

# **Investigation of an Ultra-Brief Breathing Technique for the Treatment of Physiological and Psychological Markers of Anxiety**

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## Abstract

Relaxation therapies are an attractive intervention for decreasing anxiety-related symptoms as they can be self-administered with minimal training, support or financial cost. Self-regulation of breathing is a simple and accessible intervention with minimal empirical evidence to support long-term effects of a consistent practice. The ultra-brief 4-7-8 breathing technique is one method that has substantial anecdotal support and notoriety. Despite the growing popularity of brief breathing techniques, there have been few reported empirical investigations on the effects of the long-term practice of these ultra-brief interventions. This empirical investigation will serve as a proof of principle for if there is utility in regularly practicing ultra-brief breathing interventions for individuals experiencing ongoing stress and anxiety. University students (n=109) reporting mild-to-severe symptoms of trait anxiety were randomized into three groups: a waitlist control group (n=23), a group instructed to perform an ultra-brief breathing technique twice a day (n=46), and a group instructed to perform an ultra-brief counting technique twice a day (n=41). Self-reported trait anxiety was measured at three time points over eight weeks. Repeated Measures ANOVA's indicated that participants who performed the breathing technique a minimum of four times a week reported significantly decreased trait anxiety after eight weeks, but not after four weeks, compared to participants performing a counting exercise of identical duration. Differences were specific to trait cognitive anxiety, not trait somatic anxiety. There was a significant relationship between treatment adherence and treatment efficacy for individuals who performed the breathing technique, but not for the counting technique. There were no differences in resting-state physiological markers of stress and anxiety after four weeks. Overall, these findings support the regular daily practice of the 4-7-8 breathing technique as a clinically efficacious intervention for anxiety. Further dismantling studies are required to identify and apply the active ingredients of the 4-7-8 breathing technique.

**Keywords:** breathing, relaxation, anxiety, respiration, parasympathetic, stress

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# Table of Contents

Declaration of Committee .....	ii
Ethics Statement .....	iii
Abstract .....	iv
Acknowledgements .....	v
Table of Contents .....	vi
List of Tables .....	ix
List of Figures .....	x
List of Acronyms .....	xi
<b>Chapter 1. INTRODUCTION.....</b>	<b>1</b>
1.1. Parasympathetic Nervous System .....	2
1.2. Vagal Tone .....	2
1.3. Relaxation Training .....	3
1.4. HRVB .....	4
1.5. Acute Effects .....	5
1.6. Adherence .....	5
1.7. Brief Interventions .....	7
1.7.1. Respiration Parameters .....	7
1.8. Summary .....	9
1.9. Hypotheses .....	10
1.9.1. Aim 1: efficacy of the breathing intervention in decreasing markers of anxiety .....	10
1.9.1.1. Hypotheses of anxiety markers in 3 groups over 4 weeks .....	10
1.9.1.2. Hypotheses of trait anxiety in 2 groups over 8 weeks .....	11
<b>Chapter 2. METHODS .....</b>	<b>14</b>
2.1. Study Participants .....	14
2.2. Materials .....	14
2.3. Experimental Protocol .....	16
2.3.1. Covid-19 Pandemic Resulted in Two Cohorts .....	16
2.3.2. In-Person, Pre-COVID-19 Pandemic Cohort (n = 60) .....	16
2.3.3. Online, During COVID-19 Pandemic Cohort (n = 44) .....	19
2.4. Analytical Procedures .....	20
2.4.1. Repeated Measures Mixed ANOVA .....	21
2.4.2. Nonparametric Analyses .....	22
2.4.3. Pearson's Correlation .....	22
2.4.4. Sample Size .....	22
<b>Chapter 3. RESULTS .....</b>	<b>24</b>
3.1. Descriptive Statistics .....	25
3.1.1. In-Person, Pre-Covid-19 Cohort .....	25
3.1.2. Online, During Covid-19 Cohort .....	26
3.1.3. Online and In-Person Cohort Comparison .....	26

3.1.4.	Experimental Groups .....	32
3.2.	Primary Analyses.....	35
3.2.1.	Aim 1: Efficacy of the Ultra-brief Breathing Intervention.....	35
3.2.1.1.	Mean change in trait anxiety between Breathing, Counting, and Waitlist Control group after 4 weeks .....	35
3.2.1.2.	Mean change in physiological resting state measures between the Breathing, Counting, and Waitlist Control group after 4 weeks. ....	36
3.2.1.3.	Mean change in trait anxiety between the Breathing and Counting group after 8 weeks.....	40
3.2.1.4.	Mean change in trait anxiety in the Waitlist Control group over 8 weeks.....	41
3.2.1.5.	Trait anxiety over 4 weeks in a subgroup of clinical severity.....	43
3.2.2.	Aim 2: Acceptability of the Ultra-brief Interventions.....	43
3.2.3.	Aim 3: Relationship of Adherence to Practice on Treatment Efficacy.....	45
3.2.3.1.	Mobile Application Tracked Technique Frequency.....	45
3.2.3.2.	Self-reported Technique Frequency.....	46
3.2.3.3.	Correlation between Treatment Adherence and Treatment Efficacy.....	47
3.3.	Supplementary Analysis .....	49
3.3.1.	Adequate Adherence Subgroup.....	49
3.3.1.1.	AA Mean trait anxiety after four weeks between Breathing, Counting, Waitlist Control groups .....	50
3.3.1.2.	AA: Physiological changes after 4 weeks between Breathing, Counting, Waitlist Control group.....	51
3.3.1.3.	AA: Trait anxiety after 8 weeks between the Breathing and Counting groups.....	52
3.3.1.4.	AA: Trait anxiety within the Waitlist Control group over 8 weeks .....	55
3.3.1.5.	AA: Trait Anxiety after 4 weeks in a subgroup of clinically High Anxiety .....	56
3.3.2.	Female Only Subgroup.....	57
3.3.2.1.	Female Only: Mean change in trait anxiety between Breathing, Counting and Waitlist Control group after 4 weeks .....	57
3.3.2.2.	Female Only: Mean change in trait anxiety between Breathing and Counting group after 8 weeks.....	60
<b>4.</b>	<b>DISCUSSION .....</b>	<b>61</b>
4.1.	Summary of Major Findings.....	61
4.1.1.	Four Week Follow-Up: Baseline to Week 4 .....	62
4.1.2.	Eight Week Follow-Up: Baseline to Week 8 .....	63
4.1.3.	Waitlist Control Group: Baseline to Week 8.....	65
4.1.4.	Aim 2: Treatment Acceptance .....	66
4.2.	Strengths and Limitations .....	68
4.3.	Future Research.....	70
4.4.	Conclusions.....	71
	<b>REFERENCES.....</b>	<b>73</b>
	<b>APPENDICES.....</b>	<b>83</b>
	Appendix A: Demographic Questionnaire .....	83

Appendix B: STICSA Trait .....	84
Appendix C: PANAS .....	85
Appendix D: Technique Acceptability.....	88
Appendix E: Study Completion .....	89
Appendix F: Analyses of Study Dropouts.....	90
Appendix G Statistical Output for Section 3.2.1.1: Is the mean change in trait anxiety after four weeks different between the Breathing, Counting, or Waitlist Control groups? .....	91
Appendix H Statistical Output for Section 3.2.1.2: Is the mean change in physiological resting state measures after four weeks different between the Breathing, Counting, or Waitlist Control groups? .....	92
Appendix I Statistical Output for Section 3.2.1.3: Is the mean change in trait anxiety after eight weeks different between the Breathing and Counting groups? .....	94
Appendix J Statistical Output for Section 3.2.1.4: Is the mean change in trait anxiety in the Waitlist Control group different in the first four weeks of the study, compared to the second four weeks once they begin the Breathing technique? .....	97
Appendix K Statistical Output for Section 3.2.1.5: Is there a significant change in trait anxiety after four weeks for a subgroup of clinical severity? .....	98
Appendix L Treatment Acceptance Questions by Group .....	101
Appendix M Statistical Output for Adequate Adherence Subgroup Section 3.3.1.1: .....	102
Appendix N Statistical Output for Adequate Adherence Subgroup Section 3.3.1.2: .....	104
Appendix O Statistical Output for Adequate Adherence Subgroup Section 3.3.1.3 .....	106
Appendix P Statistical Output for Adequate Adherence Subgroup Section 3.3.1.4 .....	109
Appendix Q Statistical Output for Adequate Adherence Subgroup Section 3.3.1.5 .....	111
Appendix R Statistical Output for Female-Only Subgroup Comparison 1: 3 Groups over 4 Weeks .....	114
Appendix S. Statistical Output for Female-Only Subgroup: Physiological Measures over 4 weeks.....	117
Appendix T Statistical Output for Female-Only Subgroup Comparison 3: 2 Groups over 8 Weeks .....	119



## List of Tables

Table 1	Power Calculations for 3 Groups over 2 Time Points .....	23
Table 2	Power Calculations for 2 Group by 3 Time Point Comparisons .....	23
Table 3	Descriptive Statistics for the In-Person, Pre-Covid-19 Study Cohort.....	28
Table 4	Descriptive Statistics for the Online, During-Covid-19 Study Cohort.....	30
Table 5	Descriptive Statistics Comparing the In-Person, Pre-COVID-19 cohort (n=60), and the Online, During COVID-19 cohort (n = 44).....	31
Table 6	Independent Samples T-tests comparing the In-Person, Pre-COVID-19 cohort to the Online, During COVID-19 cohort .....	32
Table 7	Descriptive Statistics for Demographic and Clinical Variables of the Total Sample (n=104) .....	33
Table 8	Pairwise Comparisons of Time Points Within the Waitlist Control Group	42
Table 9	Treatment Acceptability Mann-Whitney U Ranking Statistics of Group Differences.....	44
Table 10	Post Treatment Satisfaction Independent Samples t-test .....	45
Table 13	Objective App Usage at 4 Weeks Mann Whitney U Rankings .....	46
Table 11	Self-Reported App Frequency of Use by Group .....	46
Table 12	Treatment Adherence Wilcoxon Matched-Pairs Signed Rank Test.....	47
Table 14	Descriptive Statistics of AA Subgroup .....	49
Table 15	AA Subgroup: Pairwise Comparisons within WLC by Time Point .....	55
Table 16	AA Subgroup: Cognitive Anxiety Pairwise Comparisons within WLC.....	55
Table 17	AA Subgroup: Somatic Anxiety Pairwise Comparisons within WLC.....	56

## List of Figures

Figure 1.	Sample Selection Flow Chart .....	25
Figure 2	Estimated Marginal Means of a Repeated Measures ANOVA for Log Transformed STICSA Total Score, 3 Groups over 2 Time Points .....	36
Figure 3	Estimated Marginal Means of a Repeated Measures ANOVA for Systolic Blood Pressure .....	37
Figure 4	RMSSD Outcomes at Baseline and Week 4 Timepoints .....	39
Figure 5	Estimated Marginal Means of a Repeated Measures ANOVA for Log Transformed SDNN Measure of Heart Rate Variability .....	40
Figure 6	Estimated Marginal Means of a Repeated Measures ANOVA for Log Transformed STICSA Total .....	41
Figure 7	Estimated Marginal Means of a Repeated Measures ANOVA for Log Transformed STICSA Total within the Waitlist Control Group.....	42
Figure 8	Change in Total Anxiety relative to Self-Report App Usage at Week 8...	48
Figure 9	AA Subgroup ln(Total Anxiety), 2 Groups over 8 Weeks .....	53
Figure 10	AA Subgroup ln(Cognitive Anxiety) 2 groups over 8 weeks.....	54
Figure 11	AA Subgroup ln(Somatic Anxiety) 2 groups over 8 weeks.....	54

## List of Acronyms

ANOVA	Analysis of Variance
CBT	Cognitive Behavioral Therapy
PSNS	Parasympathetic Nervous System
HRV	Heart Rate Variability
HRVB	Heart Rate Variability Biofeedback
RMSSD	Root Mean Square of the Successive Differences
SDNN	Standard Deviation of Normal to Normal R-R Intervals
SBP	Systolic Blood Pressure
DBP	Diastolic Blood Pressure
STICSA	State-Trait Inventory for Cognitive and Somatic Anxiety
PANAS	Positive Affect Negative Affect Schedule
WLC	Waitlist Control
AA	Adequate Adherence

## Chapter 1. INTRODUCTION

In Canada, anxiety disorders have been estimated to affect 12% of the population, causing mild to severe impairment across various outcomes (Health Canada, 2002). Anxiety disorders can severely impair an individual's well-being and general functioning, as the disorders span behavioral, emotional, physiological, and cognitive states.

Anxiety-related disorders are treatable, both by pharmacological and non-pharmacological means. In 2014, the Survey on Living with Chronic Diseases in Canada (SLCDC) found that 70% of individuals with an anxiety or mood disorder reported currently taking prescription medications (Toews et al., 2016). However, while anxiety medications are effective at suppressing symptoms, many come with the burden of highly undesirable side effects, including high rates of dependency, drowsiness, impaired cognition, and sexual dysfunction (Buffett and Stewart, 2002; Struzik et al., 2004; Manzoni et al., 2008). Additionally, psychotropic medications are also costly financially (Huskamp and Shinogle, 2005).

Non-pharmacological interventions for anxiety-related disorders have been shown to have comparable efficacy to medications, including cognitive behavioral therapy (CBT) and relaxation techniques (APA, 2009; Katzman et al., 2014). While CBT is the gold-standard evidence-based therapy for treating anxiety-related disorders, practical barriers can prevent individuals from receiving these services, including: a shortage of trained professionals, long waitlists, expensive fees, extensive time commitments, and living in non-urban locations (Christensen and Hickie, 2010). Relaxation techniques offer an alternative as they can be self-administered with minimal training or support. With an absence of side effects and relative ease of administration, relaxation techniques are an effective, low cost alternative intervention for the treatment of anxiety related disorders. Relaxation training has been shown to be a reliable intervention for the treatment of anxiety, with medium-to-large effect sizes (Manzoni et al., 2008; meta-analysis).

## 1.1. Parasympathetic Nervous System

It is posited that the therapeutic benefits of relaxation and the relaxation response may be related to activation of the parasympathetic nervous system (PSNS; Jacobs, 2001; Jindal et al., 2013). Activation of the PSNS yields similar physiological characteristics as seen in the relaxation response, including decreased heart rate, decreased blood pressure, and decreased oxygen metabolism (Jindal et al., 2013; Jerath et al., 2006). If regular parasympathetic activation is a key ingredient of the efficacy of relaxation therapies, a faster and more convenient intervention targeting the PSNS may hold great promise as an accessible treatment of anxiety related symptoms.

Self-regulation of breathing has been suggested as a possible primary treatment for anxiety as it can promote an autonomic nervous system shift from a sympathetic dominant state (i.e., “fight or flight”) to a parasympathetic dominant state (rest and digest; Jerath et al., 2015). Slow paced breathing is suggested to increase the activation of the vagus nerve (Gerritsen & Band, 2018)—the main nerve of the parasympathetic nervous system (Brodal, 2016)—which is hypothesized to underlie many of the positive therapeutic outcomes associated with emotion regulation, relaxation, cognition, and well-being (Gerritsen & Band, 2018). A study investigated the effect of slow, deep breathing on the action of hyoscine-N-butylbromide (Buscopan), a parasympathetic blocker drug (Pramanik et al., 2009). In a group where the drug was not administered, five minutes of slow deep breathing resulted in decreases in blood pressure and heart rate, whereas following the administration of the drug there were no significant changes in blood pressure or heart rate. This finding suggests that the practice of slow breathing is able to modulate the autonomic nervous system through parasympathetic activity.

## 1.2. Vagal Tone

PSNS activation is not directly measured, but instead inferred by measuring processes that it effects – including heart rate and heart rate variability (HRV). While heart rate is simply a measure of frequency of heart beats in a given time period, HRV represents a more subtle measurement of the change in the time interval between successive heartbeats. It has been shown that HRV represents a more direct index of the parasympathetic nervous system than simply heart rate alone (Malik, 1996). Physiologically, the time interval between heartbeats is under constant variation,

sensitive to a variety of inputs, including movement, respiration rate, digestive processes, caffeinated beverages, and stress, among many others (Laborde et al., 2017). These continuous beat-to-beat changes largely reflect the action of the vagus nerve, the main nerve of the PSNS. Stimulation of the vagus nerve results in the release of acetylcholine at the pacemaker cells of the heart, momentarily decreasing heart rate and increasing HRV (Jalife et al., 1983). The term 'vagal tone' is also frequently used to refer to the level of PSNS activation, with high vagal tone referring to increased PSNS activation.

Psychopathologically, low vagal tone has been linked to a number of stress-related psychological conditions. Several authors have reported relationships between lower vagal tone with state and trait forms of anxiety (Miu et al., 2009; Friedman, 2007; Fuller, 1992; Watkins et al., 1998). High anxiety, but not depression, has been associated with reduced vagal tone in patients following myocardial infarction (Watkins et al., 2002). Adolescents with anxiety and antisocial behavior have been reported to have lower vagal tone (Mezzacappa et al., 1997).

However the PSNS does not solely innervate the heart. Additionally, PSNS activation stimulates digestive processes, relaxes muscles, and allows blood flow to reach reproductive organs. Additionally, higher vagal tone has been theorized to cause a variety of cognitive and psychological benefits in several models. The neurovisceral integration model posits that higher vagal tone will lead to better executive cognitive performance and emotional regulation (Thayer et al., 2009). The polyvagal theory alludes to an association with social interaction and emotion, as vagal fibers are neuroanatomically linked to the facial muscles utilized for emotional expression (Porges, 2007). And in the biological-behavioral model, higher vagal tone is seen as a reflection of functional energy which the organism is able to utilize in states of high physical activity (Grossman and Taylor, 2007). Slowed, paced breathing has been cited as a common way to achieve higher vagal tone (Lehrer, 2013; Childre, 2010).

### **1.3. Relaxation Training**

Relaxation training includes a variety of techniques which emphasize developing a relaxation response to counteract the systemic impairments of stress (Manzoni et al., 2008). The theoretical construct of a specific 'state' of relaxation was first suggested as a

protective mechanism by Walter Hess in a series of experiments in the 1930s and 1940s (Benson and Klipper, 1992). In these experiments, Hess discovered that stimulation of certain brain regions in laboratory animals would produce an effect of relaxed muscles, decreased blood pressure, and decreased breathing rate. Herbert Benson built upon this concept by popularizing the idea of the 'relaxation response' (Benson, 1975). Benson defined the relaxation response as an achievable state of decreased levels of oxygen consumption, heart rate, breathing rate, and muscle tension. Benson recognized that many techniques activated the relaxation response, including progressive muscle relaxation, autogenic training, controlled breathing techniques, guided imagery, qi gong, yoga, and various forms of meditation (Benson and Klipper, 1992). Several techniques specifically utilize breathing as the intervention's locus of control, however research of these techniques commonly suffers from poor methodology, including the lack of control groups, no group randomization, and nonspecific presentation of methods, (Brandani et al., 2017).

#### **1.4. HRVB**

One breathing-based intervention that has been applauded for its methodological rigor is heart rate variability biofeedback (HRVB; Schoenberg and David, 2014). The goal of HRVB is to maximize the amplitude of HRV oscillations. A HRVB session typically lasts for 25 minutes, but no less than 10 minutes. Throughout the session, an individual receives immediate biofeedback of an index of their HRV. The goal for the session is to keep HRV as high as possible, working towards the maximum amplitude fluctuation. The frequency that correlate with the personal maximum HRV has been called the 'resonance frequency breathing rate.' While the exact breathing frequency varies from person to person, it is reported to commonly lie between 4.5 and 7 breaths per minute (Vaschillo et al., 2002; Lehrer and Woolfolk., 2007).

A systematic review of the efficacy of HRVB on various psychiatric disorders concluded HRVB improved clinical symptoms in 70% of the studies reviewed (Schoenberg and David, 2014). The review clearly identified the high quality of the HRVB study methodologies, including a more recent meta-analysis of 24 studies of HRVB which concluded that the intervention is an effective treatment for stress and anxiety (Goessl et al., 2017).

## **1.5. Acute Effects**

Slow, paced breathing has been found to have an immediate effect on the cardio-respiratory system, influencing several parameters of HRV and blood pressure fluctuations (Edmonds et al., 2009; Park and Park, 2012; Lin et al., 2014; Van Diest et al., 2014). Heart rate increases during inspiration while arterial blood pressure decreases, and vice versa during expiration (Billman, 2011). These relationships between heart rate, blood pressure and respiration are known as cardiorespiratory coupling (Dick et al., 2014).

The physiological effects of slow breathing are restricted to the time the techniques are being performed (Russo et al., 2017). There have been few reliable findings reported immediately after a breathing-based session (Zaccaro et al., 2018). You et al., (2021) reported an experiment where slow paced breathing for a duration of five minutes resulted in increased HRV during the intervention, but had no effect on HRV indices once the slow-paced breathing was discontinued. These findings signify the transient nature of an acutely induced change. Balban et al., (2023) investigated three five-minute breathwork interventions performed for 30 days. Immediately after performing the breathing practice, participants reported significantly increased positive affect, significantly decreased negative affect, and decreased state anxiety.

## **1.6. Adherence**

Relaxation techniques generally suffer the practical problem of self-motivation, as individuals need to sustain a daily routine for therapeutic benefit (Hillenberg and Collins, 1983, Lehrer and Woolfolk 2007). Depending on the technique, relaxation techniques require up to an hour of practice a day, typically in a quiet time and space without interruptions. These requirements of location and an individual's time may be practical barriers that prevent individuals from developing a consistent daily routine.

Increased adherence to at-home practice has been shown to be associated with improved outcomes, regardless of the specific intervention studied (Kazantzis and Deane, 1999, Scheel et al., 2004). In a direct comparison of physical activity, mindfulness, and HRVB, it was reported that regardless of the specific treatment type, increased compliance to the intervention was the greatest factor in achieving stress



reduction (Van der zwan et al., 2015). In an online applied relaxation intervention, the treatment effect was moderated by how many relaxation sessions were completed, while there was no effect of type of relaxation program that was followed (Alfonsson et al., 2015). If treatment adherence is the most influential factor in determining the efficacy of relaxation interventions, then techniques with low barriers to accessibility and compliance should be prioritized as treatment interventions.

Unfortunately, obtaining high compliance rates continues to be a major obstacle for relaxation-based interventions. In a study of guided relaxation therapy for hypertension treatment, only 32% of the participants were found to average the desired once-daily, 15-minute practice over the course of 10 weeks (Hoelscher et al., 1986). A similar methodology in a sample with anxiety related difficulties reported even less compliance to the same technique over four weeks, with only 25% of participants completing the prescribed frequency of practice (Hoelscher et al., 1984). In a sample of pregnant women, participants were instructed to practice a 13-minute guided relaxation imagery audio program daily for five weeks (Chuang et al., 2015). After five weeks, there was a total adherence rate of 53%. In a qualitative analysis, reasons for nonadherence included busy schedules, unavailability of the audio program device, and a general lack of interest. In a study of a computerized CBT program, 75% of program non-completers reported that the program was too time consuming for them (Hermes et al., 2016), while another CBT study reported that the program was too difficult to fit into the daily lives of the participants (Johansson et al., 2015). Lehrer and Woolfolk (2007), suggest low compliance of stress management therapies may be due to the combined requirements of long time commitments to complete the practice and having a private location to perform the practice. It has been theorized that a strong predictor of adherence is client acceptability for the treatment, referring to the client's attitudinal judgment toward the proposed treatment. Scheel et al. (2004) suggested that a major determinant of client acceptability is the perceived difficulty of the treatment, specifically in the time, effort, and complexity required. Relaxation therapies with reduced barriers of time, effort, and complexity may therefore strengthen client acceptability and adherence to regular practice, thus increasing their efficacy.

## 1.7. Brief Interventions

In response to the logistical deterrents of time and location requirements, several brief relaxation interventions (less than 5 minutes) have been developed for greater accessibility. These so-called “ultra-brief” breathing interventions have become popularized with their ease of administration in any context, including the 3-minute breathing space of Mindfulness Based Stress Reduction (Segal, 2016; Segal et al., 2002), Dr. Andrew Weil’s 4-7-8 breath (Weil, 2000), “box breathing” (Johns, 2012), and diaphragmatic breathing (Varvogli and Darviri, 2011), among others.

When considering the differences between these breathing manipulations, there are many dimensions to consider, including the frequency of the breath (Schipke et al., 1999), the ratio of inspiration and expiration (Strauss-Blasche et al., 2000), the presence and location of a breath hold (post-inspiration or post expiration; Grossman et al., 1983), the volume of the breath inspired (Hirsch and Bishop, 1981), and whether breathing is performed orally, nasally or both.

### 1.7.1. Respiration Parameters

Typical spontaneous breathing frequencies are generally between 12 and 20 breaths per minute (Derrickson & Tortora, 2014). Respiration at slower frequencies – between 4.5 and 7 breaths per minute (Vaschillo et al., 2002; Lehrer and Wollfolk, 2007) – has been identified as an ideal breathing frequency for maximizing heart rate variability in healthy volunteers. While the exact frequency varies from person-to-person, many slow paced breathing experimental conditions are performed around six breaths per minute, as this rate has been linked to the highest vagal response (Lehrer & Gevirtz, 2014; Shaffer & Meehan, 2020).

The ratio of inhale to exhale is a variable that is frequently overlooked in studies of breathing interventions. Van Diest et al. (2014) investigated this question by comparing both respiration rates and inhale-to-exhale ratios amongst healthy volunteers. Acutely, participants self-reported stress reduction after a condition with a short inhale and long exhale (3 seconds : 7 seconds) in comparison to a long inhale and short exhale (7 seconds : 3 seconds). Physiologically, higher vagal tone in athletes was reported at breathing sequences where the exhalation was longer than the

inhalation, compared across a stable respiration frequency of six breaths per minute (Laborde et al., 2021). Other investigations support higher vagal tone (high frequency HRV) in breathing sequences with an extended exhalation (Porges 2007; Strauss-Blasche et al. 2000). An extended exhalation at a low respiration frequency was found to have the largest effect on decreasing pain perception in the presence of a moderately painful stimulus, relative to extended inhalations or at spontaneous respiration rates (Jafari et al., 2020). Other reports have shown that the ratio of inspiration to expiration has no effect on HRV indices (Klintworth et al., 2012), however this finding was reported at a respiratory rate of 13 breaths per minute. This suggests that if the ratio of inspiration to expiration is a relevant breathing parameter, it may only be relevant for slower respiration frequencies.

Respiration-induced blood pressure changes can be further increased by maintaining the inspired air within the lungs, as is the case during post-inspiration breath-holding (Reyes del Paso et al., 2014, Grossman 1983). Post inspiration pauses are known to produce a rapid and pronounced heart rate decrease, up to twenty-beat-per minute decelerations (Daly et al., 1979; Grossman, 1983; Gooden, 1994). This level of bradycardia is not only pronounced, but sudden, as most of the deceleration occurs between a single pair of beats (Angell-James and Daly, 1975; Grossman, 1983). As the breath hold volume increases, the extent of the amplitude of heart rate decrease also increases – suggesting the degree of inhalation preceding the breath-hold is also of importance (Hirsch and Bishop, 1981).

