an improvement in sleep quality after exposure to Brainwave Entrainment (BWE) treatment. This study is also consistent with previous studies showing an improvement in psychiatric and physical health symptoms after undergoing BWE. The present findings show that BWE, and more specifically Bio-tuning, may be effective as a tool for stress reduction. A note on the characteristics of the sample used in this study. Significant outcomes were found when correlating pre-treatment expectation responses. Notably, expectations of Bio-tuning's efficacy were positively correlated with measures of COVID-related stress, depression, and anxiety at post-treatment. Results like these demonstrate the complexity of working with human beings. These results present a portrait of real people with real problems, looking for real solutions, who were open to trying a different approach in the hope that it would work. As happens far too often, many of these different approaches fail to work. Notably, results from this study have shown Bio-tuning treatment's clinical potential. Indeed, although the data suggest that an efficacy expectation existed, and thus the possibility of a placebo effect, they also speak to effortful honesty in responding. Indeed, the correlation between expectation and symptoms suggests that while patients were reportedly experienc-

ing symptoms of distress, they were just as forthcoming about their expectations that Bio-tuning would work. Such an outcome is notable as it speaks to the validity of the study's results.

Another notable finding was related to the sleep quality trends that were revealed after conducting the repeated measures ANOVA. As mentioned, significant improvements occurred between Day 1 and Day 11. The Center for Neuro acoustic Research previously reported 21 days as the time needed for improvements to occur; however, the current study shows improvement occurring much earlier. While sleep quality showed significant improvement from Day 1 to Day 21, repeated measures analysis showed that most of the benefit occurred between Day 1 and Day 11. Significant correlations were also found among sleep quality at mid-treatment and COVID-related stress, sleep quality, depression and anxiety at post-treatment. Moreover, improved sleep quality at post-treatment was positively correlated with improved perceptions of one's physical and psychological health. Thus, while these data may reinforce an old adage-that with better sleep comes a better outlook on life-it also suggests that sleep may be a mediating variable regarding our outcome variables, and as such may be responsible for their improved outcomes. Future studies may consider evaluating Bio-tuning's efficacy at a different brainwave range (e.g., alpha) to obtain a better understanding of Bio-tuning's effect on reported changes in quality of life and symptoms of depression and anxiety. Although insignificant, data regarding COVID-related stress also yielded notable findings. As previously reported, no significant improvement was found in this variable between pre-and post-treatment. However, this variable showed a trend of improvement in relation to several other variables regardless of the group differences described below. As this study was carried out from September 2021 through February 2022, it is possible that this variable may have been found to be more significant if all participants were measured during the same 3-week period. As new variants emerged and more people became vaccinated, the degree of COVID-related concern often shifted sometimes from one week to the next; this occurrence likely confounded results on this variable. Future studies may wish to examine participants with a smaller timeline, and if possible, within the same timeline (i.e., within the same 3-week period).

As mentioned, a significant difference was found with regard to adherence to the COVID-19 stress variable. This means that those who adhered to treatment reported less COVID-related stress at both pre- and post-treatment. This suggests a fundamental difference in group disposition. Those who adhered to treatment may have felt more confident in their ability to follow through with the treatment and, as such, reported less concern over becoming ill; those who did not adhere reported concern as they may have expected to not follow through with treatment, and thus not receive the health benefits. However, if this explanation were true, group differences may also have been revealed in other variables, such as anxiety. An alternative explanation may be that those in the non-adherence group were specifically concerned about the COVID-19 virus, and those concerns may have interfered with their treatment adherence. This suggests that those with specific concerns may have difficulty adhering to the treatment, and thus, accountability measures may be required. Future studies may wish to incorporate accountability measures to support treatment adherence. If found to be helpful, accountability measures may also be considered as part of the Bio-tuning procedure. Despite differences in group adherence, Bio-tuning was shown to correlate positively with other outcome variables that improved. As such, although only a percentage (21%) of the group did not adhere to treatment as instructed, analysis reveals a trend in the direction of improvement. As the largest number of days reported for non-adherence was four (i.e., patient adhered for 17 of 21 days), this suggests that if a patient adheres at least a "majority" of the time, improvements may be felt. Indeed, this is in accordance with data from sleep quality analysis, which showed that patients experienced significant improvements by mid-treatment. Finally, a note regarding the significant effects of time of treatment (i.e. differences between pre- and post-treatment) on the variables of perceived physical health, sleep quality, depression, and anxiety. As shown, these variables were found to have significantly improved from pre- to post-treatment, regardless of adherence to treatment as instructed. As mentioned, while not all participants adhered to the 21-day treatment as instructed, those who did not adhere reported using the treatment at least to a point beyond mid-treatment. Although significant findings were revealed in post-treatment perceived physical health, sleep quality, depression, and anxiety, future studies may consider implementing a mid-treatment questionnaire for these variables to track significant changes and inform the Bio-tuning procedure. A few limitations exist in the interpretation of our results. We initially set out to obtain a sample of 34 participants. This was based on a power analysis with a power statistic of 0.81. While we were fortunate enough to have most participants provide responses for each variable, two of the six significant results lacked full participation. Indeed, the quality-of-life variable pertaining to perceived psychological health lacked one participant among that group. Additionally, three responses to the mid-treatment sleep quality questionnaire were not returned, however, all pre-and post-treatment responses were indeed returned. As such, those results should be interpreted with a degree of caution. Additionally, as noted earlier, not all participants adhered to the treatment as instructed. Reasons provided were that they simply "forgot," or that life events interfered with their treatment adherence (e.g., Christmas/New Year's holiday, etc). However, all participants reported eventually returning to their prescribed treatment recommendation and completing the treatment.