A recent four group randomized controlled trial compared multiple breathing techniques practiced for five minutes a day for 30 days (Balban et al 2023). They compared a “cyclic sighing” technique (inhalation through nose, prolonged exhalation through mouth), “box breathing” (equal durations of inhalation through nose, breath hold, exhalation through nose, breath hold), “hyperventilation with retention” (30 prolonged inhalations through the nose with brief exhalation, followed by a 15 second breath hold), and a mindfulness meditation control (passive observation of spontaneous breathing). All techniques analyzed resulted in an acute increase in self-reported mood after completing the 5-minute practice. The acute effect became stronger with more practice over the 30 days study. The “cyclic” sighing technique, with an emphasis on an extended exhalation, was the strongest effect and significantly stronger than the mindfulness meditation control.

## 1.8. Summary

While relaxation therapies have been shown to be effective in the management of anxiety-related symptoms, the logistical requirements of time and private location may contribute to poor adherence to a consistent self-practice. Self-regulation of breathing can promote a shift to a parasympathetic dominant state in a simple, accessible intervention. Despite the popularity of brief breathing practices, there have been few reported empirical investigations on the effects of the long-term practice of these ultra-brief interventions. As such, there is no evidence to suggest that their increased accessibility does indeed lead to higher adherence rates.

The present research investigates an ultra-brief, one-minute breathing-based intervention to be practiced twice a day for a period of eight weeks in individuals reporting mild to severe anxiety related symptoms. The controlled breathing intervention will consist of a breathing sequence that follows an inhale-retention-exhale ratio of 4-7-8 for a theoretically presumed direct activation of the parasympathetic nervous system (Weil, 2000). This ultra-brief, easily performed breathing technique can be applied without the time and location constraints of typical relaxation therapies. The investigation will serve as a proof of principle by comparison to a control group of participants instructed to engage in a counting exercise for the same amount of time at the same frequency (twice a day for a period of eight weeks). The ultra-brief breathing intervention will be assessed as a proof of principle for addressing three aims: 1) the efficacy of the breathing intervention in decreasing markers of anxiety, 2) the acceptability of the breathing intervention to participants, and 3) the relationship of adherence to practice on treatment efficacy.

In addressing the efficacy of the intervention in Aim 1, a series of repeated measures ANOVA's to analyze self-reported trait anxiety in 3 groups over 4 weeks, resting physiological variables in 3 groups over 4 weeks, self-reported trait anxiety in 2 groups over 8 weeks, self-reported trait anxiety in the waitlist control group over 8 weeks, and self-reported trait anxiety in a "High Anxiety" group over 4 weeks. Aim 2 addressed treatment acceptability of brief interventions by comparing the mean of a scale designed to assess treatment acceptability asked immediately after learning the technique. Aim 3 addressed the relationship between treatment adherence and efficacy by analyzing the correlation between treatment adherence and treatment efficacy in each group.

## 1.9. Hypotheses

The null and alternative hypothesis for each planned analysis is outlined below. The expected results are shown in bold.

### 1.9.1. Aim 1: efficacy of the breathing intervention in decreasing markers of anxiety

Treatment efficacy was addressed with five distinct families of hypotheses, analyzing self-reported trait anxiety in 3 groups over 4 weeks (3.2.1.1), resting physiological variables in 3 groups over 4 weeks (3.2.1.2), self-reported trait anxiety in 2 groups over 8 weeks (3.2.1.3), self-reported trait anxiety in the waitlist control group over 8 weeks (3.2.1.4), and self-reported trait anxiety in a “High Anxiety” group over 4 weeks (3.2.1.5).

#### 1.9.1.1. Hypotheses of anxiety markers in 3 groups over 4 weeks

Three groups of participants were compared: a group who performed an ultra-brief breathing sequence twice a day for four weeks, a group who performed an ultra-brief counting exercise for the same durations and time period, and a waitlist control. Self-reported trait anxiety and objectively measured physiological indexes were compared across groups: root mean square of the successive differences (RMSSD), standard deviation of normal to normal R-R intervals (SDNN), systolic blood pressure (SBP), diastolic blood pressure (DBP). It was hypothesized that:

- $H_{101}$ : There is no difference in the mean change in self-reported trait anxiety over four weeks between the three groups.
- $H_{1a1}$ : **The mean change of self-reported trait anxiety over four weeks is significantly different between one or more groups.**
- $H_{102}$ : There is no difference in the mean change in RMSSD over four weeks between the three groups.
- $H_{1a2}$ : **The mean change of RMSSD over four weeks is significantly different between one or more groups.**

- H1<sub>03</sub>: There is no difference in the mean change in SDNN over four weeks between the three groups.
- **H1<sub>a3</sub>: The mean change of SDNN over four weeks is significantly different between one or more groups.**
- H1<sub>04</sub>: There is no difference in the mean change in SBP over four weeks between the three groups.
- **H1<sub>a4</sub>: The mean change of SBP over four weeks is significantly different between one or more groups.**
- H1<sub>05</sub>: There is no difference in the mean change in DBP over four weeks between the three groups.
- **H1<sub>a5</sub>: The mean change of DBP over four weeks is significantly different between one or more groups.**

### **1.9.1.2. Hypotheses of trait anxiety in 2 groups over 8 weeks**

Two groups of participants were compared: a group who performed an ultra-brief breathing sequence twice a day for eight weeks, and a group who engaged in a counting exercise for the same durations and time period. Self-reported trait anxiety was measured at baseline, Week 4, and Week 8. It was hypothesized that:

- H2<sub>01</sub>: There is no difference in the mean change in self-reported trait anxiety over eight weeks between the two groups.
- **H2<sub>a1</sub>: The mean change of self-reported trait anxiety over eight weeks is significantly different between the groups.**

### **1.9.1.3. Hypotheses of trait anxiety in Waitlist Control group over 8 weeks**

An effect of time was compared over 8 weeks for the Waitlist Control individually. For the first four weeks (Baseline to Week 4) the group was not instructed to engage in any daily technique. For the second four weeks (Week 4 to Week 8), the group performed an ultra-brief breathing sequence twice a day. Self-reported trait anxiety was measured at baseline, Week 4, and Week 8. It was hypothesized that:

- H3<sub>01</sub>: There is no difference in the mean change in self-reported trait anxiety over eight weeks between the time points.
- **H3<sub>a1</sub>: The mean change of self-reported trait anxiety over eight weeks is significantly different between time points.**

#### **1.9.1.4. Hypotheses of effect of Breathing group on trait anxiety in High Anxiety subgroup over 4 weeks**

One group of participants with clinically significant levels of anxiety was formed from the Breathing group with high baseline trait anxiety, and the Waitlist Control group with high trait anxiety at Week 4 (just before they begin to perform the breathing technique). Self-reported trait anxiety before and after four weeks of performing the Breathing technique. It was hypothesized that:

- H4<sub>01</sub>: There is no difference in the mean change in self-reported trait anxiety over eight weeks between the time points.
- **H4<sub>a1</sub>: The mean change of self-reported trait anxiety over eight weeks is significantly different between time points.**

#### **1.9.2. Aim 2: Acceptability of the breathing intervention**

Two groups of participants were compared: a group who performed an ultra-brief breathing sequence twice a day for eight weeks, and a group who engaged in a counting exercise for the same durations and time period. Self-reported acceptability of the experimental techniques were measured immediately after initially learning the technique, and self-reported treatment satisfaction upon conclusion of the study. It was hypothesized that:

- **H5<sub>01</sub>: There is no difference in the mean treatment acceptance after learning the technique.**
- H5<sub>a1</sub>: The mean of treatment acceptance after learning the technique is significantly different between the groups.
- H6<sub>02</sub>: There is no difference in the mean treatment satisfaction after performing the technique for eight weeks,
- **H6<sub>a2</sub>: The mean of treatment satisfaction after performing the technique for eight weeks is significantly different between groups.**

### **1.9.3. Aim 3: Relationship between treatment adherence and treatment efficacy**

The relationship between Treatment Adherence and Treatment Efficacy was analyzed independently for each experimental group. Treatment Adherence was measured via self-report at Week 4 and Week 8. It was hypothesized that:

- $H_{7_{01}}$ : There is no relationship between treatment adherence and the change in self-reported trait anxiety in the Breathing group.
- **$H_{7_{a1}}$ : There is a positive correlation between increasing treatment adherence and a decrease in trait anxiety in the Breathing group.**
- $H_{7_{02}}$ : There is no relationship between treatment adherence and the change in self-reported trait anxiety in the Counting group.
- **$H_{7_{a2}}$ : There is a positive correlation between increasing treatment adherence and a decrease in trait anxiety in the Counting group.**



## **Chapter 2. METHODS**

### **2.1. Study Participants**

Students within Simon Fraser University (SFU) undergraduate Psychology courses were eligible to participate in the study for course credit. Interested participants filled out an online questionnaire, including the State-Trait Inventory for Cognitive and Somatic Anxiety, Trait Scale (STICSA-trait; Ree et al., 2000). Study eligibility requirements included a minimum score of 37 on the STICSA-trait scale (reported as a cut-off for mild anxiety in a student population by Van Dam et al., 2013), no current engagement in a regular contemplative practice (defined as >3 times a week) including, yoga, a formal mindfulness practice, meditation, or controlled breathing exercises, and currently not seeing a mental health professional for psychotherapy or counselling. Additionally, participants were required to be willing and able to download a mobile cell phone application compatible with either Android or I-phone operating systems.

In accordance with Tri-Council policy, the study was approved by the Simon Fraser University Research Ethics Board.

### **2.2. Materials**

Demographic information, relevant medication information, and recent substance use were collected with a questionnaire (Appendix A).

Measurement of trait anxiety was measured with the self-reported State-Trait Inventory for Cognitive and Somatic Anxiety (STICSA, Appendix B) (Ree et al., 2000). The 21-item questionnaire is composed of 11 items relating to Cognitive Anxiety and 10 items relating to Somatic Anxiety. The cognitive and somatic subscales have been supported by confirmatory factor analysis models and both subscales have been found to have high internal consistency (alphas > .87; Gros et al., 2007) and acceptable test-retest reliability ( $r_s > .65$ ; Ree et al. 2008). Test-retest correlations over the span of two months have been reported for scores on the trait somatic ( $r = 0.60$ ) and trait cognitive ( $r = 0.66$ ) subscales (Ree et al., 2008). Correlations between scores on the cognitive and somatic subscales range from  $r = .53$  to  $r = .83$  (Grös et al., 2007; Grös et al., 2010; Ree et al., 2008; Van Dam et al., 2013).

In a direct comparison with a measure of anxiety commonly used in anxiety research, the STAI, the STICSA was shown to be less strongly correlated with a measure of depression. These findings suggest that the STICSA may be a purer measure of anxiety symptomatology than the STAI (Grös et al., 2007).

State affect as a descriptive measurement was measured with the self-rated 20-item Positive Affect Negative Affect Schedule (PANAS, Appendix C) (Watson, Clark, & Tellegen, 1988). The PANAS is a reliable estimate of two broad and largely independent factors implicated in current emotional experience: positive affect and negative affect. The two scales exhibit acceptably high internal consistency (Cronbach's coefficient  $\alpha$ : .85) and low intercorrelations ( $-.15$ ). Each item is rated on a five-point Likert Scale, ranging from 1 = *Very Slightly or Not at all* to 5 = *Extremely*, to measure the extent to which the affect is being experienced at the present moment.

Acceptability of the intervention was assessed both immediately after learning the intervention (Appendix D) and upon study completion (Appendix E) with questionnaires using 5-point Likert scales to assess possible treatment accessibility barriers including perceived treatment complexity, accessibility, interference with daily life, and capability.

Participants were required to have regular access to a personal mobile cellular phone compatible with either iOS or android operating systems and be required to download the in-house developed mobile cell phone application TimerX. Intervention Adherence was inferred based on how frequently the app was used, based on a usage tracker built into the app design. Adherence was quantified by dividing the total number of times the app was used over the study period (maximum of twice per day), divided by the total number of times the app could have been used (if used twice a day everyday throughout the study).

Blood pressure was measured using an upper-arm blood pressure monitor (Model A&D UA-774) validated by the British Hypertension Society (DABL Educational Trust Ltd, n.d.). HRV data was measured using a CorSense Heart Rate Variability Finger Sensor and analyzed with the Elite HRV mobile application.

## **2.3. Experimental Protocol**

### **2.3.1. Covid-19 Pandemic Resulted in Two Cohorts**

Following completion of the electronic screening questionnaire, eligible participants were invited to participate in the study.

The total study included two cohorts of participants. The first cohort of participants completed all study data in the lab in the presence of a research assistant (n = 67). All in-person data was collected between September 2019 and March 2020. Participants were taught the brief relaxation intervention in-person and were provided with direct initial feedback on their performance upon learning the technique. These participants had physiological data measured after completing study questionnaires, including heart rate and blood pressure. Some participants in this cohort did not complete the final Week 8 time point due to the beginning of the Covid-19 lockdown restrictions taking place in Vancouver, BC. Participants were students enrolled in Psychology undergraduate courses at Simon Fraser University, predominantly attending in-person lectures. As a result of the Covid-19 restrictions, participants were unable to participate in-person in the study.

The second cohort of participants completed all study data through online questionnaires (n = 44). Data for this cohort was collected between September 2020 and April 2021. All instructions for learning and performing the technique were provided to participants via written text electronically. This cohort's data was collected amidst the Covid-19 global pandemic, at a time when in-person teaching was no longer offered at the university. No physiological data was collected from this cohort. Participants were also students enrolled in Psychology undergraduate courses at Simon Fraser University, however all courses were being delivered remotely. Participants were instructed to find a private location to complete all study questionnaires, but the environmental context of participants while engaging with the study material is unknown.

### **2.3.2. In-Person, Pre-COVID-19 Pandemic Cohort (n = 60)**

Participants electronically scheduled a lab appointment to have baseline measurements taken. Following appointment scheduling, participants were provided

recommendations for how to prepare in the 24 hours prior to coming to the lab, including:

- Try to get a good night's sleep the night before the assessment
- Try to avoid intense physical training the day before the assessment
- Try not to have a meal within two hours before the assessment
- Try to avoid caffeinated beverages two hours before the assessment
- Try to avoid alcohol for 24 hours prior to the assessment

Participants were randomly assigned to one of three groups: a Breathing group, a Counting group, and a Waitlist Control group (WLC).

Upon arriving for their scheduled appointment, participants completed an intake questionnaire including the self-reported PANAS (state) and the STICSA (trait). Resting-state physiological measurements of HRV and Blood Pressure were then taken for five minutes as participants sat quietly with soft instrumental music being played. The music consisted of guitar and light piano overlaid with sounds of lapping waves. Participants were instructed to: "Just close your eyes, listen to the music, and breathe naturally."

Participants HRV was measured with a CorSense HRV Finger Sensor on their right pointer finger. After one minute of wearing the device with soft music playing, the sensor recorded resting state HRV variables for four minutes. Then, four blood pressure readings were taken on the right arm using an upper-arm blood pressure monitor with 1-minute intervals between each reading. The average of the last three readings was used as the outcome measure for SBP and DBP measurements, respectively. Administrators were instructed to turn away from the participant as the readings were being taken to minimize observer–subject interactions.

#### Cell Phone Application Download

Participants were then instructed to download the in-house produced mobile cell phone application, 'TimerX'. Research assistants instructed participants in how to set-up the app and then explained the intervention sequence. Research assistants performed the one-minute sequence in front of the participant. Participants were then asked to perform the sequence. Feedback was provided on the participant's performance, both validating proper technique and offering corrections. If corrections were given, participants were

asked to perform the technique once more. Participants chose two times during the day to receive reminder notifications to complete the technique, which was programmed into the app by the research assistant. Finally, acceptability of the intervention was assessed immediately after learning the intervention with completion of a questionnaire assessing treatment accessibility barriers including perceived treatment complexity, accessibility, interference with daily life, and capability (Appendix D).

Participants were sent weekly reminder emails to use the mobile app 'TimerX' to complete the brief relaxation technique twice a day, every day.

Acceptability of the intervention was assessed both immediately after learning the intervention (Appendix D) and upon study completion (Appendix E) with questionnaires using 5-point Likert scales to assess possible treatment accessibility barriers including perceived treatment complexity, accessibility, interference with daily life, and capability.

***Instructions for Breathing Group:***

- Place the tip of your tongue against the ridge of tissue just behind your upper front teeth
- A 3 second inhalation through the nose
- A 5.2 second retention of the breath
- A 6 second audible exhalation through the mouth, making a 'whoosh' sound through pursed lips

This cycle was performed four times, for a total duration just under a minute. Relevant timing cues for each sequence were provided by the downloaded mobile cell phone application: 'TimerX'. The in-house developed mobile cell phone application (compatible with either iOS or android operating systems) provided visual cues combined with a user-specified auditory and/or tactile cue indicative of the breath phase to be performed throughout the breath sequence. Additionally, the cell phone application sent participants twice-daily reminders and recorded each time the application was operated by the user.

Participants were instructed to perform this one-minute breath sequence twice daily for four weeks. After the first four weeks, participants were asked to increase the duration to eight cycles per sequence (two minutes), twice a day (four minutes total per day) for the final four weeks of the study. The increase in cycles at four weeks is in accordance with the technique protocol (Weil, 2000).

### ***Instructions for Counting Group:***

This group also downloaded the mobile cell phone application: 'TimerX'. Parameters of the app were set for the app to count down from 10 to 1 in one second increments, repeating six times, for a total of one minute. Participants were asked to watch the screen for the duration of the one-minute. No mention of breathing was made. Similarly, the cell phone application sent participants twice-daily reminders and recorded each time the application was operated by the user.

Participants were instructed to perform this counting sequence twice daily for four weeks. After the first four weeks, participants were instructed to increase the duration to two minutes, twice a day (four minutes total per day) for the final four weeks of the study.

### **Waitlist Control (WLC):**

This control group came in for baseline measurements and were then given no instruction on how to spend the next four weeks. Upon returning for the Week 4 follow-up, the group downloaded the 'TimerX' app and were instructed on how to perform the breathing sequence, just as the Breathing group was instructed. The WLC was taught the technique at Week 4 to 1) help prevent drop-out and 2) in consideration of an ethical obligation to offer the control group the active treatment.

### **2.3.3. Online, During COVID-19 Pandemic Cohort (n = 44)**

Eligible participants were randomized into either the Breathing group or the Counting group and emailed a link to a study consent form and baseline questionnaire including the self-reported PANAS (state) and the STICSA (trait) measures. The questionnaire terminated with a step-by-step sequence of instructions to download the app, 'TimerX', and to learn the sequence correlated with the group they were randomized into. No Waitlist Control group was included in the online cohort.

Participants were sent weekly reminder emails to use the mobile app, 'TimerX' to complete the brief relaxation technique twice a day, every day.

At Week 4, participants were sent the Week 4 questionnaire including self-reported PANAS (state) and the STICSA (trait) measures, and a self-reported check of how

frequently they had been using the mobile app, 'TimerX'. They were then instructed to increase the duration of performing the technique to 2 minutes, twice a day, every day.

At Week 8 participants were sent the Week 8 questionnaire including the self-reported PANAS (state) measure, the STICSA (trait) measure, a self-reported check of how frequently they had been using the mobile app, 'TimerX', and a six question scale assessing treatment acceptance and satisfaction. Following submission of this questionnaire, participants were provided with a debriefing email.

## **2.4. Analytical Procedures**

The mean was used to describe each continuous outcome variable (STICSA trait anxiety, Systolic Blood Pressure (SBP), Diastolic Blood Pressure (DBP), Root Mean Square of the Successive Differences (RMSSD), Standard Deviation of Normal to Normal R-R intervals (SDNN)), along with measures of variability including the range and standard deviation. Variables were collected at the following time points:

- 1) Baseline (demographic, self-reported psychological outcomes, physiological outcomes for in-person cohort)
- 2) Week 4 time point, (self-reported psychological outcomes, physiological outcome for in-person cohort, objective treatment adherence, self-report treatment adherence)
- 3) Week 8 time point, (self-reported psychological outcomes, self-report treatment adherence)

A Chi-Square test was used to assess potential between-group differences of categorical demographic variables of sex and English as First Language speakers. Group differences in the continuous variables of age and frequency of TimerX mobile application-use was assessed with an independent samples t-test. Possible confounding effect of the variable(s) of theoretical interest or statistical interest were considered in the main statistical model. These tests were used to assess potential group differences among different cohorts of data collection (In-Person data collection vs Online data collection).

The distribution of the outcome variables (STICSA Trait Total, STICSA cognitive, STICSA somatic, SBP, DBP, RMSSD, and SDNN) were examined for assumptions of

normality, homogeneity of variance, and sphericity of the covariance matrix. Normality was assessed statistically via the Shapiro-Wilk's test. The homogeneity of variance was assessed using Levine's F test. Sphericity was assessed with Mauchly's test. In the case of statistical outliers (data points greater than three standard deviations from the mean), the data was trimmed according to Wilcoxon's recommendations for trimmed means and Winsorized variances. When outcome variables were still non-normally distributed, or the assumption of homogeneity of variances was still violated, data transformations were utilized (using the natural log). If normality assumptions are still violated, nonparametric analyses were performed.

When participants were lost to follow-up, the data were dropped from the analysis. Numbers of study drop-outs were compared between groups, and baseline data was compared between drop-outs and completers to ensure dropping-out was due to random chance.

#### **2.4.1. Repeated Measures Mixed ANOVA**

Outcome variables that met the assumptions of normality, sphericity, and homogeneity of variance were analyzed for group differences with a repeated measures mixed analysis of variance (ANOVA). In analysis, intervention group was the independent variable of interest with time as the within-subjects factor. The analysis of interest investigated an interaction between group and time, to determine if the outcome variable changes differently over time among different groups. Analyses controlled for age, sex, the effect of study cohort (In-person vs Online) and for the frequency with which the mobile app 'TimerX' was used throughout the study period. Covariates were selected as there have been repeated studies showing a relationship between measures of anxiety and age (Mahoney et al., 2015), sex (Jalnapurkar et al., 2018) and adherence (Alfonsson et al., 2015).

To limit the number of statistical tests performed, the primary outcome variable of interest of Trait Anxiety was STICSA Total score. The Cognitive Anxiety and Somatic Anxiety subscales were only investigated individually when STICSA Total indicated an effect of  $p < 0.10$ . When STICSA Cognitive Anxiety and Somatic Anxiety subscales were investigated in follow-up analyses, alpha value for significance was adjusted from 0.05 to 0.025.



Follow-up Tukey's post-hoc tests were performed when there were simple main effects to identify specific comparisons of interest.

### **2.4.2. Nonparametric Analyses**

When outcome variables remained non-normally distributed after being Winsorized and log-transformed, the non parametric Mann-Whitney U test or Wilcoxon Signed Ranks test were utilized. The Mann-Whitney U-test was used when analyzing two independent samples, whereas the Wilcoxon Signed Ranks test was used with two dependent samples.

### **2.4.3. Pearson's Correlation**

The relationship between two variables was investigated using a Pearson's correlation. This was used to determine a relationship between treatment adherence and treatment efficacy.

### **2.4.4. Sample Size**

Power calculations utilized G-Power v3.1.7 assumed a small effect size (0.3), which is a smaller effect than those observed in other relaxation training interventions in the treatment of anxiety (Cohen's  $d=0.5$ ; Manzoni et al., 2008). The power calculation was performed for a repeated measures ANOVA investigating the time\*group interaction for three groups and two time points. For this statistical test, given an alpha of 0.05 at 0.8 power, and assuming a small effect of the intervention (Cohen's  $d=0.3$ ), a minimum total sample size of  $n=81$  (27 per group) was determined to be required to show the proposed differences (see Table 1). The achieved total sample size of  $n=104$  was unevenly dispersed across the three groups, including: Breathing group ( $n=45$ ), Counting group ( $n=36$ ), and WLC ( $n=23$ ). The analyzed sample was powered at 57.9% to detect a small effect, 96.2% to detect a medium effect, and 99.9% to detect a strong effect.

**Table 1 Power Calculations for 3 Groups over 2 Time Points**

Power	Groups	Time Points	$\alpha$	Cohens d	Group sample size	Total Group sample size (15% dropout)
0.8	3	2	.05	0.3	27	31
0.85	3	2	.05	0.3	30	35
0.9	3	2	.05	0.3	34	39

The time\*group interaction comparing the Counting group and the Breathing group across three time points (2 groups x 3 time points) was also assessed for power. The achieved total sample size of n=75 was unevenly dispersed across the two groups, with Breathing group (n=42) and Counting group (n=33). The analyzed sample was powered at 75.6% to detect a small effect, 99.5% to detect a medium effect, and 99.9% to detect a strong effect

**Table 2 Power Calculations for 2 Group by 3 Time Point Comparisons**

Power	Groups	Time Points	$\alpha$	Cohens d	Group sample size	Total Group sample size (15% dropout)
0.8	2	3	.05	0.3	26	30
0.85	2	3	.05	0.3	30	35
0.9	2	3	.05	0.3	34	39

## Chapter 3. RESULTS

Of the 687 participants who completed the initial screening survey, 347 (50.5%) participants were excluded for not meeting study criteria. Specific exclusion criteria included: STICSA Total score less than pre-determined cut-off for study participation (STICSA Total < 37; n = 267, 38.9%), current access to a mental health professional (n = 59, 8.6%), current engagement with a regular contemplative practice (n = 40, 5.8%) or unwilling and/or unable to download a mobile app (n = 34, 4.9%).

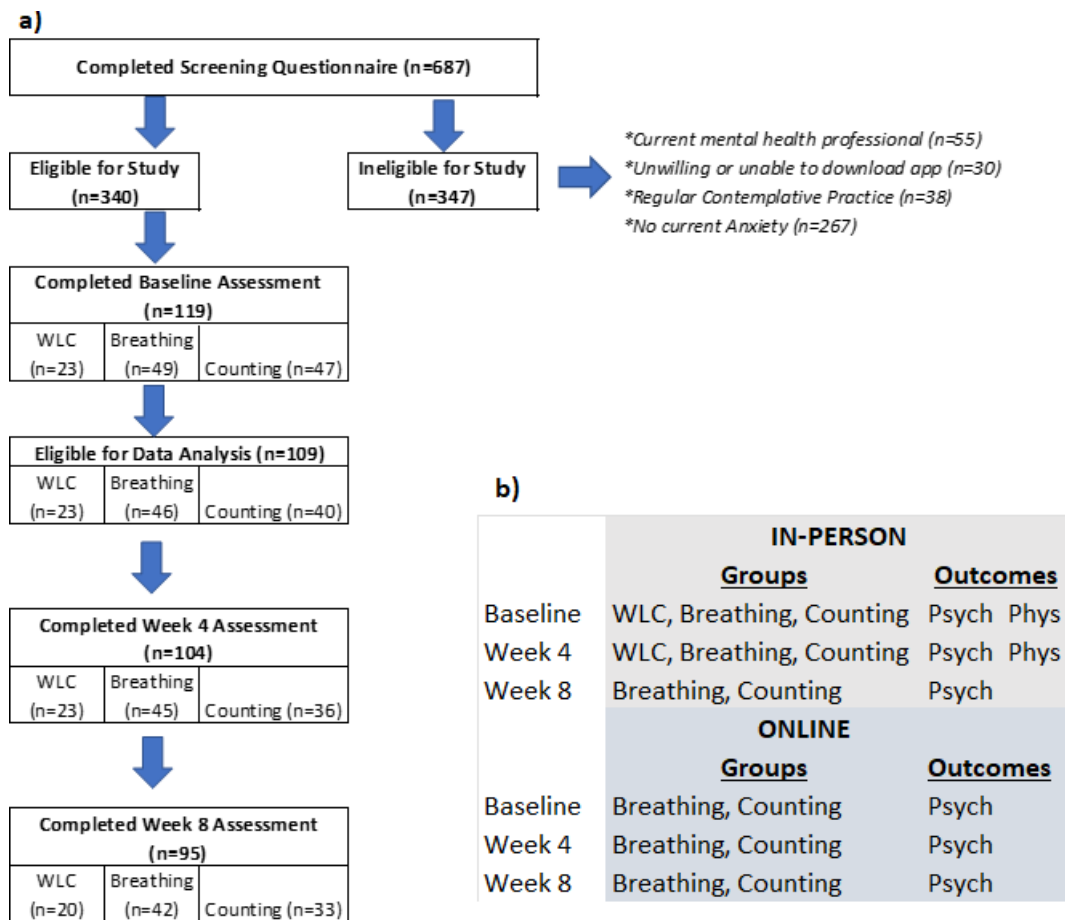
The remaining 340 participants were invited to join the 8-week study in exchange for participation credit. Of those invited, 119 (34.7%) completed the baseline assessment. 114 participants were retained after 4 weeks (4.2% drop out rate) and 104 were retained after 8 weeks (8.7% drop out rate). From baseline to study completion there was a 12.6% drop out rate. By experimental group, the drop-outs rates were 13.0% for the waitlist control, 8.1% for the breathing group, and 17% for the counting group. A chi-square test of independence was performed to examine if there was a relation between dropout rates and group. The relation between these variables was not significant,  $\chi^2(2, N = 119) = 1.714, p = .425$ . Exploratory analyses found no significant differences in any baseline variables between participants who were retained throughout eight-weeks (n=104) compared to those lost to follow-up (n=15). Statistical comparisons can be found in Appendix F.

Upon data analysis, ten participants data were excluded due to STICSA trait scores below 35 at Baseline measurement, resulting in 104 participants data eligible for analysis at Week 4, and 95 participants eligible for data analysis at Week 8.

Of the 104 participants eligible for data analysis at Week 4, 60 participants were from the in-person (pre-Covid-19) study cohort and 44 participants were from the on-line (during Covid-19) study cohort.

To the researcher's knowledge, there were no adverse effects of the study upon the study participants.

**Figure 1. Sample Selection Flow Chart**



Note: a) Flow chart of study participants moving through study b) table of the study groups, and outcome variables in the two study cohorts (in-person and online). Psych = psychological outcomes (self-reported trait anxiety), Phys = physiological outcomes (heart rate variability and blood pressure).

### 3.1. Descriptive Statistics

Descriptive statistics for demographic and outcome variables are reported for the in-person, pre-Covid-19 data cohort (n=60, section 3.1.1 below), the online, during Covid-19 data cohort (n=44, section 3.1.2), and the combined total sample (n=104, section 3.1.4 below).

#### 3.1.1. In-Person, Pre-Covid-19 Cohort

Of the 104 participants eligible for data analysis at Week 4, 60 participants were from the pre-Covid-19, in-person cohort. The three study groups consisted of the

Breathing group (n=22), the Counting group (n=15), and the Waitlist Control group (n=23). Descriptive statistics of the total in-person sample, as well as the sample partitioned into experimental study groups for baseline, Week 4, and Week 8 time points is included in Table 3.

### **3.1.2. Online, During Covid-19 Cohort**

Of the 104 participants eligible for data analysis at Week 4, 44 participants were from the Online, During-Covid-19 cohort. The online cohort only recruited participants for the Breathing (n=23) and the Counting (n=21) study groups. Descriptive statistics of the total online sample, as well as the sample partitioned into experimental study groups for baseline, Week 4, and Week 8 time points is included in Table 4.

### **3.1.3. Online and In-Person Cohort Comparison**

For the 104 participants with valid data who were retained at Week 4, group differences were explored between the Online Cohort (n=44) and the In-Person cohort (n=60) to determine if cohorts could be combined in analyses.

A Pearson Chi-Square test was used to assess potential between-cohort differences of Online cohort vs In-Person cohort for the categorical demographic variables sex and English as First Language Status. The relation between cohort and sex was insignificant,  $X^2(1, N = 104) = 3.574, p = .059$ , with a trend towards the online cohort having more women. The relation between cohort and English as First Language Status was also insignificant,  $X^2(1, N = 104) = 1.258, p = .262$ . Additionally, assessment of cohort differences in participant technique acceptance was insignificant,  $X^2(1, N = 104) = 2.314, p = .128$ , though there was a trend towards more perceived barriers to performing the technique in the online group compared to the in-person group.

A Student's t-test was used to assess potential between-cohort differences of the continuous variable of age, as well as baseline variables of total anxiety (STICSA Total), somatic anxiety (STICSA somatic), cognitive anxiety (STICSA Cognitive), positive affect (PANAS pos) and negative affect (PANAS neg), shown in Table 6.

The online cohort reported a higher score on the PANAS negative scale ( $M = 19.23, SD = 6.78$ ) than the in-person cohort ( $M = 16.53, SD = 6.00$ ), which was statistically

significant ( $t(102) = -2.142, p = 0.035$ ). Statistical significance did not withstand corrections for multiple comparisons. No other continuous variables, including the primary outcome of interest (STICSA Total), were different between cohorts. The cohorts were deemed similar enough to combine into one aggregate group for the main analyses.

**Table 3 Descriptive Statistics for the In-Person, Pre-Covid-19 Study Cohort**

**BASELINE – In-Person Cohort**

	<u>Total (n=60)</u>		<u>WLC (n=23)</u>		<u>Breathing (n=22)</u>		<u>Counting (n=15)</u>	
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range
Sex (% Female)	80.0%		78.3%		77.4%		86.7%	
First Language (% English)	75.0%		73.9%		77.3%		73.4%	
No Tx Barriers	86.7%		82.6%		90.9%		86.7%	
Age	18.88 (1.47)	17-26	18.65 (1.07)	17-22	18.77 (1.34)	17-23	19.40 (2.06)	18-26
PANAS Pos	26.15 (6.64)	12-46	26.61 (6.89)	12-41	25.59 (7.76)	14-46	26.27 (4.54)	17-33
PANAS Neg	16.53 (6.00)	10-36	17.91 (6.49)	10-36	16.50 (6.08)	10-34	14.48 (4.70)	10-27
STICSA Total	48.10 (9.70)	35-75	49.13 (10.07)	35-67	50.32 (9.9)	38-75	43.27 (9.04)	36-61
STICSA Som	21.63 (6.04)	13-44	21.91 (4.90)	14-32	22.68 (7.48)	16-44	19.67 (5.12)	13-30
STICSA Cog	26.47 (5.89)	13-38	27.22 (7.17)	13-38	23.60 (4.97)	21-38	23.60 (4.05)	17-32
Systolic BP	110.7 (9.7)	94-135	111.1 (9.9)	95-128	109.0 (9.2)	94-131	112.7 (10.3)	95-135
Diastolic BP	67.4 (6.6)	53-87	68.5 (5.05)	59-76	66.5 (6.5)	53-87	66.9 (8.6)	53-87
RMSSD	41.6 (17.8)	13-89	40.9 (16.9)	15-83	37.1 (15.1)	13-57	49.3 (21.1)	18-90
SDNN	54.1 (20.7)	13-128	52.2 (23.2)	19-127	52.4 (17.9)	25-94	59.5 (20.7)	13-101

**WEEK 4 FOLLOW-UP – In-Person Cohort**

	<u>Total (n=60)</u>		<u>WLC (n=23)</u>		<u>Breathing (n=22)</u>		<u>Counting (n=15)</u>	
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range
PANAS Pos	23.70 (7.12)	10-44	23.83 (6.41)	12-38	23.23 (8.61)	10-44	24.20 (6.14)	10-34
PANAS Neg	15.80 (5.90)	10-34	17.13 (6.36)	10-34	15.64 (6.26)	10-32	14.00 (4.23)	10-24
STICSA Total	46.72 (10.80)	27-74	48.57 (11.59)	31-74	48.36 (9.55)	29-64	41.47 (10.21)	31-60

STICSA Som	20.95 (5.90)	11-36	22.09 (6.40)	13-36	21.59 (5.03)	14-35	18.27 (5.80)	11-29
STICSA Cog	25.77 (6.27)	13-39	26.48 (6.16)	14-38	26.77 (7.00)	13-39	23.20 (4.82)	17-33
Systolic BP	108.9 (9.1)	92-133	110.9 (9.0)	96-131	105.2 (7.8)	92-125	111.6 (9.8)	100-133
Diastolic BP	67.3 (6.3)	57-87	68.7 (5.9)	60-79	65.8 (5.7)	58-79	67.5 (7.6)	57-87
RMSSD	48.1 (27.8)	11-156	41.6 (20.0)	11-90	53.3 (33.9)	14-156	50.4 (28.2)	19-134
SDNN	29.2 (27.9)	16-205	50.0 (17.0)	27-83	67.5 (37.1)	28-205	61.0 (22.6)	16-112

**WEEK 8 FOLLOW-UP – In-person Cohort**

	Total (n=54)		WLC (n=20)		Breathing (n=21)		Counting (n=13)	
	M (SD)	Range	M (SD)	Range	M (SD)	Range	M (SD)	Range
PANAS Pos	23.11 (8.51)	10-43	24.30 (9.15)	11-41	23.38 (9.16)	10-42	20.85 (6.23)	13-36
PANAS Neg	16.44 (7.25)	9-42	17.55 (8.27)	10-42	16.29 (6.60)	10-34	15.00 (6.89)	10-34
STICSA Total	43.96 (11.71)	24-70	44.80 (13.55)	25-70	44.14 (11.34)	24-61	42.38 (9.84)	28-59
STICSA Som	19.63 (5.83)	11-35	20.35 (6.87)	11-35	19.52 (4.73)	12-27	18.69 (6.05)	11-28
STICSA Cog	24.33 (7.03)	11-37	24.45 (7.70)	13-37	24.62 (7.78)	11-37	23.69 (4.83)	16-32
Tx Satisfaction	23.8 (3.5)	14-30	24.9 (2.2)	20-29	23.3 (3.8)	16-30	23.2 (4.4)	14-29
	Total (n=41)		WLC (n=15)		Breathing (n=17)		Counting (n=9)	
	M (SD)	Range	M (SD)	Range	M (SD)	Range	M (SD)	Range
Systolic BP	107.8 (8.8)	89-128	108.0 (11.1)	89-128	106.7 (8.1)	91-121	109.8 (6.4)	100-120
Diastolic BP	65.9 (5.9)	56-78	64.8 (5.8)	58-76	65.9 (6.8)	56-78	67.6 (4.0)	62-73
RMSSD	49.5 (20.9)	13-96	48.8 (19.5)	13-80	50.5 (18.1)	23-91	48.5 (29.6)	13-96
SDNN	62.8 (30.6)	19-143	69.6 (29.1)	31-124	64.7 (35.4)	27-143	49.2 (19.4)	19-79



**Table 4 Descriptive Statistics for the Online, During-Covid-19 Study Cohort**

**BASELINE – Online Cohort**

	<u>Total (n=44)</u>		<u>Breathing (n=23)</u>		<u>Counting (n=21)</u>	
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range
Sex (% Female)	93.2%		100%		85.7%	
First Language (% English)	84.1%		87.0%		81.0%	
No Tx Barriers	75.0%		78.3%		71.4%	
Age	18.82 (1.62)	17-25	18.61 (1.73)	17-25	19.05 (1.50)	17-23
PANAS Pos	27.09 (6.73)	15-42	26.17 (7.52)	15-42	28.10 (5.74)	16-37
PANAS Neg	19.23 (6.78)	10-36	18.70 (6.73)	10-36	19.81 (6.94)	10-35
STICSA Total	50.55 (9.55)	37-73	50.91 (10.1)	37-73	50.14 (9.09)	38-67
STICSA Som	22.61 (5.92)	12-36	22.91 (6.58)	13-36	22.29 (5.24)	12-33
STICSA Cog	27.93 (5.24)	11-39	28.00 (5.14)	19-37	27.86 (5.47)	11-39

**WEEK 4 FOLLOW-UP – Online Cohort**

	<u>Total (n=44)</u>		<u>Breathing (n=23)</u>		<u>Counting (n=21)</u>	
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range
PANAS Pos	25.59 (7.10)	11-41	25.61 (7.64)	15-41	25.57 (6.64)	11-37
PANAS Neg	18.30 (5.77)	9-33	18.17 (5.37)	9-28	18.43 (6.31)	11-33
STICSA Total	44.43 (8.76)	27-63	42.74 (10.04)	27-63	46.29 (6.87)	33-59
STICSA Som	20.07 (4.70)	12-30	19.30 (5.01)	12-30	20.90 (4.30)	15-29
STICSA Cog	24.36 (5.47)	15-34	23.43 (6.19)	15-33	25.38 (4.50)	16-34

**WEEK 8 FOLLOW-UP – Online Cohort**

	<u>Total (n=41)</u>		<u>Breathing (n=21)</u>		<u>Counting (n=20)</u>	
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range
PANAS Pos	26.15 (7.55)	11-43	27.57 (7.15)	13-43	24.65 (7.84)	11-39
PANAS Neg	17.63 (7.18)	9-36	16.57 (7.24)	9-36	18.75 (7.12)	10-35
STICSA Total	41.41 (8.35)	24-61	39.00 (7.54)	24-54	43.95 (8.59)	32-61
STICSA Som	18.17 (4.76)	11-31	17.43 (4.78)	11-31	18.95 (4.73)	12-28
STICSA Cog	23.24 (4.87)	13-33	21.57 (4.66)	13-30	25.00 (4.54)	17-33
Tx Satisfaction	22.6 (3.2)	14-27	23.4 (3.1)	17-27	21.9 (3.1)	14-26

**Table 5 Descriptive Statistics Comparing the In-Person, Pre-COVID-19 cohort (n=60), and the Online, During COVID-19 cohort (n = 44)**

**BASELINE**

	<u>In-person (n=60)</u>		<u>Online (n=44)</u>	
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range
Sex (% Female)	80.0%		93.2%	
First Language (% English)	75.0%		84.1%	
No Tx Barriers	86.7%		75.0%	
Age	18.88 (1.47)	17-26	18.82 (1.62)	17-25
PANAS Pos	26.15 (6.65)	12-46	27.09 (6.73)	15-42
PANAS Neg	16.53 (6.00)	10-36	19.23 (6.78)	10-36
STICSA Total	48.10 (9.70)	35-75	50.55 (9.55)	37-73
STICSA Som	21.63 (6.04)	13-44	22.61 (5.92)	12-36
STICSA Cog	26.47 (5.89)	13-38	27.93 (5.24)	19-39

**WEEK 4 FOLLOW-UP**

	<u>In-person (n=60)</u>		<u>Online (n=44)</u>	
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range
PANAS Pos	23.70 (7.13)	10-44	25.59 (7.10)	11-41
PANAS Neg	15.80 (5.90)	10-34	18.30 (5.78)	9-33
STICSA Total	46.72 (10.80)	29-74	44.43 (8.76)	27-63
STICSA Som	20.95 (5.90)	11-36	20.07 (4.70)	12-30
STICSA Cog	25.77 (6.27)	13-39	24.36 (5.47)	15-34

**WEEK 8 FOLLOW-UP**

	<u>In-Person (n=54)</u>		<u>Online (n=41)</u>	
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range
PANAS Pos	23.11 (8.51)	10-42	26.15 (7.55)	11-43
PANAS Neg	16.44 (7.25)	10-42	17.63 (7.18)	9-36
STICSA Total	43.96 (11.71)	24-70	41.41 (8.35)	24-61
STICSA Som	19.63 (5.84)	11-35	18.17 (4.76)	11-31
STICSA Cog	24.33 (7.04)	11-37	23.24 (4.87)	13-33

**Table 6 Independent Samples T-tests comparing the In-Person, Pre-COVID-19 cohort to the Online, During COVID-19 cohort**

Independent Samples Test: Online vs In-Person Cohorts							
	Levene's Test for Equality of Variances		t-test for Equality of Means				
	F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference
Age	1.044	0.309	0.214	102	0.831	0.065	0.305
Total Anxiety	0.034	0.854	-1.278	102	0.204	-2.445	1.913
Somatic Anxiety	0.258	0.612	-0.824	102	0.412	-0.980	1.189
Cognitive Anxiety	1.241	0.268	-1.312	102	0.192	-1.465	1.117
Positive Affect	0.245	0.622	-0.710	102	0.480	-0.941	1.326
Negative Affect	1.698	0.196	-2.142	102	0.035	-2.694	1.258

Note: Comparison between In-Person, Pre-COVID-19 cohort (n=60) to the Online, During COVID-19 cohort (n = 44). Levene's F Test is also shown to test the assumption of equal variances among groups. Significance set at  $\alpha < .05$ .

### 3.1.4. Experimental Groups

For the 104 participants with valid data who were retained at Week 4, 85.6% of the sample was female and 78.8% reported English as their first spoken language. Demographic information and outcome variables at baseline, Week 4, and Week 8 are reported in Table 9. In determining reliability of the self-reported Trait Anxiety scales, the STICSA Total Anxiety scale consisted of 21 items ( $\alpha=.844$ ), the STICSA Somatic subscale consisted of 11 items ( $\alpha=.822$ ) and the STICSA Cognitive subscale consisted of 10 items ( $\alpha=.797$ ). Correlations between scores on the Cognitive and Somatic subscales was .40 at baseline, .55 at Week 4, and .61 at Week 8. Correlation between Somatic and Cognitive Anxiety trait scores is lower compared to reported ranges in the literature which range from  $r = .53$  to  $r = .83$  (Grös et al., 2007; Grös et al., 2010; Ree et al., 2008; Van Dam et al., 2013).

**Table 7 Descriptive Statistics for Demographic and Clinical Variables of the Total Sample (n=104)**

<b>BASELINE</b>									
	<b><u>Total (n=104)</u></b>		<b><u>WLC (n=23)</u></b>		<b><u>Breathing (n=45)</u></b>		<b><u>Counting (n=36)</u></b>		
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	
Sex (% Female)	85.6%		78.3%		88.9%		86.10%		
First Language (% English)	78.8%		82.2%		82.2%		77.8%		
No Tx Barriers	81.7%		82.6%		84.4%		77.8%		
Age	18.86 (1.53)	17-26	18.65 (1.07)	17-22	18.69 (1.54)	17-25	19.19 (1.74)	17-26	
PANAS Pos	26.55 (6.67)	12-46	26.61 (6.89)	12-41	25.89 (7.56)	14-46	27.33 (5.29)	16-37	
PANAS Neg	17.67 (6.45)	10-36	17.91 (6.49)	10-36	17.62 (6.45)	10-36	17.58 (6.60)	10-35	
STICSA Total	49.13 (9.67)	35-75	49.13 (10.07)	35-67	50.62 (9.9)	37-75	47.28 (9.04)	36-67	
STICSA Som	22.05 (5.98)	12-44	21.91 (4.90)	14-32	22.8 (6.95)	13-44	21.19 (5.29)	12-33	
STICSA Cog	27.09 (5.64)	13-39	27.22 (7.17)	13-38	27.82 (5.01)	19-38	26.08 (5.31)	17-39	
<b>WEEK 4 FOLLOW-UP</b>									
	<b><u>Total (n=104)</u></b>		<b><u>WLC (n=23)</u></b>		<b><u>Breathing (n=45)</u></b>		<b><u>Counting (n=36)</u></b>		
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	
PANAS Pos	24.50 (7.14)	10-44	23.83 (6.41)	12-38	24.44 (8.12)	10-44	25.00 (6.38)	10-37	
PANAS Neg	16.86 (5.95)	9-34	17.13 (6.36)	10-34	16.93 (5.90)	9-32	16.58 (5.90)	10-33	
STICSA Total	45.75 (10.01)	27-74	48.57 (11.59)	31-74	45.49 (10.10)	27-64	44.42 (8.73)	31-60	
STICSA Som	20.58 (5.42)	11-36	22.09 (6.40)	13-36	20.42 (5.10)	12-35	19.81 (5.08)	11-29	
STICSA Cog	25.17 (5.96)	13-39	26.48 (6.16)	14-38	25.07 (6.74)	13-39	24.47 (4.70)	16-34	

**WEEK 8 FOLLOW-UP**

	<u>Total (n=95)</u>		<u>WLC (n=20)</u>		<u>Breathing (n=42)</u>		<u>Counting (n=33)</u>	
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range
PANAS Pos	24.42 (8.20)	10-43	24.30 (9.15)	11-41	25.48 (8.39)	10-43	23.15 (7.39)	11-39
PANAS Neg	16.96 (7.21)	9-42	17.55 (8.27)	10-42	16.43 (6.84)	9-36	17.27 (7.17)	10-35
STICSA Total	42.86 (10.42)	24-70	44.80 (13.55)	25-70	41.57 (9.86)	24-61	43.33 (8.99)	28-61
STICSA Som	19.00 (5.42)	11-35	20.35 (6.87)	11-35	18.48 (4.82)	11-31	18.85 (5.20)	11-28
STICSA Cog	23.86 (6.19)	11-37	24.45 (7.70)	13-37	23.10 (6.52)	11-37	24.48 (4.63)	16-33

## **3.2. Primary Analyses**

Statistical analyses were utilized to address the three aims of this study on 1) the efficacy of the ultra-brief breathing intervention in decreasing trait anxiety, 2) the acceptability of the ultra-brief breathing intervention, and 3) the relationship of adherence to practice on treatment efficacy.

Treatment efficacy (Aim 1) was addressed with five distinct families of analysis, analyzing self-reported trait anxiety in 3 groups over 4 weeks (3.2.1.1), resting physiological variables in 3 groups over 4 weeks (3.2.1.2), self-reported trait anxiety in 2 groups over 8 weeks (3.2.1.3), self-reported trait anxiety in the waitlist control group over 8 weeks (3.2.1.4), and self-reported trait anxiety in a “High Anxiety” group over 4 weeks (3.2.1.5). The Cognitive Anxiety and Somatic Anxiety subscales were only investigated individually when STICSA Total indicated an effect of  $p < 0.10$

Treatment acceptability (Aim 2) was addressed via between group differences of perceived barriers to performing the technique.

The relationship between treatment adherence and treatment efficacy (Aim 3) was addressed via within-group correlations.

### **3.2.1. Aim 1: Efficacy of the Ultra-brief Breathing Intervention**

#### **3.2.1.1. Mean change in trait anxiety between Breathing, Counting, and Waitlist Control group after 4 weeks**

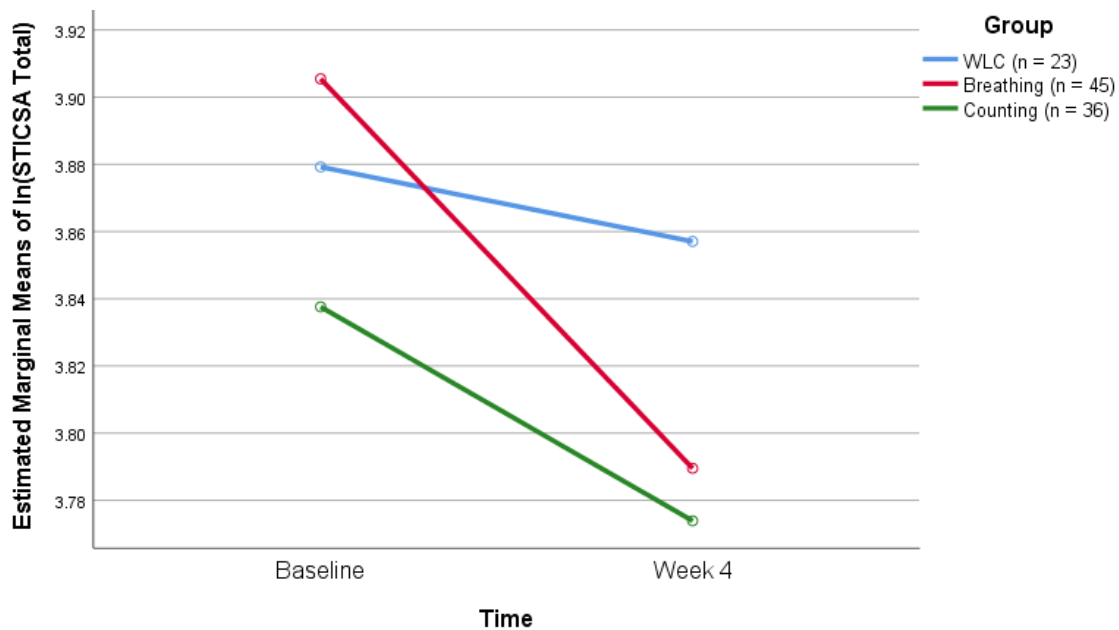
Group level distributions of the baseline STICSA Total Scores were examined to ensure assumption of repeated measures ANOVA were met. A Shapiro-Wilk test showed a significant departure from normality in all three groups, WLC, Breathing, and Counting. A log transformation was performed on STICSA Total scores, resulting in normal distributions in all three groups.

Levene’s F test indicated that the error variance of the log-transformed total anxiety scores was equal across groups. A repeated measures ANOVA, controlling for age and sex, determined that the mean of log-transformed self-reported Total Anxiety scores did not significantly differ between time points ( $F(1, 99) = .347, p = .557, \eta p^2 = .003$ ) or for

group ( $F(2, 99) = .948, p = .391$ ). There was no significant interaction between time and Group ( $F(2, 99) = 1.567, p = .214, \eta^2 = .031$ ), indicating the change in Total Anxiety scores over time was not different between groups, as shown in Figure 2.

All statistical output for analyses within Section 3.2.1.1. can be found in Appendix G.

**Figure 2** Estimated Marginal Means of a Repeated Measures ANOVA for Log Transformed STICSA Total Score, 3 Groups over 2 Time Points



Note: Estimated marginal means of a Repeated Measures ANOVA for Log Transformed STICSA Total Score. Covariates appearing in the model were evaluated at Age = 18.86 and Sex = 1.86 (male coded as 1, female coded as 2). There was no significant interaction between time and Group ( $F(2, 99) = 1.567, p = .214, \eta^2 = .031$ ).

### 3.2.1.2. Mean change in physiological resting state measures between the Breathing, Counting, and Waitlist Control group after 4 weeks.