An inherent limitation exists within the sample characteristics. At least half of the sample resides within the city of the clinician's office whose data was used in this study. Moreover, all were clients of this clinician's private psychotherapy practice. In addition, more than 80% of the sample reported (at pre-treatment) that they felt Bio-tuning would help them achieve their reason for seeking treatment at least "some." This speaks to the possibility of placebo in patient responding. The author acknowledges the possibility of

placebo effect and is aware of its potential to skew outcome scores. Unfortunately, this was a challenge that was difficult to overcome as this work was part of a dissertation study, and access to time and resources was limited. Thus, a more sophisticated study (i.e., one with a control group) was simply not feasible. As such, to reduce the likelihood of placebo responding, future studies may wish to incorporate a control group into their study. Additionally, as respondents were direct patients of the clinician collecting the data, response bias may also serve as an additional limitation in their responding. It is possible that respondents may have responded in an effort to "help" their clinician by answering in a way that would show the treatment as efficacious. For instance, responding to a depression questionnaire as "severe" at pre-treatment and "minimal" at post treatment, regardless of treatment effect. Although extreme, such a scenario is fathomable given the nature of the therapeutic relationship which often involves a supportive connection between the client and therapist. However, as mentioned previously, patients appeared to be responding honestly given the nature of the results on efficacy expectation as well as the lack of significance in the COVID-19 stress scores. Such scores suggest domain-specific and intentional responding. Instead of presenting a picture of themselves as generally "well" or "stressed," the variance in the outcomes suggests respondents were carefully considering their answers to the items. However, to reduce the possibility of response bias, future studies may consider obtaining participants with no relation to the Bio-tuning practitioner. For this study, attempts to minimize placebo and response bias were done by encouraging the patients to respond as honestly as possible to the questionnaires and informing them that their responses would have no impact on their relationship with the therapist. As noted earlier, almost all study participants reported their permanent residence as somewhere within Los Angeles County. Additionally, a large majority reported their ethnicity as Hispanic. Thus, conducting a similar study on a sample of a different ethnicity and geographic location will further inform the current data. Finally, our study only captured reported improvement instead of actual entrainment (i.e., lasting psychological effects). Thus, future studies may consider assessing patients beyond 21 days to determine whether entrainment (i.e., lasting effects) has occurred, as well as the duration of its occurrence.

Regardless of its limitations, this study has provided meaningful results. It has shown through a small clinical sample that participants are responding to the treatment favorably. An optional question on the post-treatment questionnaire asked participants for feedback about their experience. Some of the feedback received is as follows: "an immediate relief from my overthinking tendencies"; "I feel much better during the day, more relaxed when I go to sleep"; "within a week, I found myself feeling more balanced and less irritated by the 'noise' around me"; "I get better sleep, more restful and relaxed, don't feel tired when I wake up." Moreover, others reported continued use of the Bio-tuning audio file beyond the prescribed 21 days as they claim to enjoy the benefits that come with listening to their audio file. Indeed, these direct statements suggest that something more may be occurring beyond the simple enjoyment of listening to a relaxing audio file. Although there is still much to learn regarding the processes involved in a tool like Bio-tuning, or BWE in general, results like these should inspire others to examine the area further.

Clinical Implications

The results can be used to promote the wider use of Bio-tuning in a clinical setting. Prior to the results found in this study, reported benefits of the Bio-tuning equipment were based off internal studies conducted by the Center for Neuroacoustic Research. This study is the first of its kind to be conducted outside of any internal ties to the company that could be misconstrued as a conflict of interest. These results provide an objective evaluation of the Bio-tuning protocol that supports its implementation across a wide range of clinical environments. Finally, results from this study demonstrate that Bio-tuning may yield a more immediate effect than previously expected. As mentioned, results from the sleep quality questionnaire show that participants experienced significant improvements in their sleep quality between Day 1 to Day 11. As such, Bio-tuning practitioners may consider reducing their 21-day use recommendation so that it is aligned with the results of this study. Further, Bio-tuning may also be considered for evaluation as a tool that supports the immediate improvement in sleep quality. Future studies may consider tracking depression and anxiety symptoms at mid-treatment to determine the point at which the treatment is providing its most support. Indeed, if the improvement is immediate, Bio-tuning may also be considered for the treatment of severe psychiatric experiences, such as a major depressive episode.

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None.

Conflict of Interest

None of the authors have a conflict of interest.

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