The following resting state physiological measures were analyzed:

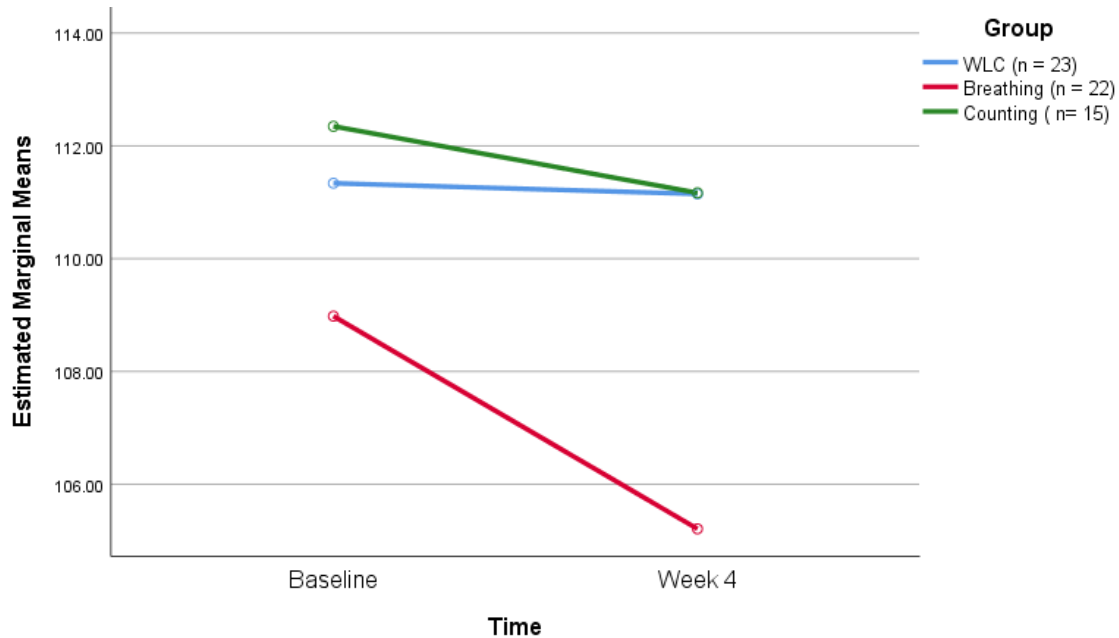
- ❖ Systolic Blood Pressure (SBP)
- ❖ Diastolic Blood Pressure (DBP)
- ❖ Root Mean Square of the Successive Differences (RMSSD)
- ❖ Standard Deviation of Normal to Normal R-R intervals (SDNN)

Distributions of physiological data outcomes of SBP, DBP, RMSSD and SDNN were assessed for normality at the group-level. All statistical output for analyses within Section 3.2.1.2. can be found in Appendix H.

### ***Systolic Blood Pressure***

SBP outcomes were normally distributed within all groups (Shapiro-Wilks  $p > .05$ ). A repeated measures ANOVA controlling for the effect of age and sex determined that the mean of SBP did not differ significantly between time points ( $F(1, 55) = 0.08, p = .779, \eta^2 = .001$ ). There was no significant effect of group ( $F(2,55)=1.942, p = .153$ ). There was no significant interaction between time and Group ( $F(2, 55) = 1.837, p = .169, \eta^2 = .063$ ), indicating the change in systolic blood pressure over time was not different between groups.

**Figure 3** Estimated Marginal Means of a Repeated Measures ANOVA for Systolic Blood Pressure



Note: Covariates appearing in the model include age and sex. There was no significant interaction between time and Group ( $F(2, 55) = 1.837, p = .169, \eta^2 = .063$ ).

### ***Diastolic Blood Pressure***

DBP outcomes were normally distributed within all groups (Shapiro-Wilks  $p > .05$ ). A repeated measures ANOVA controlling for the effect of age and sex determined that the



mean of DBP did not differ significantly between time points ( $F(1, 55) = 0.57, p = .454, \eta^2 = .01$ ). There was no significant effect of group ( $F(2,55)=1.165, p = .319$ ). There was no significant interaction between time and Group ( $F(2, 55) = 0.279, p = .758, \eta^2 = .01$ ), indicating the change in DBP over time was not different between groups.

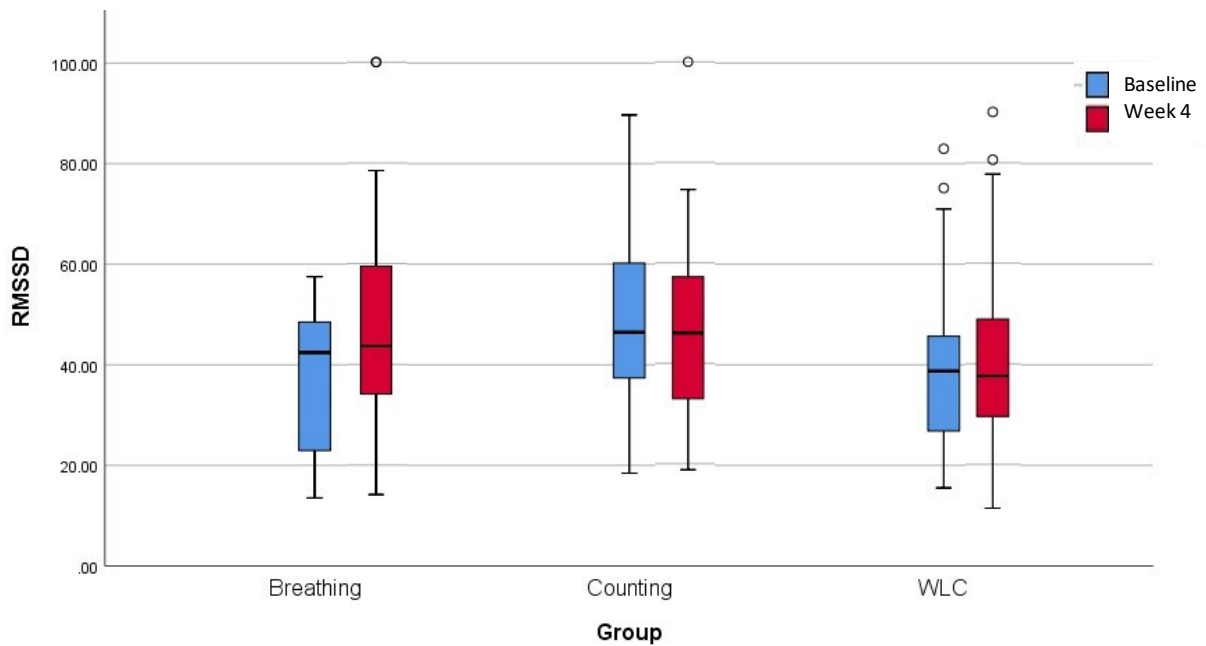
### ***RMSSD***

RMSSD outcomes by group and time point are displayed in Figure 4. The Shapiro-Wilk Test found distributions of RMSSD outcomes were nonnormally distributed at Baseline within the WLC group and the Breathing group, and at Week 4 within the Breathing group and the Counting group ( $W < .91, p < .05$ ). Log transformation of RMSSD outcomes did not provide a normal distribution within the baseline Breathing Group ( $W(22) = .873, p < .01$ ). To accommodate the non-normal distribution, the nonparametric Wilcoxon Signed Ranks test was performed separately for each of the three groups. A Bonferroni correction for the three comparisons established the  $\alpha = 0.017$  for significance testing.

In the Breathing group six participant's RMSSD outcomes decreased from Baseline to Week 4 and 16 participant's RMSSD outcomes increased from Baseline to Week 4. A Wilcoxon signed rank-test indicated that more individuals in the Breathing group had RMSSD scores that increased over four weeks than those that decreased ( $Z = -2.062, p = .039$ ). After controlling for multiple comparisons at  $\alpha = 0.017$ , this difference is interpreted as not significant.

In the Counting group, nine participants RMSSD outcomes decreased from Baseline to Week 4 and six participant's RMSSD outcomes increased from Baseline to Week 4. A Wilcoxon signed rank-test indicated that this difference was not statistically significant, ( $Z = -0.625, p = .532$ ). In the WLC group, 11 participants RMSSD outcomes decreased from Baseline to Week 4 and 12 participant's RMSSD outcomes increased from Baseline to Week 4. A Wilcoxon signed rank-test indicated that this difference was not statistically significant, ( $Z = -0.426, p = .670$ ).

**Figure 4 RMSSD Outcomes at Baseline and Week 4 Timepoints**

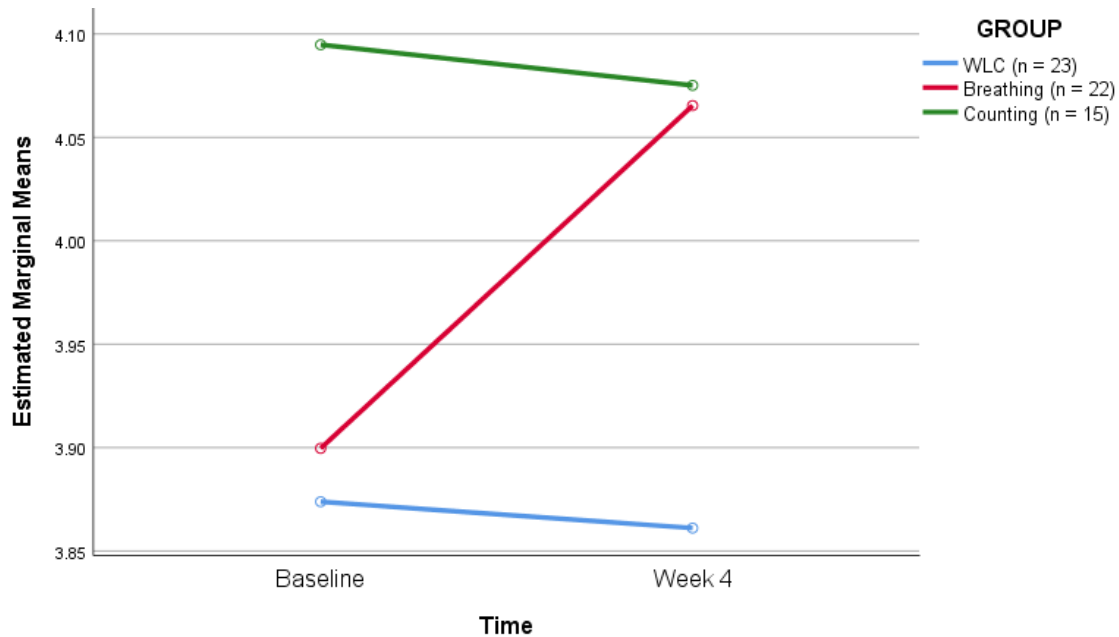


Note: Boxplots of RMSSD outcomes at baseline and four weeks. Group sample sizes Breathing n=20, Counting n=15, WLC n=23. Data represented here is nonnormally distributed.

### ***SDNN***

Distributions of SDNN outcomes were found to be nonnormally distributed at Baseline within the WLC group and at Week 4 within the Breathing group (Shapiro-Wilks  $p < .05$ ). The data were Winsorized for outlying data points, then Log transformed to obtain normal distributions within groups. A repeated measures ANOVA, controlling for age and sex, determined that the mean of log transformed SDNN did not differ significantly between time points ( $F(1, 55) = 1.683, p = .20, \eta^2 = .03$ ). There appeared to be an effect of group as shown in Figure 5, but it did not reach statistical significance ( $F(2,55) = 3.152, p = .051$ ). There was no significant interaction between time and Group ( $F(2, 55) = 1.750, p = .183, \eta^2 = .06$ ), indicating the change in log transformed SDNN over time was not statistically different between groups.

**Figure 5 Estimated Marginal Means of a Repeated Measures ANOVA for Log Transformed SDNN Measure of Heart Rate Variability**

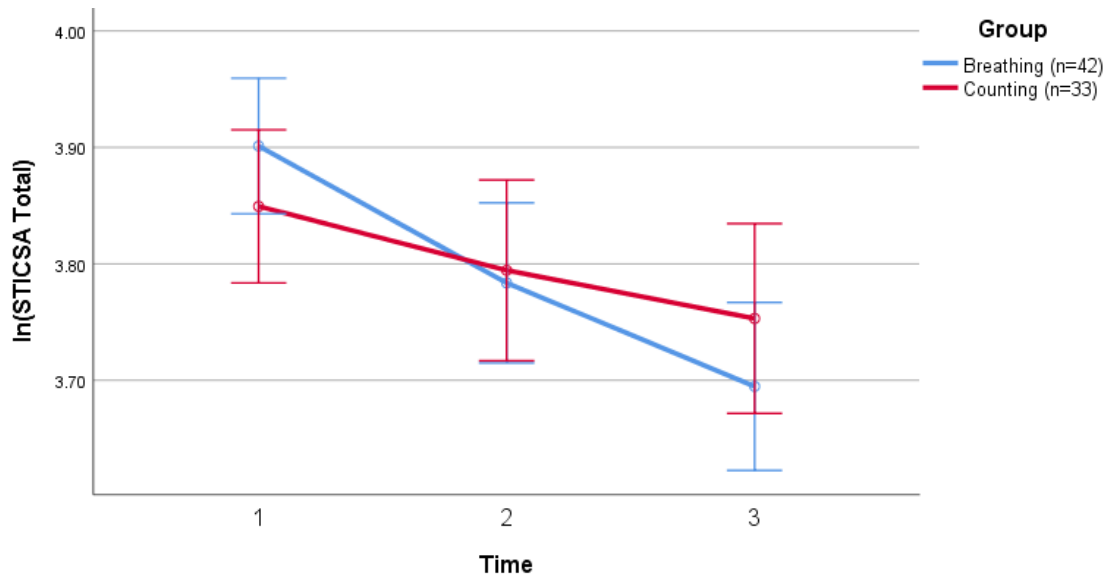


Note: Covariates appearing in the model were age and sex. There was no significant interaction between time and Group ( $F(2, 55) = 1.750, p = .183, \eta^2 = .06$ ).

### 3.2.1.3. Mean change in trait anxiety between the Breathing and Counting group after 8 weeks.

Data were analyzed using a mixed-design Repeated Measures ANOVA with a within-subjects factor of time (baseline, 4 weeks, 8 weeks) and a between-subject factor of Group (Breathing and Counting; covariates: intervention adherence, data cohort, age, and sex). Levene's test indicated that the error variance of the log-transformed total anxiety scores were equal across groups. Mauchly's test indicated that the assumption of sphericity had been violated ( $\chi^2(2) = 7.798, p = .020$ ), therefore degrees of freedom were corrected using the Huynh-Feldt estimates of sphericity ( $\epsilon = 0.992$ ). There was no significant effect of time ( $F(2, 137) = 2.511, p = 0.085$ ). There was no significant effect of group ( $F(1,69) = .019, p = .891$ ). There was no significant trend in the interaction between time and Group ( $F(2, 138) = 2.511, p = .085, \eta^2 = .035$ ). As the interaction was at  $p < .10$ , follow-up analyses were performed on STICSA subscales of Cognitive Anxiety and Somatic Anxiety. Results are visualized in Figure 6. Statistical output for analyses within Section 3.2.1.3. can be found in Appendix I.

**Figure 6 Estimated Marginal Means of a Repeated Measures ANOVA for Log Transformed STICSA Total**



Note: Covariates appearing in the model include self reported app usage, study cohort, age, and sex. Error bars represent 95% confidence intervals.

**Follow-up Analyses with STICSA subscales: cognitive and somatic (Bonferroni corrected for the 2 analyses being performed)**

Follow-up analyses investigated the STICSA subscale constructs of Cognitive Anxiety and Somatic Anxiety. Statistical significance was set to  $\alpha < .025$  to control for multiple comparisons. Mauchly’s test indicated no violations of the sphericity assumption for log transformed Cognitive Anxiety, but it did for Somatic Anxiety. A Hyun-Feldt correction was used for Somatic Anxiety analysis. For log transformed Cognitive Anxiety, the interaction between time and Group was not significant at  $\alpha < .025$ : ( $F(2, 138) = 3.207, p = .044, \eta^2 = .044$ ). There was no significant interaction between time and Group for log transformed Somatic Anxiety ( $F(2, 138) = 0.970, p = .382, \eta^2 = .014$ ).

**3.2.1.4. Mean change in trait anxiety in the Waitlist Control group over 8 weeks.**

Data were analyzed using a Repeated Measures ANOVA controlling for age and sex with a within-subjects factor of time (baseline, 4 weeks, 8 weeks) and no between-subject factor. There was no overall significant main effect of time on the log-transformed

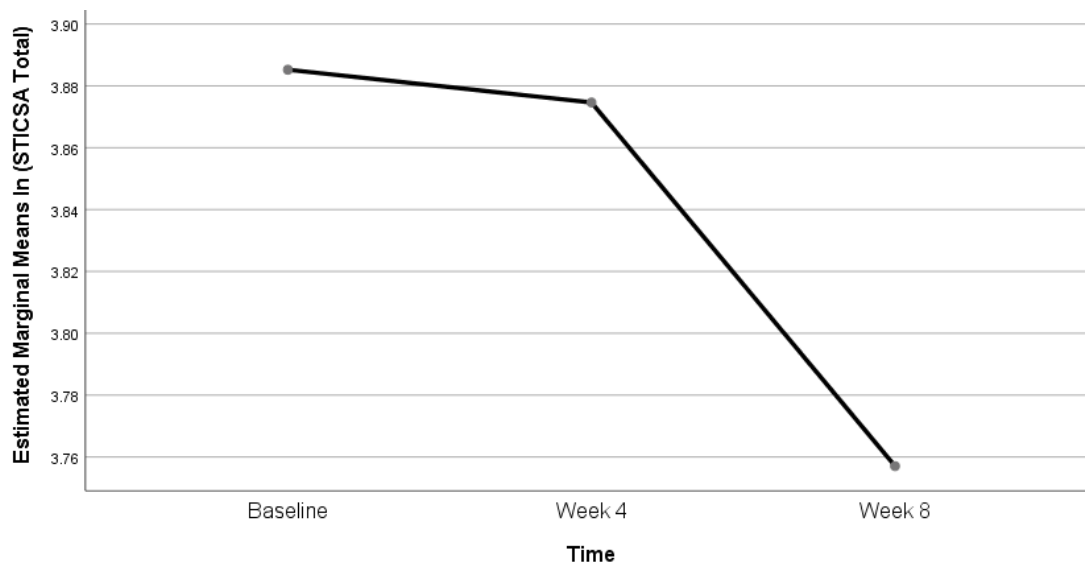
self-reported total anxiety scores ( $F(2, 34) = 2.005, p = .150, \eta^2 = .106$ ). Follow up pairwise comparisons (after Bonferroni correction) indicated a significant difference between Week 4 and Week 8 (Mean Difference = 0.118,  $p = .016$ ), but no difference between Baseline and Week 4 (Mean Difference = 0.011,  $p = 0.800$ ). Data is visualized in Figure 7. All statistical output for analyses within Section 3.2.1.4. can be found in Appendix J.

**Table 8** Pairwise Comparisons of Time Points Within the Waitlist Control Group

Time		Mean Difference	Std. Error	Sig.	95% Confidence Interval for Difference	
					Lower Bound	Upper Bound
Baseline	Week 4	0.011	0.044	0.800	-0.081	0.104
Week 4	Week 8	.118	0.044	0.016	0.024	0.211

Note: Based on estimated marginal means. Significance level set at  $\alpha = 0.025$  to adjust for multiple comparisons.

**Figure 7** Estimated Marginal Means of a Repeated Measures ANOVA for Log Transformed STICSA Total within the Waitlist Control Group



Note: Covariates appearing in the model included Age and Sex. Sample size  $n=20$ .

Post-hoc tests of STICSA subscales of Cognitive and Somatic Anxiety were performed. There was no overall significant main effect of time on the log-transformed self-reported Cognitive Anxiety scores ( $F(2, 32) = 2.063, p = .143, \eta^2 = .108$ ) or Somatic Anxiety scores ( $F(2, 32) = 1.237, p = .303, \eta^2 = .068$ ). Follow up pairwise comparisons

with an adjusted  $\alpha=0.025$  to control for multiple comparisons, indicated no significant difference between Week 4 and Week 8 in Cognitive Anxiety (Mean Difference = 0.121,  $p = .030$ ), or Somatic Anxiety (Mean Difference = 0.117,  $p = 0.034$ ).

### **3.2.1.5. Trait anxiety over 4 weeks in a subgroup of clinical severity**

In combining the Breathing group ( $n = 45$ ) and the WLC group ( $n = 23$ ), the combined sample had an average STICSA Total Anxiety score of 49.85 (SD = 10.5). Of the Breathing group, 75.6% ( $n = 34$ ) of the group had a STICSA Total score > 42 at Baseline. Of the WLC group, 60.9% ( $n = 13$ ) of the group had a STICSA Total score > 42 at Week 4. In creating the High Anxiety group by utilizing a cut-off of STICSA Total score > 42, the new group ( $n = 47$ ) had a STICSA Total average score of 54.46 (SD = 9.08, range 42-75). After 4 weeks of performing the Breathing technique, the High Anxiety group had a mean score of 47.44 (SD = 10.89, range 25-70).

As this group was nonrandomly created utilizing a minimum cut-off of STICSA (e.g., Total Anxiety > 42), the baseline distribution of the STICSA Total scores was nonnormally distributed at the first time-point according to a Shapiro-Wilks test ( $W = .91$ ,  $p = 0.002$ ). Log transformation of the STICSA Total score had no effect on the distribution ( $W = .926$ ,  $p=0.005$ ). To accommodate the non-normal distribution, the nonparametric Wilcoxon Signed Ranks test was performed.

Of the 47 participants in the High Anxiety subgroup, 32 participants reported decreased STICSA Total Anxiety scores after four weeks, 11 participants reported increased anxiety scores after four weeks, and four participants had no change. A Wilcoxon signed rank-test indicated that this difference between participants with an increased STICSA Total Anxiety score compared to a decreased score was significant at  $p<0.001$ , ( $Z = -3.874$   $p = 0.000$ ).

All statistical output for analyses within Section 3.2.1.5. can be found in Appendix K.

### **3.2.2. Aim 2: Acceptability of the Ultra-brief Interventions**

Treatment Acceptability was measured via self-report at the time of learning the technique, and via self-report at study termination. Generally, both the Counting group and the Breathing group identified minimal barriers to treatment acceptance upon

learning their intervention techniques, with over 89% of both groups disagreeing with the statements “This technique will require a lot of my time for me to accomplish” and “The technique seems complex to me”. Over 80% of both groups disagreed with the statement: “Following this technique will interrupt my daily routines”. See all response distributions in Appendix L.

Potential between group differences of a summated total score of initial perceived technique acceptability were investigated. Summated treatment acceptability scores were assessed for normality using the Shapiro-Wilks test. Scores were non-normally distributed in both groups ( $W > 0.75$ ,  $p < .0001$ ). The nonparametric Mann-Whitney U Test was applied to investigate between group differences, showing no significant difference ( $U = 812.5$ ,  $p = 0.333$ ) between the Breathing group ( $n=46$ ) and the Counting Group ( $n=40$ ) of treatment acceptability upon initially learning their respective technique.

**Table 9 Treatment Acceptability Mann-Whitney U Ranking Statistics of Group Differences**

	GROUP	N	Mean Rank	Sum of Ranks
Initial Treatment Acceptance	Breathing	46	41.16	1893.50
	Counting	40	46.19	1847.50
	Total	86		

Note: Man Whitney U test statistic = 812.50,  $p = 0.33$

Treatment Satisfaction was measured via self-report at study termination after eight weeks. Over 95% of both groups agreed that “the intervention was simple” and “the intervention was easy to perform”. When asked if “it was difficult to remember to perform the intervention”, 68% of the Breathing group disagreed, while 78% of the Counting group disagreed. Similarly, 68% of the Breathing group agreed that they “could do the intervention anywhere”, while 78% of the Counting group agreed. 20% of the Breathing group disagreed with that statement, compared to 10% of the counting group. When asked about general satisfaction with the technique, 65% of the Breathing group agreed that they were satisfied with the intervention, and similarly 64% indicated they would recommend the intervention to a friend. In the Counting group, 55% agreed that they were satisfied with the intervention, and 55% indicated they would recommend the intervention to a friend.

Potential between group differences of a summated total score of Treatment Satisfaction was investigated. The Breathing Group (n=41) had a mean Treatment Satisfaction score of 23.3 (SD=3.4) while the Counting group (n=33) had a mean score of 22.3 (SD=3.7). Levene's test suggested equal variances between groups could be assumed (F=0.025, p=.874). A Student's t-test found no significant differences between the groups  $t(72) = 1.153$ ,  $p=.253$ )

**Table 10 Post Treatment Satisfaction Independent Samples t-test**

	t	df	p	Mean Difference	Std. Error Difference	95% Confidence Interval	
						Lower	Upper
Post-Treatment Satisfaction	1.153	72	0.253	0.95344	0.82708	-0.6953	2.6021

Note: Group comparisons between mean summated post-treatment satisfaction summated scores of the Breathing group (n=41) and the Counting group (n=33)

### 3.2.3. Aim 3: Relationship of Adherence to Practice on Treatment Efficacy

Treatment Adherence was assessed both objectively through the mobile application, which tracked the number of discreet times the app was used in the first four weeks of the study, and subjectively by asking participants to report how frequently they used the app after four weeks and after eight weeks.

#### 3.2.3.1. Mobile Application Tracked Technique Frequency

Mobile app-collected data was only available for the first four weeks of the study. Analyzing app-collected data, the Counting group on average used the mobile app to perform the technique 48% of the instructed times (n=36, SD=.31) ranging from 1% to 100%, while the Breathing group used the mobile app to perform the technique 38.9% of the instructed times (n=45, SD=.27) ranging from 4% to 90%. A Shapiro-Wilks test determined that the distribution of the app-collected data within both the Breathing and the Counting group were non-normally distributed ( $W < .91$ ,  $p < .01$ ). The nonparametric Mann-Whitney U Test was performed to assess group differences in mean app-collected adherence.

The nonparametric Mann-Whitney U test was performed to evaluate whether app-collected adherence differed by group. The results indicated that there was no



significant difference between the app-collected adherence between groups ( $U = 660.00$ ,  $p = .154$ ).

**Table 11 Objective App Usage at 4 Weeks Mann Whitney U Rankings**

	Group	n	Mean Rank	Sum of Ranks
Objective App Usage	Breathing	45	37.67	1695.00
	Counting	36	45.17	1626.00
	Total	81		

### 3.2.3.2. Self-reported Technique Frequency

Self-report of app usage found that 71.1% of the Breathing group reported using the app at least once a day both after four weeks and eight weeks in the study. 11.1% of the Breathing group reported using the app less than three days a week after four weeks, which increased to 15.5% after eight weeks. In the Counting group, 75.0% reported using the app at least once a day for the first four weeks, which decreased to 61.1% after eight weeks. 8.3% of the Counting group reported using the app less than three days a week after four weeks, which increased to 19.4% after eight weeks. No participants in either group reported using the app more than twice a day.

**Table 12 Self-Reported App Frequency of Use by Group**

	Breathing (n=45)		Counting (n=36)	
	4 Weeks	8 Weeks	4 Weeks	8 Weeks
Twice a day	48.9%	44.4%	61.1%	47.2%
Once a day	22.2%	26.7%	13.9%	13.9%
4-6 times a week	17.8%	13.3%	16.7%	19.4%
2-3 times a week	4.4%	6.7%	8.3%	13.9%
Once a week	2.2%	6.7%	0.0%	2.8%
I dont use the app	4.4%	2.2%	0.0%	2.8%

A Wilcoxon matched-pairs signed-rank test indicated that the number of participants that reported a decrease in app usage compared to an increase in app usage was not significant in the Breathing group ( $Z = -.689$ ,  $p = .491$ ), but was significant

in the Counting group (-2.933,  $p=0.003$ ). In the counting group, there was a significantly larger number of participants who's app usage decreased from Week 4 to Week 8 relative to those who increased.

**Table 13 Treatment Adherence Wilcoxon Matched-Pairs Signed Rank Test**

		N	Mean Rank	Sum of Ranks
Breathing (n=45)	Frequency <b>decreased</b> at 8 Weeks vs 4 Weeks	12	10.25	123.00
	Frequency <b>increased</b> at 8 weeks vs 4 weeks	8	10.88	87.00
	Frequency remained the same	25		
Counting (n=36)	Frequency <b>decreased</b> at 8 Weeks vs 4 Weeks	13	8.46	110.00
	Frequency <b>increased</b> at 8 weeks vs 4 weeks	2	5.00	10.00
	Frequency remained the same	21		

A spearman's correlation between objective and self-report app usage found a moderate correlation between the subjective and objective measures after four weeks that was statistically significant at  $r(81)=.53$ ,  $p<.0001$ . The nonparametric Mann-Whitney U test indicated there was no difference in objective app usage between the Breathing group and the Counting group after four weeks ( $U=660.0$ ,  $p=0.154$ ). Objective data was not available for the eight-week time point.

### **3.2.3.3. Correlation between Treatment Adherence and Treatment Efficacy**

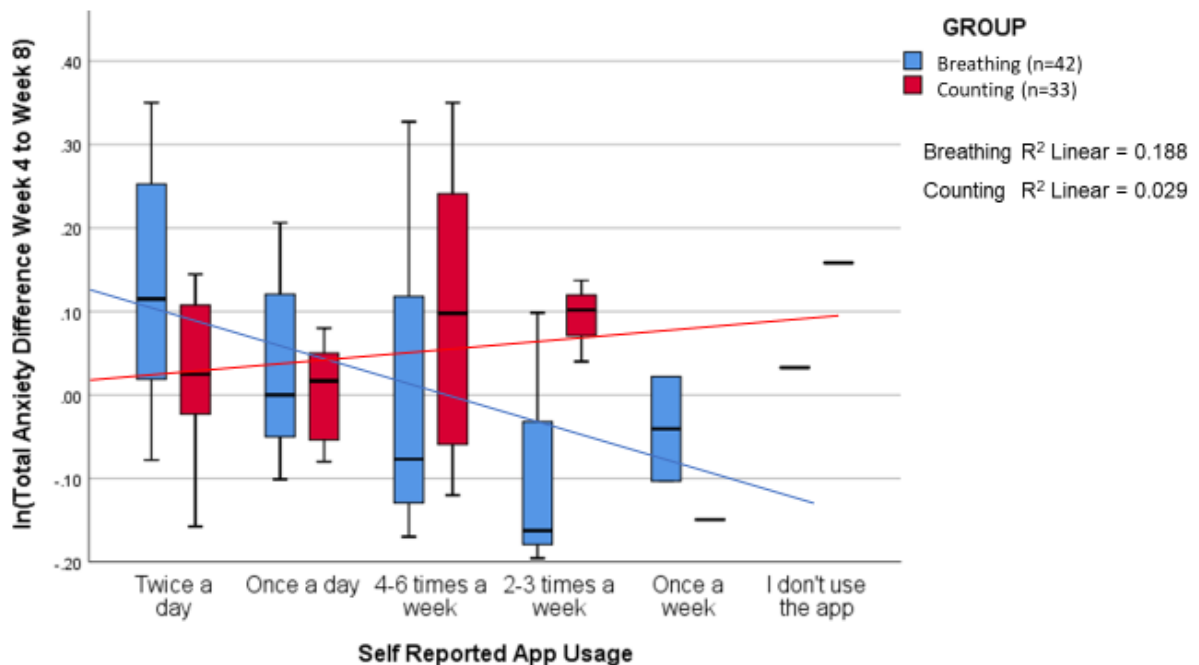
The relationship between Treatment Adherence (self-report app usage) and Treatment Efficacy (change in log-transformed self-reported Total Anxiety scores) was analyzed independently for each experimental group (Breathing group and Counting group) using a Pearson's correlation.

Correlations were analyzed between self-report app usage at Week 4 and the change in log-transformed Total Anxiety scores over the first four weeks of the study (Baseline – Week 4). Among the Breathing group, the reported frequency of technique use at Week 4 and the change in Total Anxiety scores were not significantly correlated  $r(45) = .089$ ,  $p=.563$ . Among the Counting group, the reported frequency of technique use

at Week 4 and the change in Total Anxiety scores were not significantly correlated  $r(36) = -.141, p=.412$ .

Correlations were analyzed between self-report app usage at Week 8 and the change in Windsorized log-transformed Total Anxiety scores over the second four weeks of the study (Week 4 – Week 8). Results are displayed in Figure 8. Among the Breathing group, the reported frequency of technique use at Week 8 and the change in Total Anxiety scores were significantly positively correlated  $r(42) = .433, p=.004$ . Among the Counting group, the reported frequency of technique use at Week 8 and the change in total anxiety scores were not correlated  $r(33) = -.169, p=.346$ .

**Figure 8 Change in Total Anxiety relative to Self-Report App Usage at Week 8**



### Treatment Adherence Follow-up Analyses with STICSA subscales

Follow-up analyses investigated the STICSA subscale constructs of Cognitive Anxiety and Somatic Anxiety within the Breathing group from Week 4 to Week 8. Statistical significance was set to  $\alpha < .025$  to control for multiple comparisons. The reported frequency of technique use at Week 8 was significantly positively correlated to both the change in Cognitive Anxiety scores ( $r(42) = .368, p=.016$ ) and Somatic Anxiety scores ( $r(42) = .392, p=.010$ ).

### 3.3. Supplementary Analysis

#### 3.3.1. Adequate Adherence Subgroup

A subgroup of participants was identified that did not sufficiently adhere to performing the designated technique. Of the 81 participants assigned to an experimental group, six participants (n=3 Breathing group, n=3 Counting group) reported performing their designated technique three times per week or less in both the Week 4 and Week 8 follow-up. As these participants did not adequately engage with the intervention at either time point, they were excluded in these supplementary analyses to determine treatment efficacy for those who performed the intervention.

Demographic information and outcome variables at baseline, Week 4, and Week 8 for the subgroup who sufficiently adhered to the assigned intervention (AA) are presented in Table 14.

**Table 14 Descriptive Statistics of AA Subgroup**

##### **BASELINE: ADEQUATE ADHERENCE SUBGROUP**

	<u>Breathing (n=42)</u>		<u>Counting (n=33)</u>	
Sex (% Female)	90.5%		87.9%	
First Language (% English)	83.3%		78.8%	
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range
Age	18.67 (1.53)	17-25	19.00 (1.32)	17-23
PANAS Pos	26.00 (7.79)	14-46	27.48 (5.32)	16-37
PANAS Neg	17.05 (5.61)	10-36	17.85 (6.83)	10-35
STICSA Total	50.40 (9.84)	37-75	47.58 (9.33)	36-67
STICSA Som	22.57 (7.02)	13-44	21.39 (5.32)	12-33
STICSA Cog	27.83 (4.92)	19-38	26.18 (5.33)	17-39

##### **WEEK 4 FOLLOW-UP: ADEQUATE ADHERENCE SUBGROUP**

	<u>Breathing (n=42)</u>		<u>Counting (n=33)</u>	
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range
PANAS Pos	24.40 (8.41)	10-44	25.06 (6.65)	10-37
PANAS Neg	16.38 (5.50)	9-32	16.64 (6.10)	10-33
STICSA Total	44.43 (9.55)	27-64	44.85 (8.71)	31-60
STICSA Som	20.07 (4.99)	12-35	20.03 (5.15)	11-29

STICSA Cog	24.36 (6.36)	13-38	24.82 (4.73)	16-34
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**WEEK 8 FOLLOW-UP: ADEQUATE ADHERENCE SUBGROUP**

	<u>Breathing (n=40)</u>		<u>Counting (n=32)</u>	
	<i>M (SD)</i>	Range	<i>M (SD)</i>	Range
PANAS Pos	25.85 (8.40)	10-43	23.41 (7.36)	11-39
PANAS Neg	15.68 (6.05)	9-36	17.50 (7.16)	10-35
STICSA Total	40.70 (9.26)	24-61	43.59 (9.00)	28-61
STICSA Som	18.12 (4.65)	11-31	18.97 (5.23)	11-28
STICSA Cog	22.57 (6.21)	11-37	24.62 (4.63)	16-33

Descriptive statistics for demographic and clinical variables, including mean (M), standard deviation, and range for the group level samples of participants who reported adequate adherence to performing the technique (at least 3 days a week or more).

**3.3.1.1. AA Mean trait anxiety after four weeks between Breathing, Counting, Waitlist Control groups**

Analyses within the AA subgroup compared the Breathing group (n=42), Counting group (n=33) and the WLC (n=23) from Baseline to Week 4. A repeated measures ANOVA, controlling for age and sex, determined there was no significant interaction between time and Group ( $F(2, 93) = 2.484, p = 0.089, \eta^2 = 0.051$ ), indicating the change in total anxiety scores over the four week time period was not significantly different between groups. All statistical output for analyses within Section 3.3.1.1. can be found in Appendix M.

**Follow-up Analyses with STICSA subscales of Adequate Adherence subgroup:**

Follow-up analyses investigated the STICSA subscale constructs of Cognitive Anxiety and Somatic Anxiety. Statistical significance was set to  $\alpha < .025$  to control for multiple comparisons. For log transformed Cognitive Anxiety, there was a medium effect size, but no significant interaction between time and Group using an  $\alpha < .025$ : ( $F(2, 93) = 3.295, p = .041, \eta^2 = .066$ ), indicating no change in cognitive anxiety scores over time dependent on which group a participant was in. There was no significant interaction between time and Group for log transformed Somatic Anxiety ( $F(2, 93) = 1.083, p = .343, \eta^2 = .023$ ).

### **3.3.1.2. AA: Physiological changes after 4 weeks between Breathing, Counting, Waitlist Control group**

Analyses within the AA subgroup compared the Breathing group (n=20), the WLC (n=23) and the counting group (n=13). All statistical output for analyses within Section 3.3.1.2. can be found in Appendix N.

#### ***Systolic Blood Pressure (AA)***

SBP outcomes were normally distributed within both groups (Shapiro-Wilks  $p > .05$ ). A repeated measures ANOVA controlling for the effect of age and sex determined there was no significant interaction between time and Group ( $F(2, 51) = 2.039, p = .141, \eta^2 = .074$ ), indicating the change in systolic blood pressure over time was not different between groups.

#### ***Diastolic Blood Pressure (AA)***

DBP outcomes were normally distributed within both groups (Shapiro-Wilks  $p > .05$ ). A repeated measures ANOVA controlling for the effect of age and sex determined there was no significant interaction between time and Group ( $F(2, 51) = 0.094, p = .910, \eta^2 = .004$ ), indicating the change in systolic blood pressure over time was not different between groups.

#### ***RMSSD (AA)***

The Shapiro-Wilk Test found distributions of RMSSD outcomes were non-normally distributed. Log transformation of RMSSD outcomes did not achieve normality. To accommodate the non-normal distribution, the nonparametric Wilcoxon Signed Ranks test was performed separately for each of the three groups. A Bonferroni correction for the three comparisons established the  $\alpha = 0.017$  for significance testing.

In the Breathing group five participant's RMSSD outcomes decreased from Baseline to Week 4 and 15 participant's RMSSD outcomes increased from Baseline to Week 4. A Wilcoxon signed rank-test indicated that this difference between participants with an increased RMSSD compared to a decreased RMSSD was not significant at the predetermined  $\alpha = 0.017$  ( $Z = -2.203, p = .028$ ).

In the Counting group, eight participants RMSSD outcomes decreased from Baseline to Week 4 and five participant's RMSSD outcomes increased from Baseline to Week 4. A Wilcoxon signed rank-test indicated that this difference was not statistically significant, ( $Z = -0.524$   $p = .600$ ). In the WLC group, 11 participants RMSSD outcomes decreased from Baseline to Week 4 and 12 participant's RMSSD outcomes increased from Baseline to Week 4. A Wilcoxon signed rank-test indicated that this difference was not statistically significant, ( $Z = -0.426$   $p = .670$ ).

### ***SDNN (AA)***

Distributions of SDNN outcomes were found to be non-normally distributed (Shapiro-Wilks  $p < .05$ ). The data was Log transformed to obtain normal distributions within groups. A repeated measures ANOVA controlling for the effect of age and sex determined there was a medium effect, but no significant interaction between time and Group ( $F(2, 51) = 2.026$ ,  $p = .142$ ,  $\eta^2 = .074$ ).

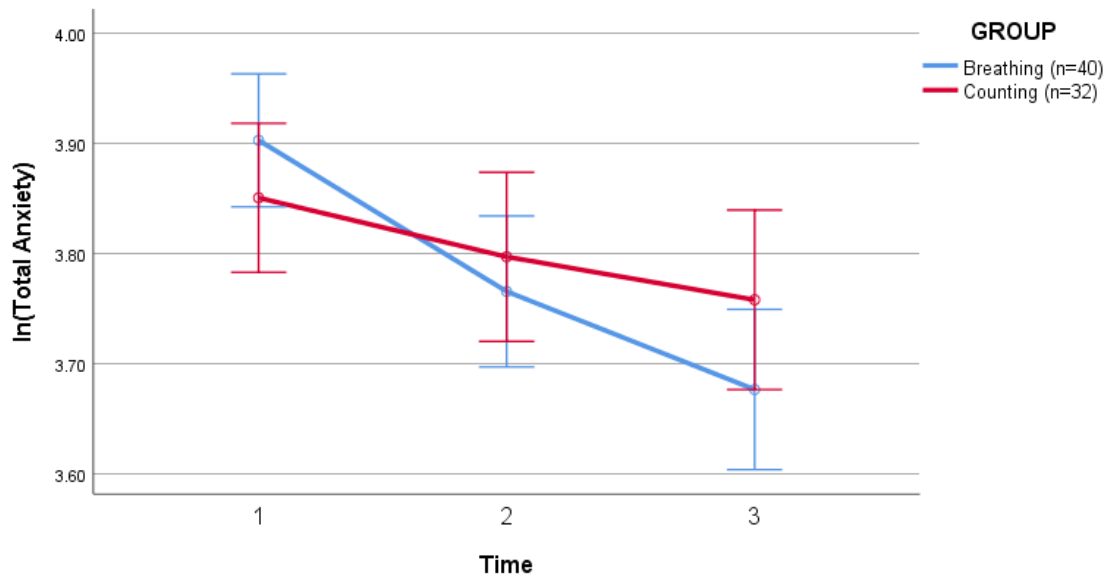
### **3.3.1.3. AA: Trait anxiety after 8 weeks between the Breathing and Counting groups**

Analyses within the AA subgroup compared the Breathing group ( $n=40$ ) and the Counting group ( $n=32$ ) from Baseline to Week 8. A Shapiro Wilks test confirmed that all outcome variables of log transformed STICSA Total and subscales were  $p > .05$ . Data were analysed using a mixed-design Repeated Measures ANOVA with a within-subjects factor of time (baseline, 4 weeks, 8 weeks) and a between-subject factor of Group (Breathing and Counting; covariates: intervention adherence, data cohort, age, and sex). Levene's test indicated that the error variance of the log-transformed total anxiety scores were equal across groups. Mauchly's test indicated that the assumption of sphericity was met ( $p>0.05$ ). Upon analyzing Total Anxiety, there was a significant interaction between Time and Group ( $F(2, 132) = 3.722$ ,  $p = .027$ ,  $\eta^2 = .053$ ), indicating a greater decrease in Total Anxiety scores over time in the Breathing group compared to the Counting group, as visualized in Figure 9. Follow-up analyses were performed for the Cognitive and Somatic subscales of the STICSA, using a corrected alpha = .025 to control for multiple comparisons. Repeated Measures ANOVA of the Cognitive Anxiety subscale yielded a significant interaction of time and Group ( $F(2, 132) = 5.571$ ,  $p = .005$ ,  $\eta^2 = .078$ ) while the Somatic Anxiety subscale indicated no significant interaction ( $F(2, 132) = 1.023$ ,  $p = .362$ ,  $\eta^2 = .015$ ). Participants in the Breathing group who completed the eight week intervention

and adhered to the instructed treatment, experienced significant decreases in self-reported trait Cognitive Anxiety (see Figure 10), but not Somatic Anxiety (see Figure 11). There was a medium-large effect of the intervention in these treatment adhering participants.

All statistical output for analyses within Section 3.3.1.3. can be found in Appendix O.

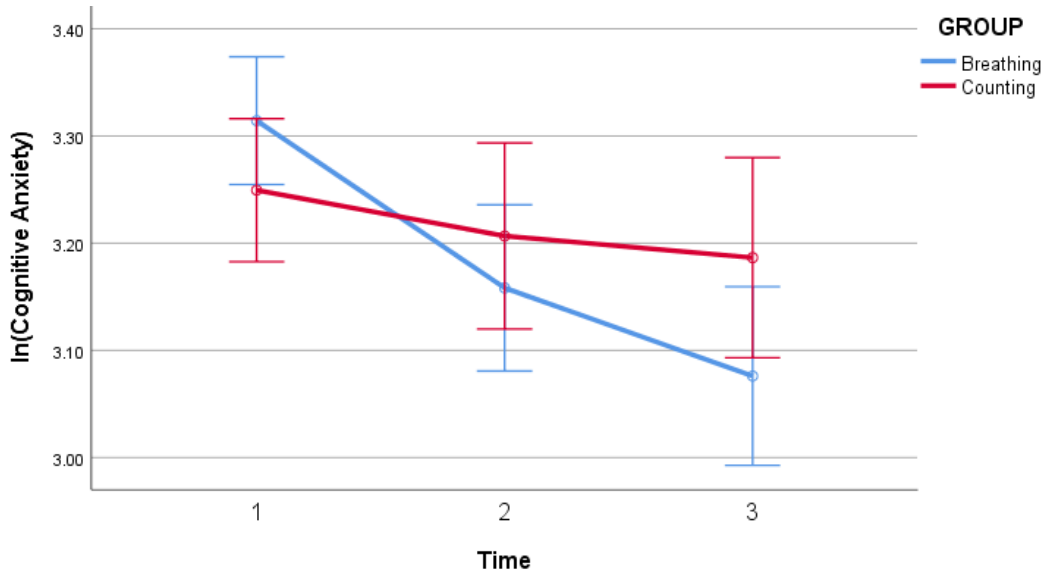
**Figure 9 AA Subgroup In(Total Anxiety), 2 Groups over 8 Weeks**



Covariates appearing in the model are evaluated at: Cohort = 1.5, Self Reported App Usage = 2.9, Age = 18.9, Sex = 1.9 Error bars: 95% CI

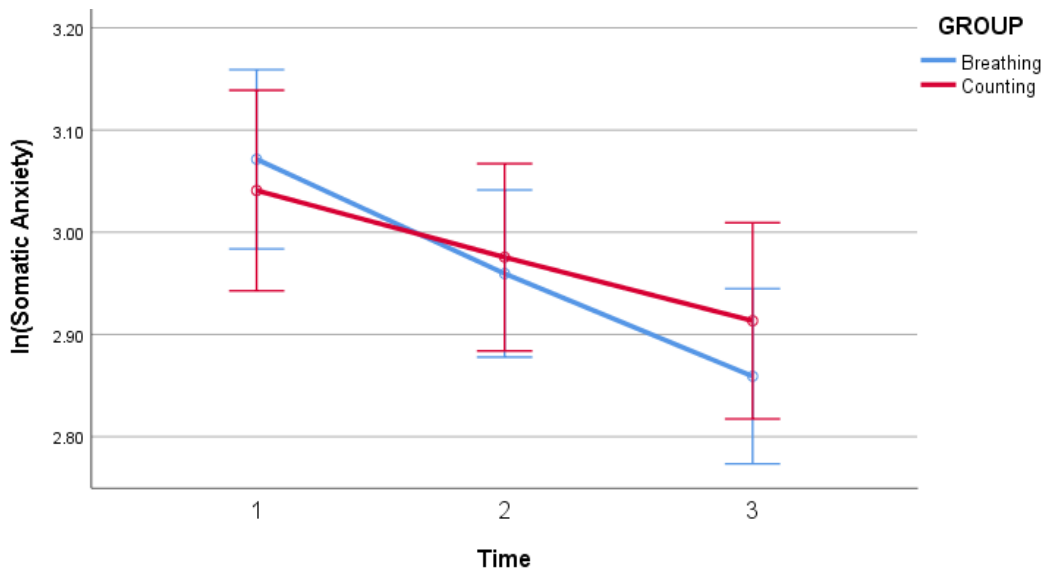


**Figure 10 AA Subgroup In(Cognitive Anxiety) 2 groups over 8 weeks**



Covariates appearing in the model are evaluated at: Self Reported App Usage= 2.9, Cohort = 1.6, Sex = 1.9, Age = 18.9 Error bars: 95% CI

**Figure 11 AA Subgroup In(Somatic Anxiety) 2 groups over 8 weeks**



Covariates appearing in the model are evaluated at: Self Reported App Usage = 2.9, Cohort = 1.6, Sex = 1.9, Age = 18.9, Error bars: 95% CI

### 3.3.1.4. AA: Trait anxiety within the Waitlist Control group over 8 weeks

Of the Waitlist Control group (n=20), 19 participants reported adequate technique adherence and were thus included in the AA subgroup. Data were analyzed using a Repeated Measures ANOVA controlling for age and sex with a within-subjects factor of time (baseline, 4 weeks, 8 weeks) and no between-subject factor. There was no overall significant main effect of time on the log-transformed self-reported Total Anxiety scores ( $F(2, 32) = 1.784, p = .184, \eta^2 = .100$ ). Follow up pairwise comparisons with an  $\alpha=0.025$  for multiple comparisons, indicated a significant difference between Week 4 and Week 8 (Mean Difference = 0.125,  $p = .016$ ), but no difference between Baseline and Week 4 (Mean Difference = 0.013,  $p = 0.776$ ).

All statistical output for analyses within Section 3.3.1.4. can be found in Appendix P.

**Table 15 AA Subgroup: Pairwise Comparisons within WLC by Time Point**

Time		Mean Difference	Std. Error	Sig.	95% Confidence Interval for Difference	
					Lower Bound	Upper Bound
Baseline	Week 4	0.013	0.046	0.776	-0.085	0.112
Week 4	Week 8	.125*	0.046	0.016	0.027	0.222

Based on estimated marginal means. \* refers to the mean difference is significant at the .025 level in adjustment for multiple comparisons.

Post-hoc tests of STICSA subscales of Cognitive and Somatic anxiety were performed. There was no overall significant main effect of time on the log-transformed self-reported Cognitive Anxiety scores ( $F(2, 32) = 1.843, p = .175, \eta^2 = .103$ ) or Somatic Anxiety scores ( $F(2, 32) = 1.118, p = .339, \eta^2 = .065$ ). Follow up pairwise comparisons with an adjusted  $\alpha=0.025$  to control for multiple comparisons, indicated no significant difference between Week 4 and Week 8 in Cognitive Anxiety (Mean Difference = 0.126,  $p = .032$ ), or Somatic anxiety (Mean Difference = 0.126,  $p = 0.028$ ).

**Table 16 AA Subgroup: Cognitive Anxiety Pairwise Comparisons within WLC**

Time		Mean Difference	Std. Error	Sig.	95% Confidence Interval for Difference	
					Lower Bound	Upper Bound
Baseline	Week 4	0.017	0.048	0.719	-0.083	0.118

Week 4	Week 8	.126	0.054	0.032	0.012	0.240
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Based on estimated marginal means. \*The mean difference is significant at the .025 level in adjustment for multiple comparisons.

**Table 17 AA Subgroup: Somatic Anxiety Pairwise Comparisons within WLC**

Time		Mean Difference	Std. Error	Sig.	95% Confidence Interval for Difference	
					Lower Bound	Upper Bound
Baseline	Week 4	0.001	0.063	0.990	-0.135	0.133
Week 4	Week 8	.126	0.052	0.028	0.015	0.238

Based on estimated marginal means. \*The mean difference is significant at the .025 level in adjustment for multiple comparisons.

### 3.3.1.5. AA: Trait Anxiety after 4 weeks in a subgroup of clinically High Anxiety

Of the High Anxiety subgroup (n=47), 45 participants reported adequate technique adherence and were thus included in the AA subgroup. Normality for STICSA outcome measures of STICSA Total, Cognitive Subscale, and Somatic Subscale were assessed with a Shapiro-Wilks test. STICSA Cognitive subscales were normally distributed. Log-transformation resulted in normal distributions of the Somatic subscale outcomes, but non-normal distributions of the Total Anxiety score. All statistical output for analyses within Section 3.3.1.5. can be found in Appendix Q.

The AA subgroup STICSA Total score was analyzed with the nonparametric Wilcoxon Signed Ranks test. Of the 45 participants in the High Anxiety subgroup, 31 participants reported decreased STICSA Total Anxiety scores after four weeks, 10 participants reported increased anxiety scores after four weeks, and four participants had no change. A Wilcoxon signed rank-test indicated that this difference between participants with an increased STICSA Total Anxiety score compared to a decreased score was significant at  $p < 0.001$ , ( $Z = -3.963$   $p = 0.000$ ).

The AA subgroup of Cognitive subscale and log transformed Somatic subscale were analyzed using a Repeated Measures ANOVA controlling for adherence, cohort, age and sex with a within-subjects factor of time and no between-subject factor. There was a significant effect of estimated means over time in both the Cognitive (Wilks' Lambda = 0.625,  $F(1,40) = 24.008$ ,  $p < .001$ ,  $\eta^2 = .375$ ) and log transformed Somatic

(Wilks' Lambda = 0.744,  $F(1,40) = 13.728$ ,  $p = .001$ ,  $\eta^2 = .256$ ) outcomes, such that estimated means decreased from time 1 to time 2 in the AA subgroup of the High Anxiety subgroup.

### **3.3.2. Female Only Subgroup**

Of total study participants, men only accounted for 15% of the sample. When investigated by experimental group, men made up 21.7% of WLC ( $n=5$ ), 11.1% of Breathing group ( $n=5$ ), and 13.9% of Counting group ( $n=5$ ). When men are excluded from analysis, sample sizes are WLC  $n=18$ , Breathing  $n=40$ , Counting  $n=31$ . Exploratory analyses were completed for mean Trait anxiety between Breathing, Counting, and WLC over four weeks (section 3.2.1.1) and between only Breathing and Counting groups over eight weeks (section 3.2.1.3). Statistical results are reported in Appendices Q and R.

#### **3.3.2.1. Female Only: Mean change in trait anxiety between Breathing, Counting and Waitlist Control group after 4 weeks**

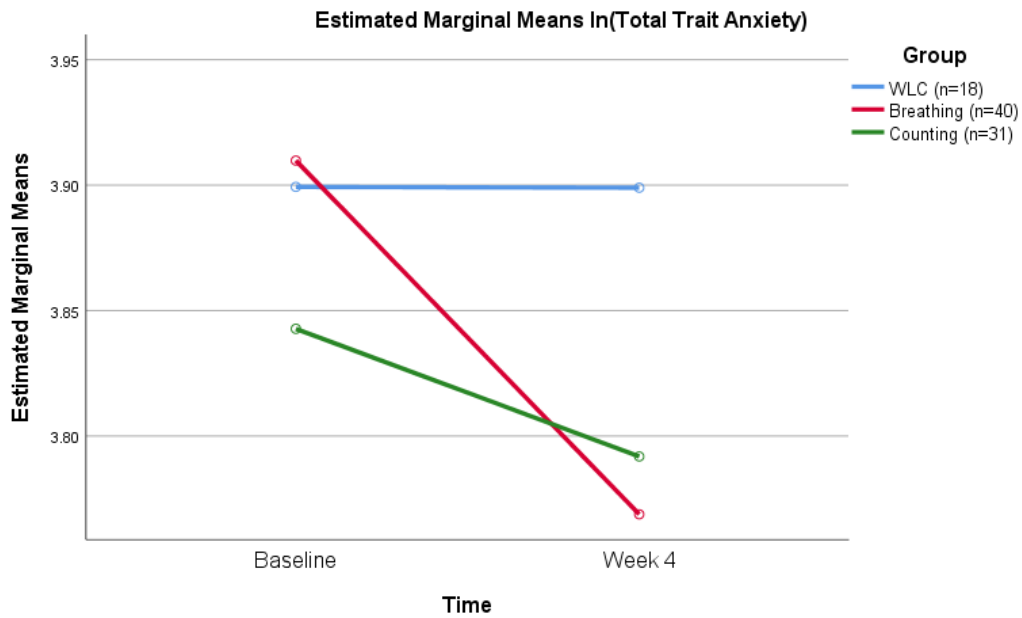
##### ***Self-reported Trait Anxiety***

A repeated measures ANOVA controlling for age determined there was a significant interaction between time and Group ( $F(2, 85) = 3.442$ ,  $p = .037$ ,  $\eta^2 = .075$ ), indicating the change in Total Anxiety scores over time was different between the three groups. This difference was also present in the subscale of Cognitive Anxiety ( $F(2, 85) = 4.232$ ,  $p = .018$ ,  $\eta^2 = .091$ ), but not in Somatic Anxiety ( $F(2, 85) = 1.689$ ,  $p = .191$ ,  $\eta^2 = .038$ ). Full analyses are reported in Appendix R, data is visualized in Figure 12.

##### ***Systolic Blood Pressure***

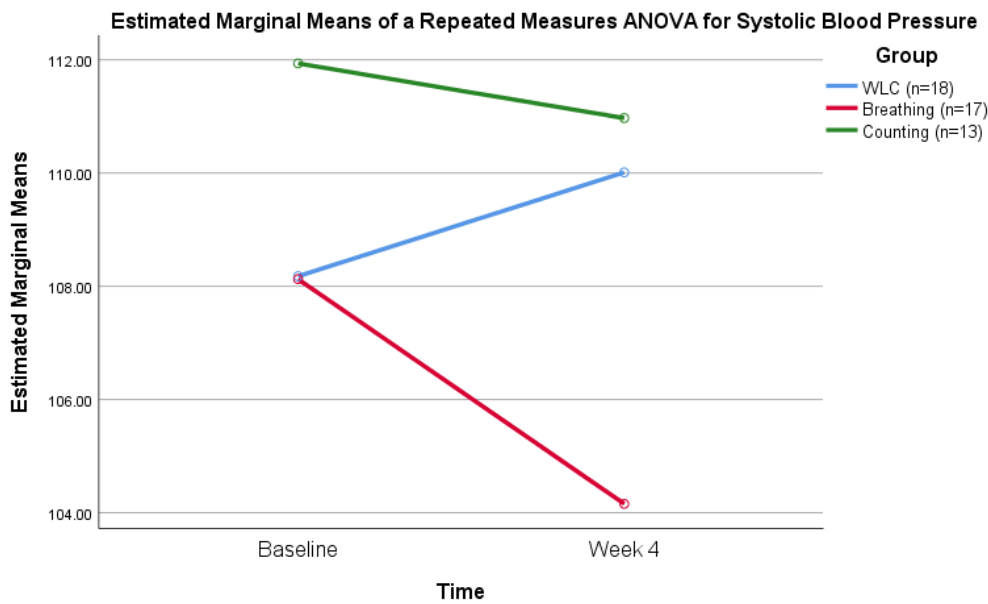
SBP outcomes were normally distributed within all groups (Shapiro-Wilks  $p > .05$ ). A repeated measures ANOVA controlling for the effect of age determined that the mean of SBP did not differ significantly between time points ( $F(1, 44) = 0.572$ ,  $p = .454$ ,  $\eta^2 = .013$ ). There was a significant interaction between time and Group ( $F(2, 44) = 4.268$ ,  $p = .020$ ,  $\eta^2 = .162$ ), indicating the change in resting systolic blood pressure over time was different between groups. Data is visualized in Figure 13.

**Figure 12 FEMALE In(Total Anxiety) 3 groups over 4 weeks**



Note: Covariate Age appeared in the model as: Age = 18.74. Time x Group interaction significant at p=0.037.

**Figure 13 FEMALE Systolic Blood Pressure 3 groups over 4 weeks**



Note: Covariate Age appeared in the model as: Age = 18.73. Time x Group interaction significant at p=0.02

### **Diastolic Blood Pressure**

DBP outcomes were normally distributed within all groups (Shapiro-Wilks  $p > .05$ ). A repeated measures ANOVA controlling for the effect of age and sex determined that the mean of DBP did not differ significantly between time points ( $F(1, 44) = 0.00$ ,  $p = .589$ ,  $\eta^2 = .00$ ). There was no significant interaction between time and Group ( $F(2, 44) = 0.481$ ,  $p = .622$ ,  $\eta^2 = .021$ ), indicating the change in DBP over time was not different between groups.

### **RMSSD**

The Shapiro-Wilk Test found distributions of RMSSD outcomes were non-normally distributed, despite data undergoing Windsorization and log-transformation. To accommodate the non-normal distribution, the nonparametric Wilcoxon Signed Ranks test was performed separately for each of the three groups. A Bonferroni correction for the three comparisons established the  $\alpha = 0.017$  for significance testing.

In the Breathing group four participant's RMSSD outcomes decreased from Baseline to Week 4 and 13 participant's RMSSD outcomes increased from Baseline to Week 4. A Wilcoxon signed rank-test indicated that this difference between participants with an increased RMSSD compared to a decreased RMSSD was not significant at the predetermined  $\alpha = 0.017$  ( $Z = -1.965$ ,  $p = .049$ ). In the Counting group, eight participants RMSSD outcomes decreased from Baseline to Week 4 and five participant's RMSSD outcomes increased from Baseline to Week 4. A Wilcoxon signed rank-test indicated that this difference was not statistically significant, ( $Z = -0.384$   $p = .701$ ). In the WLC group, nine participants RMSSD outcomes decreased from Baseline to Week 4 and nine participant's RMSSD outcomes increased from Baseline to Week 4. A Wilcoxon signed rank-test indicated that this difference was not statistically significant, ( $Z = -0.065$   $p = .948$ ).

### **SDNN**

To achieve normal distributions of SDNN outcomes (Shapiro-Wilks  $p > 0.05$ ), data was Windsorized and log-transformed. A repeated measures ANOVA controlling for the

effect of age determined there was no significant interaction between time and Group ( $F(2, 44) = .885, p = .420, \eta^2 = .039$ ) for SDNN outcomes.

### 3.3.2.2. Female Only: Mean change in trait anxiety between Breathing and Counting group after 8 weeks

Re-analysis of Section 3.2.1.3. found that a repeated measures ANOVA controlling for age, cohort, and frequency of app usage determined there was a significant interaction between time and Group ( $F(2, 122) = 3.486, p = .034, \eta^2 = .054$ ), indicating the change in total anxiety scores over time was different between the Breathing group and the Counting group. This difference was also present in the subscale of Cognitive Anxiety ( $F(2, 85) = 5.124, p = .007, \eta^2 = .077$ ), but not in Somatic Anxiety ( $F(2, 122) = 0.820, p = .443, \eta^2 = .013$ ). Full analyses are reported in Appendix T.

## **4. DISCUSSION**

This study examined the efficacy of regularly practicing an ultra-brief, one-minute breathing-based intervention on resting state physiological measures and self-reported measures of trait anxiety in university students self-reporting mild-to-severe anxiety-related symptoms. Despite the growing popularity of brief breathing techniques, there have been few reported empirical investigations on these ultra-brief interventions. Additionally, the effects of repeated practice of these interventions and the effect of technique adherence in these interventions is of interest. Understanding more about whether ultra brief breathing techniques are clinically efficacious and what influences that efficacy is important as brief interventions are highly accessible, low cost and easily self-administered.

The present study is seemingly the first to examine the clinical efficacy of regularly practicing an ultra-brief breathing technique over an extended time course (eight weeks), with control groups for comparison. This research is the first empirical study to examine the 4-7-8 breathing technique (Weil, 2000), a widely popularized ultra brief breathing technique with common anecdotal claims of promoting relaxation and reducing anxiety (Aim 1). Finally, this study assesses technique acceptance (Aim 2) and adherence to the technique (Aim 3) to increase understanding in factors related to treatment efficacy.

### **4.1. Summary of Major Findings**

In brief, there was no effect of the ultra-brief 4-7-8 Breathing technique on markers of trait anxiety compared to a counting technique and waitlist control in the analyses of the total sample. There were no group differences after four weeks in self-reported trait anxiety or in resting state physiological markers of stress and anxiety after four weeks. There were no group differences in self-reported trait anxiety after eight weeks.

However, in a subgroup of participants that sufficiently adhered to the technique (performing the technique four times or more per week), the group that performed the Breathing technique reported significantly decreased Total Trait Anxiety after eight weeks, but not four weeks, compared to participants performing a counting exercise of



identical duration. Upon follow-up subscale analyses, differences were specific to trait Cognitive Anxiety, not trait Somatic Anxiety. There were no differences in resting state physiological markers of stress and anxiety after four weeks.

Results indicated that both the Breathing and Counting groups perceived minimal barriers to integrating the technique into their daily life. There were no differences of initial perceived barriers between the two technique types. Assessment of treatment adherence found no difference in the average treatment adherence between the Breathing group and Counting group. Increased treatment adherence was found to be positively correlated with increased treatment effect only for the Breathing group, there was no correlation between treatment adherence and treatment efficacy in the Counting group.

#### **4.1.1. Four Week Follow-Up: Baseline to Week 4**

In the second set of analyses, a three-group design was utilized to control for the placebo effect over four weeks. The four-week waitlist control group was compared to two experimental groups: the ultra-brief breathing group and the ultra-brief counting group. Contrary to expectations, there were no differences in self-reported trait anxiety between the Breathing group, Counting group, and Waitlist Control after four weeks of instructed twice-daily practice in the total sample analyses. However, in the Female-only sub-analyses there was a significant effect of group over time as shown in Figure 12.

There were no statistically significant differences in any physiological resting state measures after four weeks, including measures of blood pressure (systolic and diastolic), and heart rate variability (RMSSD, SDNN). However, there was a nearly-significant effect of RMSSD values specific to the Breathing group, such that more individuals were measured to have an increase in RMSSD values from baseline to 4 weeks relative to those who had a decrease in RMSSD values. This effect was not seen in the Counting group or waitlist group. Additionally, in the sub-sample of the female-only analyses, there was a significant difference in systolic blood pressure between baseline and Week 4, with the Breathing group exhibiting a greater decrease over time than the other groups.

However, the mixed findings reported in this study differs from an investigation of device guided slow breathing performed for three minutes, five times a day (Wang et al., 2021). That investigation reported decreases in diastolic blood pressure after four weeks, which was maintained at eight weeks. However, they reported no treatment effect for systolic blood pressure, or the HRV measurements of RMSSD or SDNN. Our results showed a potential effect of the breathing technique on systolic blood pressure and RMSSD, but not diastolic blood pressure. The differences in these findings does not render confidence for these physiological findings.

The presence of a stronger physiological effect in the Breathing group than the other groups is congruent with physiological theory. Slow, paced breathing has been found to have an immediate effect on the cardio-respiratory system, (Lin et al., 2014; Van Diest et al., 2014). Blood pressure naturally varies, decreasing during inspiration and increasing during expiration (Billman, 2011). While the mechanism of a longer term change in resting state physiological measures is unknown, frequent practice of techniques that acutely effect these measures may have a long-lasting effect. Future research is indicated on this matter.

As data collection of physiological outcome variables was most effected by the Covid-19 changes to the study protocol, these analyses were the lowest powered in this study. Additionally, physiological variables have high day-to-day variability within an individual, making detection of a reliable difference statistically difficult. Thus, the results reported here should not be interpreted as conclusive, as the effect may have statistically emerged with a larger sample size, or disappeared altogether. The effect sizes that were observed after four weeks in this study, which may be due simply to random chance, are still less than the purposed medium-to-large effect sizes reported by traditional relaxation therapies (Manzoni et al., 2008). However, these findings may indicate that a larger sample size followed over the full eight weeks would be of interest.

#### **4.1.2. Eight Week Follow-Up: Baseline to Week 8**

In the third set of analyses, a two-group design was utilized to compare the two experimental groups over three time points spanning eight weeks. Among participants who performed their assigned ultra-brief relaxation technique at least four times per

week, there was a statistically significant interaction between time and group, such that changes in Trait Anxiety over the eight weeks were dependent on whether a participant was in the Breathing group or the Counting group. Participants in the Breathing group reported larger decreases in Trait Anxiety over the eight weeks, when controlling for the frequency of technique adherence, study cohort, age, and sex. The observed effect was specific to Cognitive Anxiety, and was not present in Somatic Anxiety. Changes in Trait Anxiety were clinically significant as well as statistically significant. Estimated marginal means indicated an average decrease of 10 points on the STICSA trait total scale across eight weeks, a difference large enough to move an individual from above a cut-off indicative of severe clinical anxiety, (STICSA Total >43; Van Dam et al., 2013) to below a cut-off indicative of mild anxiety, (STICSA Total=37; Van Dam et al., 2013).

This clear and large difference provides empirical merit for the claim that performing the 4-7-8 breathing technique regularly for eight weeks decreases trait anxiety. The results suggest that a significant effect of breathing only emerged after eight weeks of practice, but not at four. Of note, participants are instructed to increase their duration of practice to two minutes, twice a day from Week 4 to Week 8. This theoretically leads to twice the duration of practice in the second four weeks of the study. This increase in practice may in part explain why an effect emerged at 8 weeks but not at 4 weeks.

However, in analyzing the Female only subgroup, there was a significant effect of breathing that emerged over the other groups at both Week 4 and Week 8. This may indicate there is efficacy at Week 4, but this study was only powered enough to detect the stronger effect observed at Week 8. The eight-week treatment effect is similar to an investigation of a once daily 13-minute guided meditation that reported a medium-large effect size decrease in anxiety, mood disturbance, and fatigue at eight weeks, but not four weeks (Basso et al., 2019).

The specificity of the present findings to Cognitive Anxiety, but not Somatic Anxiety, also merits highlighting. Upon further examination of the results, both the Counting and Breathing groups decreased in trait Somatic Anxiety over time. While no group difference emerged, both groups reported less trait Somatic Anxiety over the course of the eight weeks. However, trait Cognitive Anxiety was only decreased in the

Breathing group. The counting technique seemed to have no effect on self-reported trait Cognitive Anxiety over time.

The cognitive dimension of anxiety reflects symptoms associated with thought processes, including worry, intrusive thoughts, and lack of concentration, while the somatic dimension includes symptoms such as hyperventilation, sweating, trembling, and palpitations (Ree et al., 2008). An intervention effect solely on the cognitive dimension was also reported in an investigation by McEvoy et al., (2017), which compared a control group to a progressive muscle relaxation and an attention training protocol. They reported both experimental conditions were associated with greater changes in cognitive, but not somatic, anxiety (McEvoy et al., 2017). Investigations by the developer of the STICSA also found differential predictive merits of the cognitive and somatic trait scales of the STICSA (Ree et al., 2008). They reported that cognitive trait scales were predictive of both cognitive and somatic state scores during a stress induction task, whereas somatic trait scales were not predictive of either cognitive or somatic state scores.

#### **4.1.3. Waitlist Control Group: Baseline to Week 8**

The Waitlist Control Group acted solely as a waitlist control for the first four weeks of the study before performing the breathing technique for one minute, twice a day for the second four weeks of the study. Consistent with expectations about the Waitlist control group, no change was observed in the first four weeks of the study while participants were in the waitlist period. However, there was a significant decrease in trait anxiety between Week 4 and Week 8, after participants had been taught and instructed to practice the breathing technique. These findings indicate that compared to performing no daily task, the same individuals reported decreased trait anxiety after four weeks of being instructed to perform the 4-7-8 breathing technique twice daily. While significant, care must be taken in interpreting this result as this may be the result of a placebo effect. Participants may have expected that performing the technique would lead to therapeutic benefit, and self-reported their trait anxiety after four weeks in accordance with this expectancy effect.

#### **4.1.4. Aim 2: Treatment Acceptance**

The results reported here indicated that both of the ultra-brief relaxation techniques had minimal perceived barriers to the techniques, both at the time of learning the technique and at study completion. As proposed by Scheel et al. (2004), perceived difficulty of treatment, specifically in the time, effort, and complexity of the treatment, are major determinants of treatment acceptability. In the present study, immediately after learning the techniques, over 90% of both groups disagreed with the statements “This technique will require a lot of my time for me to accomplish” and “The technique seems complex to me”. Over 80% of both groups disagreed with the statement: “Following this technique will interrupt my daily routines”.

At study completion, the vast majority of both groups (>95%) agreed the intervention was simple and easy to perform. The largest barriers the participants of this study identified were in remembering to perform the intervention and feeling comfortable performing the intervention anywhere. Despite low perceived barriers at the time of learning the treatment and at study completion, only 65% of the Breathing group and 55% of the Counting group reported that they were “satisfied with the intervention”. This relatively low rate of satisfaction indicates that while these ultra brief techniques might be useful tools for some individuals, they will not be found useful for everyone.

#### **4.1.5. Aim 3: Treatment Adherence**

Despite both techniques receiving high participant acceptance, treatment adherence remained relatively low in both groups. The Counting group on average used the mobile app to perform the technique 48% of the instructed times (approximately once per day), while the Breathing group used the mobile app to perform the technique 38% of the instructed times. A meta-analysis of treatment adherence of mindfulness-related intervention home practice across 43 studies found a pooled estimate of 64% of the assigned amount of practice completed by participants, equating to approximately 30 minutes per day (Parsons et al., 2017). One of the largest perceived barriers the participants of this study identified was simply in remembering to perform the intervention. As one-minute is such a brief time, it is possible that the intervention is too brief to be appropriately integrated into an individual’s daily schedule. Such a brief intervention could be easily overlooked or left out. This brevity in time may

also have effects on a participant's belief that the treatment will be efficacious. This effect might be mitigated by greater education at the time of learning the technique about any empirically-supported clinical efficacy of the ultra-brief breathing technique. Additionally, more structured reminders and scheduling tools may be required to remind individuals to take a brief moment for their practice. More research is required to understand motivations related to at-home treatment adherence in ultra-brief techniques compared to longer and more complex interventions.

There was no difference in treatment adherence between groups. However, within the Breathing group, increased treatment adherence was positively correlated with increased treatment effect. This finding could be interpreted as reflecting that either the performing of the 4-7-8 technique more frequently leads to greater treatment effects, or that the participants experiencing the greatest treatment effects were more motivated to perform the 4-7-8 technique. However, there was no correlation of treatment adherence and treatment effect within the Counting group, suggesting that this effect is specific to the Breathing group. Furthermore, the Counting group had a significantly larger number of participants with app usage decreasing from week four to week eight relative to those who increased. For the Breathing group, there was no difference in the number of participants that reported a decrease in app usage compared to an increase in app usage in week four relative to week eight. Anecdotally, the originator of the 4-7-8 breathing technique advises that immediately following technique administration, the user may experience transient sensations of tingling or light headedness (Weil, 2000). It is possible that this immediate experiential shift may be interpreted as evidence that the technique is actively helping them, prompting motivation to continue. Balban et al. (2023) also theorize that breathwork interventions that create an acute shift in state likely encourage adherence because people feel better during the intervention.

Alternatively, the breathing group may have been noticing a beneficial effect of the treatment on their anxiety levels, leading to increased motivation to continue to perform the technique throughout the second half of the study. Nonetheless, this stability in treatment adherence over time is promising for the 4-7-8 technique, and further supports that establishing greater technique motivation and adherence at the time of learning the technique may lead to greater adherence throughout the eight

weeks. Future qualitative work will be needed to understand the motivations for this difference in adherence to the technique over time.

## **4.2. Strengths and Limitations**

The conclusions drawn in the current study are supported by the rigor of the methodological design, allowing multiple comparisons of interest. As research of breathing-related techniques typically suffers from poor methodology (Brandani et al., 2017), the inclusion of comparison groups and larger group sample sizes makes this investigation a noteworthy addition to the literature of brief breathing techniques. The experimental breathing technique was compared to a counting technique tailored to reproduce many of the possible experimental and expectation effects that may have been present. These include receiving the same experimental rationales, performing daily practice of a brief technique for an equivalent time through the same mobile application, and identical exposures to study survey materials and research assistant contact. Additionally, the use of three time points instills greater confidence in observing persistent changes over time than a two time-point design.

Several limitations existed within this study. This study consisted of a university sample, which was predominately female and highly educated. Additionally, participants had the incentive of receiving course credit for participating in a research study, which is an external motivation that likely provided a buffer against study drop out rates. Data for the study was collected in the context of a university setting spanning a school semester. As such, the first time point was taken within the first three weeks of the term, when student stress may be relatively low. The final eight-week testing point was taken prior to the exam period of students. Therefore, despite the findings of this study having implications for future research into clinical populations, the generalizability of our findings from this sample is limited.

Regarding generalization of the experimental outcomes reported here, it should be noted this study differed from many studies in the administration of the STICSA trait scale. Commonly, studies will administer the STICSA state scale immediately followed by the STICSA trait scale, however this study omitted state scale measurement. Therefore, care should be taken if directly comparing STICSA numerical outcomes to other investigations use of the STICSA trait scales. Additionally, the STICSA trait scale

was the predominant study outcome. While it has robust psychometric properties (Barros et al., 2022; Ree et al., 2008; Gros et al., 2007) as a self-report measure, it is subject to potential biases and sources of error. Unfortunately, objective physiological measures of anxiety were solely collected before Covid-19 restrictions took effect, which left the objective measures of anxiety underpowered to produce confidence in the reported physiological findings.

The onset of Covid-19 midway through study recruitment yielded an unexpected shift in study methodology. Prior to the Covid-19 restrictions, participants were randomized into three groups, completed self-report measurements in a controlled environment monitored by a research assistant, and had objective physiological measurements performed. Following Covid-19 restrictions, the waitlist control group was discontinued, physiological measurements were discontinued, and all self-report data were provided through unmonitored online surveys. Despite participants being instructed to complete surveys in a secure and private location, there is no assurance where participants were when completing study surveys. Demographic analyses showed very few differences between the two cohorts. However, the online, during Covid-19 cohort reported significantly higher negative PANAS scores (negative state affect) than the in-person, pre-Covid-19 cohort. There was also a nonsignificant trend towards the online cohort identifying more barriers to technique acceptance when learning the ultra-brief experimental technique online.

One can only speculate whether the cohort-based differences reflect an effect of environmental context or an effect of stressors related to the Covid-19 pandemic. While research findings are mixed, some investigations have found that completing questionnaires online may lead to higher scores overall than in-person responding for some questionnaires (Buchanan, 2003; Buchanan and Smith, 1999) suggesting that internet administration leads to higher scores overall. Others have found no difference between in-person and online administration of mood-related questionnaires (Fouladi et al., 2002). This trend could explain the higher PANAS negative scores reported in the online cohort reported here. However, resting state negative affect could also be reasonably interpreted as a result of the larger context of the Covid-19 pandemic. Despite these cohort-differences, there were no statistical differences in trait anxiety between cohorts, the primary outcome of the study. Thus analyses were conducted by combining the two cohorts, and statistically controlling for the effect of cohort. However,



it was not logistically possible to control for the effect of study cohort in all analyses. For instance, analyses of physiological data is only representative of the pre-Covid-19 study cohort. Additionally, as participants in the waitlist control group were all from the pre-Covid-19 study cohort, the variable of 'study cohort' was not included in analyses including the waitlist control group.

Additionally, in part due to the numerous statistical analyses performed in this study, it was decided to only perform analyses on STICSA Subscales for Cognitive and Somatic Anxiety when there was at least an identifiable trend ( $p < 0.10$ ) in the STICSA Total Anxiety outcome. As the total score is a summated score of the two subscales, it provides more complete coverage of the construct of anxiety and more points of discrimination to differentiate individuals. However, in choosing to not analyze the Cognitive and Somatic subscales for each analysis, it is possible that some findings went unidentified in this approach.

### **4.3. Future Research**

While this research supports the 4-7-8 breathing technique as more effective at decreasing self-reported symptoms of trait anxiety than a counting exercise, it is unknown which aspect(s) of the technique are responsible for the clinical efficacy. Future studies are necessary to deconstruct the 4-7-8 breathing technique into its active ingredients. Jafari et al (2020) have undertaken a dismantling approach to breathing techniques in understanding breathing's effect on pain perception by comparing differences in pain perception among four conditions: a group that breathes spontaneously, a group that breathes in a controlled rhythm at a frequency that matches their spontaneous breathing frequency, a slow breathing condition with an elongated inhalation, and a slow breathing condition with an elongated exhalation. The Jafari et al study showed that when a mild pain stimulus was introduced, all three groups that consciously engaged attention with their breathing reported lower scores of pain than a group that was spontaneously breathing. There were no differences between the three breathing-engaged groups. This suggests that paying attention to breathing has a general effect on pain perception, regardless of breathing frequency or inhalation-exhalation ratio. However, when a moderate pain stimulus was introduced, a tiered response of pain perception emerged, such that slow breathing with an

elongated exhalation was the most effective breathing strategy for lowering pain perception.

While the mechanism of breathing's effect on pain perception may be different than breathing's effect on trait anxiety, a similar dismantling study design would provide clarity to what the active ingredient(s) of the 4-7-8 technique are among the various components of breathing that are manipulated within the technique, including 1) focusing on the breath 2) breathing at a slower frequency 3) breathing with an elongated exhalation, and 4) introducing a retention of the breath.

Finally, the intervention effect of this study was only confirmed after eight weeks, not after four. Between week four and week eight, participants increased the duration of the breathing sequence from one minute to two minutes. It is unclear if the treatment gains were due to performing the technique for the additional four weeks, or for performing the breathing sequence at two-minute intervals. Future comparisons of interest would also include a group that performs the one-minute interval throughout the entirety of the eight weeks, and a group that performs the two-minute interval from the study outset.

Additionally, having eight-week treatment outcomes on physiological measurements would be of interest. Due to methodological obstacles presented by the onset of the Covid-19 pandemic, data collection of physiological measurements could only power analyses of treatment outcomes at four weeks for the present study. As differences in the treatment outcome of trait anxiety was only observed to occur at eight weeks, having baseline physiological measurements of eight-week outcomes would be of interest for future research. Outcomes of comparisons with an eight-week waitlist control would also be of interest.

As technique adherence was correlated with anxiety outcome in the breathing group, factors that predict increased treatment adherence should be better understood. Post-study qualitative interviews would help identify these factors.

#### **4.4. Conclusions**

To summarize, the present study was the first to investigate the efficacy of regularly practicing the ultra-brief 4-7-8 breathing technique on measures of anxiety. In

this predominantly female sample of university students self-reporting mild to severe symptoms of anxiety, performing the 4-7-8 breathing technique for eight weeks led to significantly decreased trait anxiety of a medium effect size compared to participants performing a counting exercise of identical duration. Differences were specific to trait cognitive anxiety, not trait somatic anxiety. Differences were specific to individuals who performed the breathing technique a minimum of 4 times a week for the duration of the required eight weeks. There were no significant differences in resting state physiological markers or trait anxiety after four weeks, though these effects may appear given a larger sample size. In the Breathing group, but not the Counting group, increased treatment adherence was positively correlated with increased treatment effect. This investigation supports the conclusion that regular daily practice of the ultra-brief 4-7-8 breathing technique can be of clinical utility, though more benefit is likely to come when practiced with more regularity (twice a day) for 8 weeks.

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# APPENDICES

## Appendix A: Demographic Questionnaire

### DEMOGRAPHIC QUESTIONNAIRE

A BRIEF, ONE MINUTE RELAXATION THERAPY

ID code \_\_\_\_\_

How old are you? \_\_\_\_\_

What is your gender?  female  male  transgender

What is your race/ethnicity?

- |   |  |
|---|--|
| <input type="radio"/> Asian or Pacific Islander | <input type="radio"/> Black / African American   |
| <input type="radio"/> Indian                    | <input type="radio"/> First Nations / Indigenous |
| <input type="radio"/> White/ Caucasian          | <input type="radio"/> Other _____                |

Do you currently use tobacco?

- Yes, on a regular basis  Yes, but only once in awhile  
 Not anymore, I quit  No, I have never used tobacco

How often do you have a drink containing alcohol?

- never  
 monthly or less  
 2-4 times a month  
 2-3 times a week  
 4 or more times a week

Are you taking any:  cardioactive medication  antihypertensive medication  
 antidepressant medication  antipsychotic medication

Please list any additional medications you are currently taking:


Do you currently have a practice of meditation, yoga, or breathing exercises?  yes  no

Other: \_\_\_\_\_

If yes, how frequently do you practice?  everyday  a couple times a week  once a week

Are you regularly meeting with a counsellor or psychologist?  yes  no

## Appendix B: STICSA Trait

DIRECTIONS: Below is a list of statements which can be used to describe how people feel. Beside each statement are four numbers which indicate how often each statement is true of you (e.g., 1 = *not at all*, 4 = *very much so*). Please read each statement carefully and select the number which best indicates how often, in general, the statement is true of you.

	Not at All	A Little	Moderately	Very Much So
1. My heart beats fast	1	2	3	4
2. My muscles are tense	1	2	3	4
3. I feel agonized over my problems	1	2	3	4
4. I think that others won't approve of me	1	2	3	4
5. I feel like I'm missing out on things because I can't make up my mind	1	2	3	4
6. I feel dizzy	1	2	3	4
7. My muscles feel weak	1	2	3	4
8. I feel trembly and shaky	1	2	3	4
9. I picture some future misfortune	1	2	3	4
10. I can't get some thought out of my mind	1	2	3	4
11. I have trouble remembering things	1	2	3	4
12. My face feels hot	1	2	3	4
13. I think the worst will happen	1	2	3	4
14. My arms and legs feel stiff	1	2	3	4
15. My throat feels dry	1	2	3	4
16. I keep busy to avoid uncomfortable thoughts	1	2	3	4
17. I cannot concentrate without irrelevant thoughts intruding	1	2	3	4
18. My breathing is fast and shallow	1	2	3	4
19. I worry that I cannot control my thoughts as well as I would like to	1	2	3	4
20. I have butterflies in my stomach	1	2	3	4
21. My palms feel clammy.	1	2	3	4

## **Appendix C: PANAS**

Indicate the extent you feel this way right now



	Very slightly or not at all	A little	Moderately	Quite a bit	Extremely
Interested	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Distressed	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Excited	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Upset	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Strong	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Guilty	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Scared	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Hostile	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Enthusiastic	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Proud	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Irritable	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Alert	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Ashamed	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>

Inspired	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Nervous	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Determined	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Attentive	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Jittery	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Active	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Afraid	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>

## Appendix D: Technique Acceptability

Select how much you agree with each of the following statements

	Strongly agree	Somewhat agree	Neither agree nor disagree	Somewhat disagree	Strongly disagree
The technique seems complex to me	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Following this technique will interrupt my daily routines	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
The technique will require a lot of my time for me to accomplish	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
I am capable of performing the technique	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>

## Appendix E: Study Completion

Select how much you agree with each of the following statements

	Strongly agree	Somewhat agree	Neither agree nor disagree	Somewhat disagree	Strongly disagree
I was satisfied with the intervention.	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
The intervention was simple.	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
The intervention was easy to perform	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
It was difficult to remember to do the intervention	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
I could do the intervention anywhere	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
I would recommend this intervention to a friend	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>

## Appendix F: Analyses of Study Dropouts

Independent Samples Test Between Study Drop-outs (n=15) and Study Completers (n=104)								
		t-test for Equality of Means						
	Equal Variance Assume	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% CI	
							Lower	Upper
Experimental Group	yes	-0.733	117	0.465	-0.151	0.206	-0.558	0.257
Cohort	yes	0.098	117	0.922	0.01346	0.137	-0.258	0.284
Age	no	-0.731	15.287	0.476	-0.487	0.667	-1.906	0.932
Sex	no	1.524	16.055	0.147	0.199	0.130	-0.078	0.475
English Second Language	yes	-1.148	117	0.253	-0.131	0.114	-0.358	0.095
STICSA Total	yes	-0.300	117	0.765	-0.900	2.997	-6.836	5.036
TA: Complex	yes	0.330	116	0.742	0.08673	0.263	-0.434	0.607
TA: Routine	yes	0.836	116	0.405	0.22265	0.266	-0.304	0.750
TA: Time	yes	-0.80	116	0.424	-0.17411	0.216	-0.603	0.255
TA: Capable	yes	-0.33	116	0.740	-0.05243	0.157	-0.365	0.260
TA: Total	yes	0.263	117	0.793	0.16987	0.646	-1.110	1.450
TA: Any Disagreement	no	1.511	24.271	0.144	0.11603	0.076	-0.042	0.274
Any current mindfulness techniques	yes	-1.82	117	0.071	-0.235	0.129	-0.491	0.020
PANAS positive	yes	-0.584	117	0.560	-1.080	1.849	-4.742	2.581
PANAS negative	yes	0.542	117	0.589	0.946	1.746	-2.511	4.404

\*All data from Baseline. TA= Technique Acceptance

**Appendix G Statistical Output for Section 3.2.1.1: Is the mean change in trait anxiety after four weeks different between the Breathing, Counting, or Waitlist Control groups?**

Shapiro-Wilk's Test of Normality				
	Group	Statistic	df	Sig.
STICSA Total	WLC	0.914	23	0.05
	Breathing	0.926	45	0.007
	Counting	0.923	36	0.015
ln(STICSA Total)	WLC	0.932	23	0.123
	Breathing	0.954	45	0.071
	Counting	0.945	36	0.072

**Table 10.** Shapiro Wilk's test of normality of the distribution of the outcome variable STICSA Total Score, and the outcome variable after a log-transformation. Distributions analyzed at a group level, with Waitlist Control (WLC, n = 23, Breathing n= 46, Counting n = 33).

Tests of Within-Subjects Effects ln(Total Anxiety) 3 Groups*2 Timepoints							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.008	1	0.008	0.347	0.557	0.003	0.090
Time * Age	0.013	1	0.013	0.578	0.449	0.006	0.117
Time * Sex	0.004	1	0.004	0.169	0.682	0.002	0.069
Time * Group	0.071	2	0.035	1.567	0.214	0.031	0.325
Error(Time)	2.241	99	0.023				

**Table 11.** Statistical output of the Within-Subjects Effects of a repeated measures ANOVA controlling for age and sex of three groups (WLC n = 23, Breathing n = 45, Counting n = 36) over two timepoints (Baseline and Week 4).

**Tests of Between-Subjects Effects**

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	13.628	1	13.628	213.639	.000
Age	.001	1	.001	.023	.879
Sex	.048	1	.048	.756	.387
Group	.121	2	.060	.948	.391
Error	6.315	99	.064		

**Appendix H Statistical Output for Section 3.2.1.2: Is the mean change in physiological resting state measures after four weeks different between the Breathing, Counting, or Waitlist Control groups?**

Tests of Within-Subjects Effects of Systolic Blood Pressure							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	1.636	1	1.636	0.08	0.779	0.001	0.059
Time * Age	3.281	1	3.281	0.16	0.691	0.003	0.068
Time * Sex	56.579	1	56.579	2.762	0.102	0.048	0.372
Time * Group	75.276	2	37.638	1.837	0.169	0.063	0.367
Error(Time)	1126.77	55	20.487				

*Computed using alpha = .05*

**Table 13.** Statistical output of the Within-Subjects Effects of a repeated measures ANOVA controlling for age and sex of three groups (WLC n = 23, Breathing n = 22, Counting n = 15) over two timepoints (Baseline and Week 4)

**Tests of Between-Subjects Effects**

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	4348.919	1	4348.919	32.103	.000
Age	442.580	1	442.580	3.267	.076
Sex	459.730	1	459.730	3.394	.071
Group	526.057	2	263.028	1.942	.153
Error	7450.786	55	135.469		

**Table 14.** Statistical output of the Between-Subjects Effects of a repeated measures ANOVA controlling for age and sex of three groups (WLC n = 23, Breathing n = 22, Counting n = 15) over two timepoints (Baseline and Week 4)

Tests of Within-Subjects Effects of Diastolic Blood Pressure							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	6.621	1	6.621	0.57	0.454	0.01	0.115
Time * Age	1.565	1	1.565	0.135	0.715	0.002	0.065
Time * sex	23.976	1	23.976	2.064	0.156	0.036	0.292
Time * GROUP	6.475	2	3.238	0.279	0.758	0.01	0.092
Error(Time)	638.953	55	11.617				

*Computed using alpha = .05*

**Table 15.** Statistical output of the Within-Subjects Effects of a repeated measures ANOVA controlling for age and sex of three groups (WLC n = 23, Breathing n = 22, Counting n = 15) over two timepoints (Baseline and Week 4).

**Tests of Between-Subjects Effects DBP**

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	1394.684	1	1394.684	19.983	.000
Age	183.002	1	183.002	2.622	.111
Sex	26.357	1	26.357	.378	.541
Group	162.655	2	81.327	1.165	.319
Error	3838.727	55	69.795		

**Table 16.** Statistical output of the Between-Subjects Effects of a repeated measures ANOVA controlling for age and sex of three groups (WLC n = 23, Breathing n = 22, Counting n = 15) over two timepoints (Baseline and Week 4). DBP

<b>Tests of Within-Subjects Effects of ln(SDNN)</b>							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.109	1	0.109	1.683	0.2	0.03	0.247
Time * Age	0.105	1	0.105	1.614	0.209	0.029	0.239
Time * Sex	0.042	1	0.042	0.645	0.425	0.012	0.124
Time * Group	0.227	2	0.113	1.75	0.183	0.06	0.351
Error(Time)	3.565	55	0.065				

*Computed using alpha = .05*

**Table 17.** Statistical output of the Within-Subjects Effects of a repeated measures ANOVA controlling for age and sex of three groups (WLC n = 23, Breathing n = 22, Counting n = 15) over two timepoints (Baseline and Week 4).

**Tests of Between-Subjects Effects ln(SDNN)**

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	11.819	1	11.819	88.625	.000
Age	.116	1	.116	.872	.355
Sex	1.117	1	1.117	8.376	.005
Group	.841	2	.420	3.152	.051
Error	7.335	55	.133		

**Table 18.** Statistical output of the Between-Subjects Effects of a repeated measures ANOVA controlling for age and sex of three groups (WLC n = 23, Breathing n = 22, Counting n = 15) over two timepoints (Baseline and Week 4).



**Appendix I Statistical Output for Section 3.2.1.3: Is the mean change in trait anxiety after eight weeks different between the Breathing and Counting groups?**

Levene's Test of Equality of Error Variances for Log-Transformed Total Anxiety				
	F	df1	df2	Sig.
Baseline	0.005	1	73	0.941
Week 4	0.328	1	73	0.568
Week 8	0.00	1	73	0.993

Tests of Within-Subjects Effects ln(Total Anxiety)							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.008	1.984	0.004	0.182	0.832	0.003	0.078
Time * Adherence	0.070	1.984	0.035	1.605	0.205	0.023	0.334
Time * Cohort	0.088	1.984	0.044	2.012	0.138	0.028	0.408
Time * Sex	0.018	1.984	0.009	0.402	0.668	0.006	0.114
Time * Age	0.020	1.984	0.010	0.459	0.631	0.007	0.123
Time * Group	0.110	1.984	0.055	2.511	0.085	0.035	0.494
Error(Time)	3.018	136.918	0.022				

**Table 19.** Statistical output of the Within-Subjects Effects of a repeated measures ANOVA of two groups (Breathing n = 42, Counting n = 33) over three timepoints (Baseline, Week 4, Week 8) while controlling for data cohort, intervention fidelity, age, and sex. Sphericity corrections made with Huynh-Feldt estimates of sphericity.

Tests of Between-Subjects Effects ln(Total Anxiety)

	Type III SoS	df	Mean Square	F	Sig.	Partia Eta^2	Observed Power
Intercept	14.805	1	14.805	155.548	0	0.693	1
Adherence	0.029	1	0.029	0.307	0.581	0.004	0.085
Cohort	0.008	1	0.008	0.087	0.769	0.001	0.06
Sex	0.045	1	0.045	0.472	0.494	0.007	0.104
Age	0.02	1	0.02	0.211	0.647	0.003	0.074
Group	0.002	1	0.002	0.019	0.891	0	0.052
Error	6.568	69	0.095				

Table 20. Statistical output of the Between-Subjects Effects of a repeated measures ANOVA of two groups (Breathing n = 42, Counting n = 33) over three timepoints (Baseline, Week 4, Week 8) while controlling for data cohort, intervention fidelity, age, and sex.

**Follow-up Analyses with STICSA subscales: cognitive and somatic (Bonferroni corrected for the 2 analyses being performed)**

Tests of Within-Subjects Effects In(Cognitive Anxiety)							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.002	2	0.001	0.028	0.972	0.000	0.054
Time * Adherence	0.086	2	0.043	1.599	0.206	0.023	0.334
Time * Cohort	0.134	2	0.067	2.469	0.088	0.035	0.489
Time * Sex	0.017	2	0.008	0.309	0.735	0.004	0.098
Time * Age	0.004	2	0.002	0.076	0.927	0.001	0.061
Time * Group	0.173	2	0.087	3.207	0.044	0.044	0.605
Error(Time)	3.731	138	0.027				

**Table 21.** Statistical output of the Within-Subjects Effects of a repeated measures ANOVA of two groups (Breathing n = 42, Counting n = 33) over three timepoints (Baseline, Week 4, Week 8) while controlling for data cohort, intervention adherence, age, and sex

Tests of Between-Subjects Effects In(Cognitive Anxiety)							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Intercept	10.863	1	10.863	95.009	0.000	0.579	1.000
Adherence	0.013	1	0.013	0.114	0.736	0.002	0.063
Cohort	0.001	1	0.001	0.006	0.937	0.000	0.051
Sex	0.164	1	0.164	1.437	0.235	0.020	0.219
Age	0.009	1	0.009	0.077	0.782	0.001	0.059
Group	0.024	1	0.024	0.210	0.648	0.003	0.074
Error	7.889	69	0.114				

**Table 22.** Statistical output of the Between-Subjects Effects of a repeated measures ANOVA of two groups (Breathing n = 42, Counting n = 33) over three timepoints (Baseline, Week 4, Week 8) while controlling for data cohort, intervention fidelity, age, and sex.

Tests of Within-Subjects Effects In(Somatic Anxiety)							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.041	2.000	0.020	0.683	0.507	0.010	0.163
Time * Adherence	0.074	2.000	0.037	1.246	0.291	0.018	0.268
Time * Cohort	0.054	2.000	0.027	0.907	0.406	0.013	0.204
Time * Sex	0.022	2.000	0.011	0.362	0.697	0.005	0.107
Time * Age	0.072	2.000	0.036	1.212	0.301	0.017	0.261
Time * Group	0.058	2.000	0.029	0.970	0.382	0.014	0.216
Error(Time)	4.111	138.000	0.030				

**Table 23.** Statistical output of the Within-Subjects Effects of a repeated measures ANOVA of two groups (Breathing n = 42, Counting n = 33) over three timepoints (Baseline, Week 4, Week 8) while controlling for data cohort, intervention fidelity, age, and sex. A Hyun-Feldt correction was used to control for violations of sphericity.

Tests of Between-Subjects Effects In(Somatic Anxiety)							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Intercept	9.191	1	9.191	58.035	0.000	0.457	1.000
Adherence	0.044	1	0.044	0.281	0.598	0.004	0.082
Cohort	0.018	1	0.018	0.113	0.738	0.002	0.063
Sex	0.000	1	0.000	0.002	0.966	0.000	0.050
Age	0.034	1	0.034	0.217	0.643	0.003	0.074
Group	0.003	1	0.003	0.017	0.896	0.000	0.052
Error	10.927	69	0.158				

**Table 24.** Statistical output of the Between-Subjects Effects of a repeated measures ANOVA of two groups (Breathing n = 42, Counting n = 33) over three timepoints (Baseline, Week 4, Week 8) while controlling for data cohort, intervention fidelity, age, and sex.

**Appendix J Statistical Output for Section 3.2.1.4: Is the mean change in trait anxiety in the Waitlist Control group different in the first four weeks of the study, compared to the second four weeks once they begin the Breathing technique?**

<b>Tests of Within-Subjects Effects of the WLC over 8 Weeks In(Tot Anx)</b>							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.106	2	0.053	2.005	0.150	0.106	0.385
Time * Age	0.113	2	0.057	2.150	0.132	0.112	0.410
Time * Sex	0.007	2	0.004	0.135	0.874	0.008	0.069
Error(Time)	0.898	34	0.026				

**Table 25.** Statistical output of the Within-Subjects Effects of a repeated measures ANOVA controlling age and sex of the WLC (n = 20) over three timepoints (Baseline, Week 4, Week 8)

**Tests of Within-Subjects Effects In(Cognitive Anxiety) within WLC**

	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Time	0.130	2	0.065	2.063	0.143	0.108
Time * Age	0.126	2	0.063	2.007	0.150	0.106
Time * Sex	0.015	2	0.008	0.243	0.785	0.014
Error(Time)	1.067	34	0.031			

**Ln(Cognitive Anxiety) Pairwise Comparisons within WLC**

Time		Mean Difference	Std. Error	Sig.	95% Confidence Interval for Difference	
					Lower Bound	Upper Bound
Baseline	Week 4	.015	0.045	0.740	-0.080	0.110
Week 4	Week 8	.121	0.051	0.030	0.013	0.229

Based on estimated marginal means

\*. significant at the .025 level

**Tests of Within-Subjects Effects In(Somatic Anxiety) within WLC**

	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Time	0.088	2	0.044	1.237	0.303	0.068
Time * Age	0.112	2	0.056	1.578	0.221	0.085
Time * Sex	0.019	2	0.010	0.269	0.766	0.016
Error(Time)	1.207	34	0.035			

**Appendix K Statistical Output for Section 3.2.1.5: Is there a significant change in trait anxiety after four weeks for a subgroup of clinical severity?**

**Tests of Normality**

	Shapiro-Wilk Statistic	df	Sig.
HighCog1	.953	47	.059
HighCog2	.958	47	.088
HighSom1	.954	47	.061
HighSom2	.963	47	.144

**High Anxiety Cognitive controlling for adherence, cohort, age, sex**

Tests of Within-Subjects Effects Cognitive							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	19.955	1	19.955	1.381	0.247	0.032	0.209
Time * Adherence	0.632	1	0.632	0.044	0.835	0.001	0.055
Time * Cohort	44.233	1	44.233	3.062	0.087	0.068	0.401
Time * Sex	42.364	1	42.364	2.932	0.094	0.065	0.387
Time * Age	60.348	1	60.348	4.177	0.047	0.090	0.515
Error(Time)	606.767	42	14.447				

Estimated Means Cognitive				
Time	Mean	Std. Error	95% Confidence Interval	
			Lower Bound	Upper Bound
1	29.872 <sup>a</sup>	0.616	28.630	31.115
2	26.277 <sup>a</sup>	0.946	24.367	28.186
Covariates are evaluated with: Self Reported App Usage = 2.89, Cohort = 1.4, Sex = 1.89, Age = 18.64.				

Pairwise Comparisons Cognitive						
Time		Mean Diff	St Error	Sig	95% CI	
					Lower	Upper
1	2	3.596*	0.784	0.000	2.013	5.178
2	1	-3.596*	0.784	0.000	-5.178	-2.013
Based on estimated marginal means						

Multivariate Tests of Estimated Marginal Means Cognitive							
	Value	F	Hypothesis df	Error df	Sig.	Partial Eta Squared	Observed Power
Wilks' lambda	0.666	21.032	1.000	42.000	0.000	0.334	0.994
Each F tests the multivariate effect of Time. These tests are based on the linearly independent pairwise comparisons among the estimated marginal means.							

### High Anxiety Somatic controlling for adherence, cohort, age, sex

Tests of Within-Subjects Effects Somatic							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	1.268	1	1.268	0.068	0.795	0.002	0.058
Time * Adherence	0.443	1	0.443	0.024	0.878	0.001	0.053
Time * Cohort	65.726	1	65.726	3.549	0.067	0.078	0.453
Time * Sex	2.202	1	2.202	0.119	0.732	0.003	0.063
Time * Age	0.010	1	0.010	0.001	0.981	0.000	0.050
Error(Time)	777.811	42	18.519				

Estimates Means Somatic				
Time	Mean	Std. Error	95% Confidence Interval	
			Lower Bound	Upper Bound

1	24.596 <sup>a</sup>	1.033	22.511	26.681
2	21.170 <sup>a</sup>	0.811	19.534	22.806

Pairwise Comparisons Somatic						
(I) Time		Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval for Difference	
					Lower Bound	Upper Bound
1	2	3.426 <sup>*</sup>	0.888	0.000	1.634	5.217

Multivariate Tests of Estimated Marginal Means Somatic							
	Value	F	Hypothesis df	Error df	Sig.	Partial Eta Squared	Observed Power
Wilks' lambda	0.738	14.89	1.000	42.000	0.000	0.262	0.965

## Appendix L Treatment Acceptance Questions by Group

<b>The technique seems complex to me</b>			
	GROUP		Total
	Breathing	Counting	
strongly disagree	31	23	54
somewhat disagree	8	9	17
neither	1	0	1
somewhat agree	4	3	7
strongly agree	0	1	1
	44	36	80

<b>Following this technique will interrupt my daily routine</b>			
	GROUP		Total
	Breathing	Counting	
strongly disagree	26	18	44
somewhat disagree	10	10	20
neither	4	4	8
somewhat agree	4	4	8
strongly agree	0	0	0
	44	36	80

<b>The technique will require a lot of my time to accomplish</b>			
	GROUP		Total
	Breathing	Counting	
strongly disagree	34	24	58
somewhat disagree	7	8	15
neither	0	1	1
somewhat agree	3	3	6
strongly agree	0	0	0
	44	36	80

<b>I am capable of performing the technique</b>			
	GROUP		Total
	Breathing	Counting	
strongly disagree	1	0	1
somewhat disagree	0	0	0
neither	0	2	2
somewhat agree	8	6	14
strongly agree	35	28	63
	44	36	80



**Appendix M Statistical Output for Adequate Adherence Subgroup  
Section 3.3.1.1:**

<b>Tests of Within-Subjects Effects In(Total Anxiety) of Adequate Adherence</b>							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	.0001	1	.0001	0.004	0.952	0.000	0.050
Time * Age	0.003	1	0.003	0.134	0.715	0.001	0.065
Time * Sex	0.001	1	0.001	0.026	0.873	0.000	0.053
Time * GROUP	0.110	2	0.055	2.484	0.089	0.051	0.487
Error(Time)	2.050	93	0.022				

<b>Tests of Between-Subjects Effects In(Total Anxiety) of Adequate Adherence</b>								
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power	
Intercept	10.666	1	10.666	166.081	0.000	0.641	1.000	
Age	0.003	1	0.003	0.049	0.826	0.001	0.055	
sex	0.031	1	0.031	0.479	0.491	0.005	0.105	
GROUP	0.082	2	0.041	0.642	0.528	0.014	0.155	
Error	5.973	93	0.066					

**Follow-up Analyses with STICSA subscales of Adequate Adherence subgroup:**

<b>Tests of Within-Subjects Effects In(Cognitive Anxiety) of AA</b>							
Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.002	1	0.002	0.065	0.800	0.001	0.057
Time * Age	0.000	1	0.000	0.002	0.964	0.000	0.050
Time * Sex	0.000	1	0.000	0.000	0.996	0.000	0.050
Time * Group	0.161	2	0.080	3.295	0.041	0.066	0.612
Error(Time)	2.266	93	0.024				

<b>Tests of Between-Subjects Effects In(Cognitive Anxiety) AA</b>					
Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	7.644	1	7.644	91.344	0.000
Age	0.003	1	0.003	0.039	0.844
sex	0.009	1	0.009	0.102	0.750
GROUP	0.044	2	0.022	0.265	0.768
Error	7.251	93	0.084		

<b>Tests of Within-Subjects Effects In(Somatic Anxiety) of AA</b>							
Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.007	1	0.007	0.217	0.642	0.002	0.075
Time * Age	0.018	1	0.018	0.545	0.462	0.006	0.113
Time * sex	0.000	1	0.000	0.015	0.904	0.000	0.052
Time * Group	0.072	2	0.036	1.083	0.343	0.023	0.235
Error(Time)	3.086	93	0.033				

<b>Tests of Between-Subjects Effects In(Somatic Anxiety) AA</b>					
Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	6.146	1	6.146	59.897	0.000
Age	0.005	1	0.005	0.050	0.824
sex	0.118	1	0.118	1.154	0.285
GROUP	0.130	2	0.065	0.632	0.534
Error	9.542	93	0.103		

**Appendix N Statistical Output for Adequate Adherence Subgroup  
Section 3.3.1.2:**

**Tests of Within-Subjects Effects SYSTOLIC**

	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	10.494	1	10.494	0.533	0.469	0.010	0.111
Time * Age	44.229	1	44.229	2.246	0.140	0.042	0.313
Time * sex	79.006	1	79.006	4.012	0.051	0.073	0.502
Time * GROUP	80.314	2	40.157	2.039	0.141	0.074	0.401
Error(Time)	1004.258	51	19.691				

**Tests of Between-Subjects Effects SYSTOLIC**

	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Intercept	3412.063	1	3412.063	26.219	0.000	0.340	0.999
Age	93.764	1	93.764	0.721	0.400	0.014	0.132
sex	492.295	1	492.295	3.783	0.057	0.069	0.479
GROUP	264.266	2	132.133	1.015	0.369	0.038	0.217
Error	6636.873	51	130.135				

**Tests of Within-Subjects Effects DIASTOLIC**

	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	3.903	1	3.903	0.320	0.574	0.006	0.086
Time * Age	01.158	1	01.158	0.095	0.759	0.002	0.061
Time * sex	15.255	1	15.255	1.250	0.269	0.024	0.195
Time * GROUP	2.293	2	1.147	0.094	0.910	0.004	0.064
Error(Time)	622.411	51	12.204				

**Tests of Between-Subjects Effects DIASTOLIC**

---

	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Intercept	1097.695	1	1097.695	15.100	0.000	0.228	0.968
Age	43.017	1	43.017	0.592	0.445	0.011	0.117
sex	35.299	1	35.299	0.486	0.489	0.009	0.105
GROUP	126.006	2	63.003	0.867	0.426	0.033	0.191
Error	3707.453	51	72.695				

**Tests of Within-Subjects Effects In(SDNN)**

	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.056	1	0.056	0.812	0.372	0.016	0.143
Time * Age	0.051	1	0.051	0.741	0.393	0.014	0.135
Time * sex	0.027	1	0.027	0.389	0.536	0.008	0.094
Time * GROUP	0.277	2	0.138	2.026	0.142	0.074	0.399
Error(Time)	3.487	51	0.068				

**Tests of Between-Subjects Effects In(SDNN)**

	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Intercept	6.715	1	6.715	52.334	0.000	0.505	1.000
Age	0.032	1	0.032	0.249	0.620	0.005	0.078
sex	0.639	1	0.639	4.977	0.030	0.089	0.591
GROUP	0.625	2	0.312	2.434	0.098	0.087	0.468
Error	6.543	51	0.128				

## Appendix O Statistical Output for Adequate Adherence Subgroup Section 3.3.1.3

### AA Subgroup Tests of Within-Subjects Effects In(Total Anxiety)

	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.002	2	0.001	0.040	0.961	0.001	0.056
Time * Adherence	0.063	2	0.032	1.494	.228	0.022	0.314
Time * Cohort	0.068	2	0.034	1.597	0.206	0.024	0.333
Time * Sex	0.000	2	0.000	0.01	0.990	0.005	0.100
Time * Age	0.014	2	0.007	0.319	0.727	0.010	0.149
Time * Group	0.158	2	0.079	3.722	0.027	0.053	0.674
Error(Time)	2.799	132	0.021				

a. Computed using alpha = .05

### AA Subgroup Tests of Between-Subjects Effects In(Total Anxiety)

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Intercept	13.623	1	13.623	146.634	0.000	0.690	1.000
Adherence	0.121	1	0.121	1.303	0.258	0.019	0.203
Cohort	0.0001	1	0.0001	.000	0.990	0.000	0.050
Sex	0.022	1	0.022	0.232	0.631	0.004	0.076
Age	0.045	1	0.045	0.488	0.487	0.007	0.106
Group	0.021	1	0.021	0.229	0.634	0.003	0.076
Error	5.284	66	0.091				

### AA Subgroup Tests of Within-Subjects Effects In(Cognitive Anxiety)

	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.005	2	0.002	0.098	0.907	0.001	0.065
Time * Adherence	0.090	2	0.045	1.841	0.163	0.027	0.378
Time * Cohort	0.106	2	0.053	2.158	0.120	0.032	0.435

Time * Sex	0.002	2	0.001	0.046	0.955	0.001	0.057
Time * Age	0.007	2	0.004	0.145	0.865	0.002	0.072
Time * Group	0.274	2	0.137	5.571	0.005	0.078	0.849
Error(Time)	3.243	132	0.025				
Computed using alpha = .025 for Multiple Comparisons							

<b>AA Subgroup Tests of Between-Subjects Effects In(Cognitive Anxiety)</b>							
Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Intercept	9.854	1	9.854	85.895	0.000	0.565	1.000
Adherence	0.093	1	0.093	0.810	0.372	0.012	0.144
Cohort	0.004	1	0.004	0.036	0.851	0.001	0.054
Sex	0.095	1	0.095	0.829	0.366	0.012	0.146
Age	0.020	1	0.020	0.172	0.680	0.003	0.069
Group	0.051	1	0.051	0.446	0.507	0.007	0.101
Error	7.572	66	0.115				

<b>AA Subgroup Tests of Within-Subjects Effects In(Somatic Anxiety)</b>							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.032	2	0.016	0.516	0.598	0.008	0.134
Time * Adherence	0.057	2	0.029	0.939	0.394	0.014	0.210
Time * Cohort	0.038	2	0.019	0.614	0.543	0.009	0.151
Time * Sex	0.011	2	0.005	0.175	0.840	0.003	0.077
Time * Age	0.062	2	0.031	1.008	0.368	0.015	0.223
Time * Group	0.063	2	0.031	1.023	0.362	0.015	0.226
Error(Time)	4.040	132	0.031				
Computed using alpha = .025 for Multiple Comparisons							

**AA Subgroup Tests of Between-Subjects Effects In(Somatic Anxiety)**

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Intercept	8.717	1	8.717	56.475	0.000	0.461	1.000
Adherence	0.138	1	0.138	0.897	0.347	0.013	0.154
Cohort	0.003	1	0.003	0.019	0.890	0.000	0.052
Sex	0.002	1	0.002	0.015	0.903	0.000	0.052
Age	0.081	1	0.081	0.524	0.471	0.008	0.110
Group	0.009	1	0.009	0.059	0.809	0.001	0.057
Error	10.188	66	0.154				

## Appendix P Statistical Output for Adequate Adherence Subgroup Section 3.3.1.4

AA Subgroup: Tests of Within-Subjects Effects of the WLC over 8 Weeks							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.099	2	0.049	1.784	0.184	0.100	0.345
Time * Age	0.105	2	0.052	1.887	0.168	0.106	0.363
Time * Sex	0.009	2	0.005	0.165	0.848	0.010	0.073
Error(Time)	0.886	32	0.028				

Table 25. Statistical output of the Within-Subjects Effects of a repeated measures ANOVA controlling for age and sex of the WLC ( $n = 19$ ) over three timepoints (Baseline, Week 4, Week 8)

### AA Subgroup Tests of Between-Subjects Effects In(Somatic Anxiety)

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Intercept	2.463	1	2.463	18.603	0.001	0.538	0.981
Age	0.001	1	0.001	0.006	0.939	0.000	0.051
Sex	0.036	1	0.036	0.268	0.612	0.016	0.078
Error	2.118	16	0.132				

### AA Subgroup: Pairwise Comparisons within WLC by Time Point

Time		Mean Difference	Std. Error	Sig.	95% Confidence Interval for Difference	
					Lower Bound	Upper Bound
Baseline	Week 4	0.013	0.046	0.776	-0.085	0.112
Week 4	Week 8	.125*	0.046	0.016	0.027	0.222

Based on estimated marginal means

\*. The mean difference is significant at the .025 level in adjustment for multiple comparisons.

### AA Subgroup: Cognitive Anxiety Pairwise Comparisons within WLC

Time		Mean Difference	Std. Error	Sig.	95% Confidence Interval for Difference	
					Lower Bound	Upper Bound
Baseline	Week 4	0.017	0.048	0.719	-0.083	0.118
Week 4	Week 8	.126	0.054	0.032	0.012	0.240

Based on estimated marginal means



\*. The mean difference is significant at the .025 level in adjustment for multiple comparisons.

<b>AA Subgroup: Somatic Anxiety Pairwise Comparisons within WLC</b>						
Time		Mean Difference	Std. Error	Sig.	95% Confidence Interval for Difference	
					Lower Bound	Upper Bound
Baseline	Week 4	0.001	0.063	0.990	-0.135	0.133
Week 4	Week 8	.126	0.052	0.028	0.015	0.238

Based on estimated marginal means

\*. The mean difference is significant at the .025 level in adjustment for multiple comparisons.

## Appendix Q Statistical Output for Adequate Adherence Subgroup Section 3.3.1.5

### AA Subgroup for Total Anxiety: Wilcoxon Signed Ranks

		N	Mean Rank	Sum of Ranks
HighAnx2 - HighAnx1	Negative Ranks	31 <sup>a</sup>	23.74	736.00
	Positive Ranks	10 <sup>b</sup>	12.50	125.00
	Ties	4 <sup>c</sup>		
	Total	45		

a. HighAnx2 < HighAnx1

b. HighAnx2 > HighAnx1

c. HighAnx2 = HighAnx1

### Test Statistics

	HighAnx2 - HighAnx1
Z	-3.963 <sup>b</sup>
Asymp. Sig. (2-tailed)	.000

a. Wilcoxon Signed Ranks Test

b. Based on positive ranks.

AA Subgroup Cognitive Tests of Within-Subjects Effects							
	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	2.904	1	2.904	0.207	0.651	0.005	0.073
Time *Adherenc e	6.267	1	6.267	0.447	0.507	0.011	0.100
Time * Cohort	24.224	1	24.224	1.729	0.196	0.041	0.250
Time * Age	24.642	1	24.642	3.219	0.080	0.074	0.417
Time * Sex	45.101	1	45.101	1.759	0.192	0.042	0.253
Error(Time)	560.490	40	14.012				

AA Subgroup Cognitive Estimated Means			
Measure:			
Time	Mean	Std. Error	95% Confidence Interval

			Lower Bound	Upper Bound
1	29.894 <sup>a</sup>	0.616	28.598	31.090
2	25.978 <sup>a</sup>	0.944	24.070	27.886

AA Subgroup Cognitive Estimated Means Pairwise Comparisons						
Time	Mean Difference	Std. Error	Sig.	95% Confidence Interval for Difference		
				Lower Bound	Upper Bound	
1   2	3.867*	0.789	0.000	2.272	5.462	

AA Subgroup Cognitive Estimated Means Multivariate Tests							
	Value	F	Hypothesis df	Error df	Sig.	Partial Eta Squared	Observed Power
Wilks' lambda	0.625	24.008 <sup>a</sup>	1.000	40.000	0.000	0.375	0.998

AA Subgroup In(Somatic) Tests of Within-Subjects Effects							
Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Observed Power
Time	0.003	1	0.003	0.081	0.777	0.002	0.059
Time * Adherence	0.001	1	0.001	0.035	0.852	0.001	0.054
Time * Cohort	0.125	1	0.125	3.392	0.073	0.078	0.436
Time * Age	0.000	1	0.000	0.005	0.944	0.000	0.051
Time * Sex	0.001	1	0.001	0.026	0.872	0.001	0.053
Error(Time )	1.476	40	0.037				

AA Subgroup In(Somatic) Estimated Marginal Means				
Time	Mean	Std. Error	95% Confidence Interval	
			Lower Bound	Upper Bound
1	3.155	0.043	3.069	3.242
2	3.005	0.040	2.923	3.087

AA Subgroup In(Somatic) Estimated Means Pairwise Comparisons						
Measure:						
(I) Time	Mean Difference (I- J)	Std. Error	Sig.	95% Confidence Interval for Difference		
				Lower Bound	Upper Bound	
1	2	.150*	0.041	0.001	0.068	0.232

AA Subgroup In(Somatic) Estimated Means Multivariate Tests							
	Value	F	Hypothesis df	Error df	Sig.	Partial Eta Squared	Observed Power
Wilks' lambda	0.744	13.728 <sup>a</sup>	1.000	40.000	0.001	0.256	0.951

## Appendix R Statistical Output for Female-Only Subgroup Comparison 1: 3 Groups over 4 Weeks

		Tests of Normality					
		Kolmogorov-Smirnov <sup>a</sup>			Shapiro-Wilk		
	GROUP	Statistic	df	Sig.	Statistic	df	Sig.
STICSA.tot	1	.186	18	.099	.887	18	.034
	Breathing	.195	40	.001	.917	40	.006
	Counting	.134	31	.166	.944	31	.105
STICSA.tot_2	1	.126	18	.200*	.969	18	.788
	Breathing	.116	40	.185	.969	40	.336
	Counting	.144	31	.101	.956	31	.228
InTOT	1	.190	18	.085	.908	18	.080
	Breathing	.171	40	.005	.947	40	.059
	Counting	.101	31	.200*	.963	31	.342
InTOT2	1	.124	18	.200*	.972	18	.834
	Breathing	.154	40	.018	.951	40	.085
	Counting	.118	31	.200*	.955	31	.218

\*. This is a lower bound of the true significance.

a. Lilliefors Significance Correction

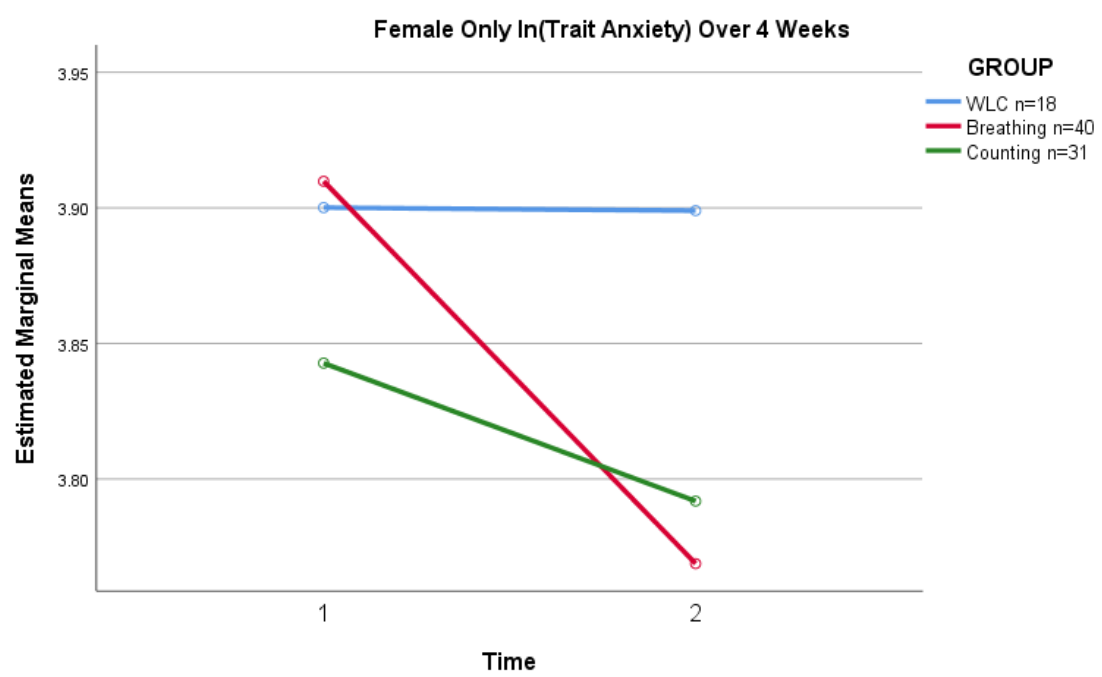
### FEMALE: In(Total Anxiety) Within-Subjects Effects

		Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Time	Sphericity Assumed	0.001	1	0.001	0.024	0.877	0.000
Time * Age	Sphericity Assumed	6.230E-05	1	6.230E-05	0.003	0.956	0.000
Time * GROUP	Sphericity Assumed	0.143	2	0.072	3.442	0.037	0.075
Error(Time)	Sphericity Assumed	1.767	85	0.021			

### Tests of Between-Subjects Effects

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Intercept	12.126	1	12.126	195.999	.000	.698
Age	.022	1	.022	.350	.556	.004
GROUP	.156	2	.078	1.259	.289	.029

Error	5.259	85	.062		
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Covariates appearing in the model are evaluated at the following values: Age = 18.74

COGNITIVE ANXIETY: Female Only

**FEMALE: In(Cognitive Anxiety) Within-Subjects Effects**

		Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Time	Sphericity Assumed	0.006	1	0.006	0.289	0.592	0.003
Time * Age	Sphericity Assumed	0.002	1	0.002	0.098	0.755	0.001
Time * GROUP	Sphericity Assumed	0.188	2	0.094	4.232	0.018	0.091
Error(Time)	Sphericity Assumed	1.892	85	0.022			

**FEMALE: In(Cognitive Anxiety) Tests of Between-Subjects Effects**

	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Intercept	8.005	1	8.005	111.618	.000	.568

Age	.053	1	.053	.746	.390	.009
GROUP	.152	2	.076	1.063	.350	.024
Error	6.096	85	.072			

SOMATIC ANXIETY: Female Only

**FEMALE: In(Somatic Anxiety) Within-Subjects Effects**

		Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Time	Sphericity Assumed	0.002	1	0.002	0.062	0.805	0.001
Time * Age	Sphericity Assumed	0.005	1	0.005	0.148	0.701	0.002
Time * GROUP	Sphericity Assumed	0.113	2	0.057	1.689	0.191	0.038
Error(Time)	Sphericity Assumed	2.844	85	0.033			

**FEMALE: In(Somatic Anxiety) Tests of Between-Subjects Effects**

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Intercept	7.945	1	7.945	76.770	.000	.475
Age	.003	1	.003	.024	.876	.000
GROUP	.173	2	.087	.836	.437	.019
Error	8.797	85	.103			

Appendix S. Statistical Output for Female-Only Subgroup: Physiological Measures over 4 weeks

**FEMALE: Systolic Blood Pressure Within-Subjects Effects**

		Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Time	Sphericity Assumed	9.857	1	9.857	0.572	0.454	0.013
Time * Age	Sphericity Assumed	11.689	1	11.689	0.678	0.415	0.015
Time * Group	Sphericity Assumed	147.197	2	73.599	4.268	0.02	0.162
Error(Time)	Sphericity Assumed	758.779	44	17.245			

**FEMALE: SBP Tests of Between-Subjects Effects**

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Intercept	4402.634	1	4402.634	33.900	.000	.435
Age	89.671	1	89.671	.690	.410	.015
Group	419.071	2	209.535	1.613	.211	.068
Error	5714.400	44	129.873			

**FEMALE: Diastolic Blood Pressure Within-Subjects Effects**

		Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Time	Sphericity Assumed	0.002	1	0.002	0.000	0.989	0.000
Time * Age	Sphericity Assumed	0.029	1	0.029	0.002	0.962	0.000
Time * Group	Sphericity Assumed	11.836	2	5.918	0.481	0.622	0.021
Error(Time)	Sphericity Assumed	541.801	44	12.314			

**FEMALE: DBP Tests of Between-Subjects Effects**

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Intercept	813.491	1	813.491	10.179	.003	.188
Age	41.897	1	41.897	.524	.473	.012



Group	54.439	2	27.220	.341	.713	.015
Error	3516.582	44	79.922			

**FEMALE: Tests of Between-Subjects Effects**

	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Intercept	6.385	1	6.385	44.841	.000	.505
Age	.223	1	.223	1.566	.217	.034
Group	1.131	2	.566	3.973	.026	.153
Error	6.265	44	.142			

**FEMALE: SDNN Within-Subjects Effects**

		Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Time	Sphericity Assumed	3.226E-05	1	3.226E-05	0.000	0.983	0.000
Time * Age	Sphericity Assumed	9.803E-05	1	9.803E-05	0.001	0.970	0.000
Time * Group	Sphericity Assumed	0.119	2	0.060	0.885	0.420	0.039
Error(Time)	Sphericity Assumed	2.962	44	0.067			

## Appendix T Statistical Output for Female-Only Subgroup Comparison 3: 2 Groups over 8 Weeks

### FEMALE In(Total Anxiety) Mauchly's Test of Sphericity<sup>a</sup>

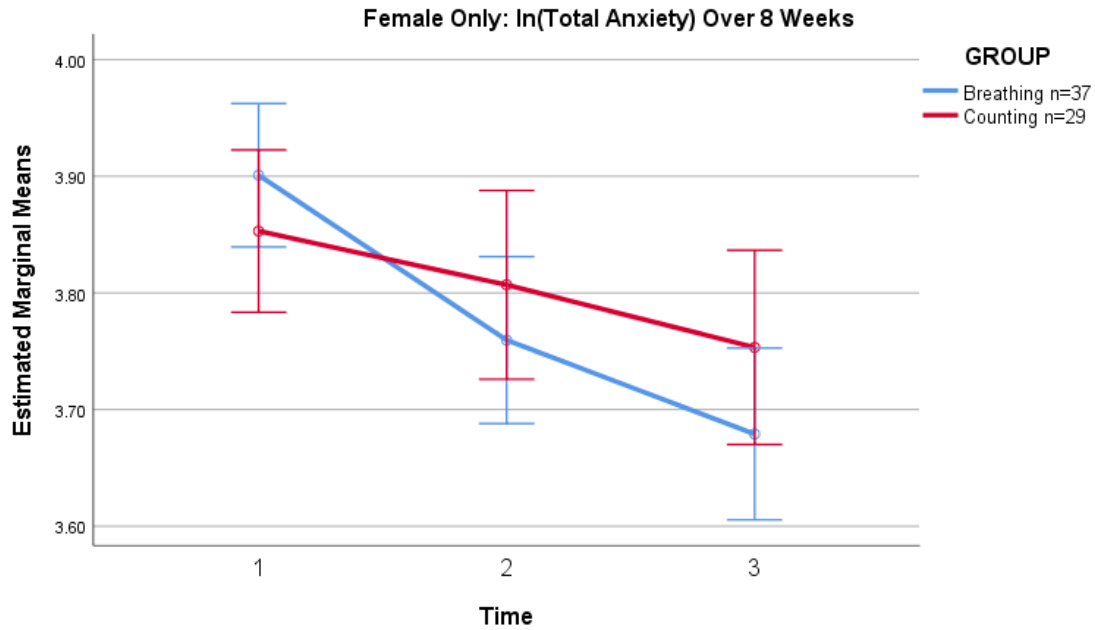
Within Subjects Effect	Mauchly's W	Approx.	df	Sig.	Epsilon <sup>b</sup>		
		Chi-Square			Greenhouse-Geisser	Huynh-Feldt	Lower-bound
Time	.924	4.758	2	.093	.929	1.000	.500

### Female Only: In(Total Anxiety) Within-Subjects Effects

Source		Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Time	Sphericity Assumed	0.004	2	0.002	0.118	0.889	0.002
Time * Cohort	Sphericity Assumed	0.048	2	0.024	1.281	0.281	0.021
Time * Age	Sphericity Assumed	0.014	2	0.007	0.377	0.687	0.006
Time * App Freq	Sphericity Assumed	0.016	2	0.008	0.423	0.656	0.007
Time * GROUP	Sphericity Assumed	0.131	2	0.066	3.486	0.034	0.054
Error(Time)	Sphericity Assumed	2.297	122	0.019			

### FEMALE In(Total Anxiety) Tests of Between-Subjects Effects

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Intercept	14.004	1	14.004	149.605	.000	.710
ONLINE	.005	1	.005	.049	.826	.001
Age	.000	1	.000	.001	.972	.000
appfreq3W	.002	1	.002	.022	.881	.000
GROUP	.029	1	.029	.307	.581	.005
Error	5.710	61	.094			



Covariates appearing in the model are evaluated at the following values: Cohort = 1.6, Age = 18.8, Self Reported App Freq = 2.8  
Error bars: 95% CI

**Female Only: In(Cognitive Anxiety) Mauchly's Test of Sphericity**

Within Subjects Effect	Mauchly's W	Approx.			Epsilon		
		Chi-Square	df	Sig.	Greenhouse-Geisser	Huynh-Feldt	Lower-bound
Time	.949	3.121	2	.210	.952	1.000	.500

**Female Only: In(Cognitive Anxiety) Within-Subjects Effects**

Source		Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Time	Sphericity Assumed	0.004	2	0.002	0.093	0.912	0.002
Time * Cohort	Sphericity Assumed	0.067	2	0.033	1.442	0.240	0.023
Time * Age	Sphericity Assumed	0.015	2	0.007	0.315	0.731	0.005
Time * App Frequency	Sphericity Assumed	0.053	2	0.026	1.148	0.321	0.018
Time * GROUP	Sphericity Assumed	0.236	2	0.118	5.124	0.007	0.077
Error(Time)	Sphericity Assumed	2.813	122	0.023			

**Female Only: In(Cognitive Anxiety) Tests of Between-Subjects Effects**

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Intercept	8.836	1	8.836	82.070	.000	.574
ONLINE	.001	1	.001	.012	.915	.000
Age	.029	1	.029	.266	.608	.004
appfreq3W	.000	1	.000	.001	.975	.000
GROUP	.087	1	.087	.809	.372	.013
Error	6.567	61	.108			

**Mauchly's Test of Sphericity**

Within Subjects Effect	Mauchly's W	Approx. Chi-Square	df	Sig.	Epsilon		
					Greenhouse-Geisser	Huynh-Feldt	Lower-bound
Time	.916	5.248	2	.073	.923	1.000	.500

**Female Only: In(Somatic Anxiety) Within-Subjects Effects**

Source		Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Time	Sphericity Assumed	0.027	2	0.013	0.485	0.617	0.008
Time * Cohort	Sphericity Assumed	0.037	2	0.018	0.662	0.518	0.011
Time * Age	Sphericity Assumed	0.042	2	0.021	0.752	0.474	0.012
Time * App Freq	Sphericity Assumed	0.000	2	0.000	0.005	0.995	0.000
Time * GROUP	Sphericity Assumed	0.045	2	0.023	0.820	0.443	0.013
Error(Time)	Sphericity Assumed	3.382	122	0.028			

### Tests of Between-Subjects Effects

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Intercept	9.935	1	9.935	60.746	.000	.499
ONLINE	.021	1	.021	.130	.720	.002
Age	.034	1	.034	.211	.648	.003
appfreq3W	.011	1	.011	.065	.800	.001
GROUP	.005	1	.005	.032	.859	.001
Error	9.977	61	.164			